International Union of Soil Sciences®

SUSTAINABLE SOIL MANAGEMENT AS A KEY TO PRESERVE SOIL BIODIVERSITY AND STOP ITS DEGRADATION

EDITORS:

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International
Decade of Soils
2015-2024

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2022

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Please cite this publication as follows:

Reyes-Sánchez, L. B., Horn, R., Costantini, E. A.C. (eds.) 2022: **Sustainable soil management as a key to preserving soil biodiversity and stopping its degradation**. International Union of Soil Sciences (IUSS). Vienna, Austria.

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Sponsor: International Union of Soil Sciences (IUSS)

ISBN: 979-8-9862451-0-2

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Foreword

Most organisms in terrestrial ecosystems develop at least a part of their life cycle in soil habitats or live in it, making the soil resource the most important reservoir of global biodiversity. Soil biodiversity plays a fundamental role in the proper functioning of all terrestrial ecosystems as well as economic, social, and human services that guarantee both human well-being and the existence of life on Earth.

Furthermore, soil healthy, nutritious food, climate change mitigation, as well as many other toral issues essential to our life, all depend on the knowledge of soil biodiversity.

Against this background intends the IUSS, as the leader in Soil Sciences at the worldwide level, to foreground the study of edaphic biodiversity from the broad interdisciplinary perspective, in order to promote the advancement of its area of knowledge.

In this regard, Soil Biodiversity was proposed as a current topic of great scientific, social, economic, and political importance for the present IUSS book as a document that had to be written from an interdisciplinary perspective and starting on the current state of the art in this area. A book written from the perspective of various points of view and interactions since the different areas of knowledge of soil sciences offered by the authors.

That is why the book contains both experimental data and conceptual information, didactic experiences, and reviews organized in four different sections showing points of view and interactions from the following knowledge areas of soil sciences:

* Soil Biodiversity perspectives from biological sciences point of view

* Interdisciplinary perspectives from agronomical point of view

* Interdisciplinary perspectives from soil physical point of view

* Interdisciplinary perspectives from soil chemistry and education point of view

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This book is the first of the Open Access Book Series of the IUSS.

Dr. Laura Bertha Reyes Sánchez — UNAM, Mexico
President of the International Union of Soil Sciences — May 19, 2022

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Chapter 1

The link between the soil and the human microbiome-Holobiont theory as key concept for the human future

Katharina M. Keiblinger, Martin Grassberger and Winfried E. H. Blum

Summary

Microbes are essential for life on earth. The recently proposed "*one health*" concept emphasizes the importance of the microbiome and its diversity on ecosystem and human health.

In this chapter, losses in microbial diversity including both soils and human (intestine) and potential consequences on the functioning of the respective systems and interlinkages are depicted. In particular, the use of plant protection agents and antimicrobials in human medicine as well as resistance in soils is discussed in the prospect of the declining biodiversity. The development of the human microbiome and how environmental microbes can proliferate over large distances and impact human health and diseases are outlined. Sustainable agricultural management is of crucial importance for terrestrial ecosystem functioning in the face of changing climate. We discuss potential steps for the latter, in combination with a human lifestyle that is re-connecting to a microbial rich and diverse nature and show how essential this is to maintain and retrieve human health.

Introduction

Since human presence, aboveground species diversity has dropped by 1000-fold (Joppa et al. 2016). Total biomass of humans and domesticated animals (mainly livestock) greatly surpasses wild mammals (Bar-On et al. 2018). The world population together with its animal husbandry has reached about 97% of the total weight of all vertebrates on earth. The remaining 3%, which are not domesticated, are constantly declining further.

This is a clear signal, indicating that the global natural resources are becoming more and more under threat. This human induced impact has not only caused an enormous loss of soils, for example through sealing and erosion, but also the deterioration of soil quality, especially for the production of food and fiber and the maintenance of adequate environmental living conditions for humans at a worldwide level (Blum, 2017). In this context, especially the use of antibiotics for maintaining and increasing the stock of animal husbandry has deteriorated the quality of agricultural soils through a reduction of their biodiversity and that of their vegetation cover.

The "*one health*" approach (*Table 1*) is of high relevance in times of a pandemic crisis, as it aims to include not only the issues of food safety but also to control diseases such as zoonoses and combat antibiotic resistance. To ensure planetary health it is crucial to account the linkages between our environment the ecosystems, biodiversity and human health (Zhu et al. 2019).

The need for food and fiber is still increasing globally and is suggested to further rise up to 70% by 2050 (Singh and Trivedi, 2017). To tackle todays and future challenges regarding food production under increasingly harsh climatic conditions is of high priority. Conventional farming practices had enabled higher agricultural productivity in the previous decades and, hence, contribute to better access to food and alleviate poverty. However, this was to a large extent achieved by massive use of agrochemicals such as fertilizers and pesticides. The excessive use may counteract food security and human health (Singh and Trivedi, 2017). Soils under arable use need to be maintained by sustainable production systems, that they can further contribute to climate change mitigation, and we should emphasize on a management that is conserving and improving its biodiversity (FAO et al. 2020).

The European commission has launched the Mission of Soil Health which aims for improving soil health under sustainable management to ensure essential ecosystem function/services such as soil carbon sequestration and the production of nutritious and safe food (*https://ec.europa.eu/info/publications/caring-soil-caring-life_en accessed 15.6.2021*).

Soil biodiversity and ecosystem functioning

In the biosphere microbes are omnipresent and thereby provide a pillar for life on earth. The soil inhabits an enormous diversity of fauna and microbes, which are of crucial importance for human life to provide food, feed, fiber and medicine (Raaijmakers and Mazzola, 2016). To get a concept and put this into numbers, a single gram of soil is inhabited by a few thousand microbial species and hence, soils belong to the most diverse environments globally (Allison and Martiny, 2008).

In the Anthropocene (*see definition in Table 1*), global biodiversity has declined, by a strong and irrevocable reduction in plant biodiversity and also soil biodiversity (Geisen et al. 2019). Consequently also plant host associated organisms are lost, but the degree of species extinction is unknown (Geisen et al. 2019).

Even though the importance of the soil microbial diversity for ecosystem functions is widely acknowledged, the role of individual species and the potential impact on plant and human health is far from understood (Raaijmakers and Mazzola, 2016; Wagg et al. 2018). However, the threats to global (soil) biodiversity are manifold, with one main driver the human induced ecosystem changes (Geisen et al. 2019). Still causal links between loss in biodiversity and ecosystem functioning are under debate (Maron et al. 2018), with limited "functional redundancy" among soil microbes, until a critical limit for biodiversity loss/species extinction is reached (Delgado-Baquerizo et al. 2016).

The latter suggests a decoupling of soil biodiversity and ecosystem functioning, with a loss of some taxa, "generalized functions" remain via functional redundancy while rare functions can severely reduce ecosystem stability (Tibbett et al. 2020; Bardgett and van der Putten, 2014). This is in compliance with the concept of "*keystone taxa*" (*Table 1*), that are proposed to be highly relevant for stable and proper ecosystem functioning (Banerjee et al. 2018).

Plants and soil microbes evolved together, and this resulted in mutualistic associations, with benefits for both partners. With plant breeding programs mainly focusing on crop yields, these relationships have suffered by the loss of key beneficial species.

Soil microbiome and its interaction with plants and plant protection agents.

The soil microbiome is suggested to be interrelated with its environment, this includes not only the abiotic climatic and mineral soil components, but also their living elements such as the plants and soil biota (Wallenstein, 2017). The microbiome of plants is called the phytomicrobiome and covers the bacteria and fungi that occur on various plant compartments such as root, shoot leaf, seed and flower. These microbes play a crucial role in plant productivity and health (Chouhan et al. 2021). The functional aspects of the phytomicrobiome (*Table 1*) can be promoted by environmentally friendly agricultural management approaches and thereby promote the resource use efficiency of crops, crop yield and fitness including their resilience to (a)biotic stresses (Chouhan et al. 2021).

Fostering the crop microbiome can provide a huge potential to agriculture and food production by (i) improved productivity and quality by increased crop health through integrated management practices adapted to the climatic conditions, (ii) control of pathogens and pests in a more sustainable way, trough reduced application of chemical products and hence more friendly approaches for environmental and human health, (iii) more efficient resource (i.e. water, and soil nutrient) use, for more resilient and healthy crops under a changing climate.

Changing climate also promotes the proliferation of invasive species as well as pathogens and pests (Finch et al. 2021). To safeguard natural habitats (i.e. pristine environments) and agricultural products, the “phytomicrobiome” and microbial inoculants, may promote positive environmental and social outcomes (Singh and Trivedi, 2017).

Agrochemicals such as pesticides are widely used in agriculture, with about half of them were herbicides applied in 2010 (Sharma et al. 2019). Herbicides show detrimental effects on non-target organisms, such gut microbiota of honey bees (Motta et al. 2018), however, how this affects the soil microbial community structure and functions is still unclear. A recent study on three widely used herbicides including glyphosate, dicamba and glufosinate demonstrated an increase in antibiotic resistance genes and mobile genetic elements, without clear changes in bacterial community abundance and diversity (Liao et al. 2021). Herbicides exposure was reported to not only select for mutations that are advantageous in their metabolism for xenobiotic degradation, but also to increase herbicide resistance in bacteria (Rainio et al. 2021). In this regard, the scientific community is unaware about potential evolutionary impacts of herbicide application for the ecology of soil microbial communities and the functional consequences (Liao et al. 2021). This should be considered for the increase in antibiotic resistance genes by herbicides. However, also for antibiotic resistance genes and potential pathogens introduced with fertilization of untreated manure as these can be probably re-localize their resistance from beneficial or harmless soil microbes to pathogens for humans.

The human microbiome and its evolution

In the course of the evolution of multicellular organisms, an increasingly complex digestive tract developed, which usually consists of a more or less long tube with a “front” and “rear” opening. While ingesting food, all such multicellular organisms naturally also take in microbes from their environment, some of which feel so comfortable in the hosts intestine that they settle permanently.

And yet the bacterial community in the intestine differs considerably from species to species and from individual to individual. Certain types of bacteria are highly specialized in their animal (or plant) host as a result of a long shared evolutionary coexistence. These organisms therefore often have a very close relationship with their host during its entire lifetime. Other microorganisms, on the other hand, only colonize the host temporarily, but interact with the local microorganisms during their passage through the host intestine.

Our body consists of around 30 trillion somatic cells and 38 trillion bacteria (Sender et al. 2016). Most of these bacteria are friendly or indifferent towards us and usually feel quite comfortable in the warm, oxygen-poor environment of our intestines with a daily supply of nutrients. Together, it is estimated that these intestinal bacteria weigh up to 0.2 kg (Sender et al. 2016). The importance of our microbiome (*Table 1*) can be recognized primarily from the fact that our body cells only have around 23,000 genes each, but we carry somewhere between 2 million and possibly even 20 million bacterial genes in our microbiome. In a cross-study meta-analysis (covering 3,655 samples from 13 studies) of metagenomes (*Table 1*) from two human body niches, the mouth and gut, found staggering genetic heterogeneity in the dataset, identifying a total of 45.666.334 non-redundant genes (approx. half of them oral and half of them in the gut) at the 95% identity level (Tierney et al. 2019). That means we are probably 99% microbe, at least from a genetic point of view.

This additional, variable genome, the microbiome, ultimately leads to considerable inter-individual differences in the genetic makeup of different people. Our microbiome therefore increases our genetic diversity considerably. The importance of this overall bacterial genome is of such great importance for our organism that we are now talking about our "second genome" (Grice and Segre, 2012). The fact that the entirety of the microbiota in our body is now referred to as "a separate organ" shows the importance which our microbial companions have gained in biomedical research (Baquero and Nombela, 2012).

Glossary

Concept	Definition	Citation
One health	*The one health concept extends the previously proposed "one medicine" and ecosystem health concept, to the whole ecosystem".*	(Zinsstag et al. 2011)
Anthropocene	*„A new human-dominated geological epoch"*	(Lewis and Maslin 2015)
Functional redundancy hypothesis	*"The ability of one microbial taxon to carry out a process at the same rate as another under the same environmental conditions"*	(Allison and Martiny 2008)
Keystone taxa	*Keystone taxa, are essential for proper ecosystem functioning*	(Trivedi et al. 2017)
Microbiome	This term was initially used to refer to the Genes within microbes, but the term is currently being used as descriptive term for the microorganisms themselves (i.e. the microbiota)	

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Concept	Definition	Citation
Metagenome	Refers to the entire genetic material in a sample and is composed of the entire genetic material of all the involved organisms.	
Phytomicrobiome	The microbiome of plants, including bacteria and fungi that occur on various plant compartments	(Chouhan et al. 2021)
α-diversity	Also termed richness, is a measure of the biodiversity of a habitat, it describes the number of species that occur in a habitat.	
β-diversity	This is a measure of the difference in species biodiversity between different communities spatially distributed in a landscape	(Brevik et al. 2020; Romanelli et al. 2015)
coprolith	Also coprolite, is fossilized stools, and derives from paleofeces	Wibowo et al. 2021
Biodiversity hypothesis or "old friend hypothesis"	Replacing the "Hygiene hypothesis" The importance of human immune system, by vital exposure to microbes that were already present during human (and mammalian) evolution, which could persist in small hunter-gatherer groups as microbiota.	(Rook et al. 2017; von Hertzen et al. 2011)
Hygiene hypothesis	Suggests that limited exposure to microorganisms leads to disordered regulation of the immune system with can cause increased inflammatory disorders. Vice versa contacts with communalist and pathogenic microbes can stimulate immune functions	(Rook 2009)
(human) holobiont	Is an entire living to be seen as a whole "ecosystem" that not only includes about 20,000 genes of the human, but also of up to a million genes from their bacteria, which are located in the intestines, among others.	(Rosenberg and Zilber-Rosenberg 2013)
Flying microbes	These originate from a terrestrial source and can spread across the troposphere around the globe	(Lopatina et al. 2019)
Farm effect	The protective effect of farms against allergic diseases and the high numbers of microbes there is now known in science as the "farm effect"	(Depner, et al. 2020)

Table 1. Glossary.

Ecosystem services of our microbiome

Short chain fatty acids (SCFAs) are among the best-known and apparently most important metabolic products of our microbiome with regard to our health. These bacterial metabolic products, which are also produced in inconceivable quantities in the bovine rumen and thereby supply the grass-eating host with energy, are also produced in our large intestine are created as a by-product when intestinal bacteria break down complex carbohydrates. Further the SCFAs not only protect the lining of our intestines, but also have an anti-inflammatory effect on the whole body and in addition influence sugar and fat metabolism (Morrison and Preston, 2016).

Likewise, SFCAs have a positive effect on our immune system, promote the formation of new blood vessels and support our stem cells. The three main forms of SFCAs acetate, propionate and butyrate play the main role. Propionate and butyrate reduce the cholesterol level and inflammatory processes, and prevent the formation of arteriosclerotic changes in the vascular wall associated with high blood pressure (Bartolomaeus et al. 2019, Kim et al. 2018) and activate the cells of the immune system (Nastasi et al. 2015).

Numerous phytonutrients get into the large intestine through their connection with the complex structures of the plant cell walls. There they are released by the microbial activity or are even converted by bacteria. This means that the intestinal bacteria are also able to produce chemical compounds that were not originally present in the plant (Braune and Blaut, 2016).

In connection with the plant building material lignin, the so-called ferulic acid, for example, also reaches the large intestine, where it is released by intestinal bacteria. Other bacteria within the intestinal microbial community then convert this ferulic acid into other phenolic acids, which in turn are absorbed by the body, where they act as antioxidants (Duncan et al. 2016).

On the other hand, some food components can also be converted into harmful compounds by intestinal bacteria. For example, the conversion of the inherently harmless phosphatidylcholine from animal products such as meat or eggs can be converted into the atherosclerosis-promoting metabolite trimethylamine and thus endanger cardiovascular health in the long term (Wang et al. 2011). A variety of other food ingredients counteract these negative consequences.

Studies have shown that gut bacteria mediate the interaction between protein metabolism and the host's immune response. Amino acids can be metabolized into numerous microbial breakdown products, and these metabolites are involved in various physiological functions that may be related to both health and diseases of the host. Higher amino acid levels and undigested protein lead to an increase in pathogenic microorganisms with an associated higher risk of metabolic diseases. The proteins ingested with food can also lead to potentially harmful metabolites such as indol, phenol and cresol in the course of metabolism by the intestinal bacteria. These metabolites may also contribute to an increased risk of colon cancer (Louis et al. 2014).

So-called xenobiotics are chemical compounds that are actually foreign to the biological cycle of an organism. The list ranges from drugs to pesticides. With its immense diversity, the human microbiome has a high metabolic capacity that considerably exceeds the metabolic potential of its host. As a result, the human intestinal microbiome is also able to metabolize xenobiotics in food through to pharmaceutical ingredients (Abdelsalam et al. 2020). In the course of this biotransformation, the microbiome can alter the half-lives of xenobiotics, their potential biological effects on the human body, and the speed and extent with which they reach the bloodstream or their biological targets (Koppel et al. 2017). The microbial enzymes can activate drug precursors (so-called prodrugs), inactivate drugs or increase or decrease their toxicity. The microbiome thus contributes to the fact that people respond differently to drugs due to the different microbiome compositions and the different enzymatic activity of different types of bacteria.

Some metabolic products of bacteria are used for "communication" with their host. The SCFAs produced by bacteria regulate fat storage, hunger and satiety hormones as well as the consumption preferences of their host, which in turn influences the composition of the microbiome and thus can have serious long-term consequences for health and behavior (Tremaroli and Bäckhed, 2012).

Our daily diet influences the composition of the gut microbiota and its SCFA production to a considerable extent. The resulting interaction between nutrition, microbiota and their metabolites is in turn associated with the development of serious metabolic disorders such as obesity and diabetes. Initially, however, the question often arose whether the changes in the gut microbiota are the cause or the consequence of these diseases.

Bacterial metabolic products such as SCFAs and numerous other compounds develop their effect not only in the intestine, but also reach all imaginable target organs in our body via the bloodstream. Up to 30% of the molecules in our blood are of bacterial origin (Nicholson et al. 2012). We are permanently flooded with bacterial information molecules that influence the physiology and metabolism of distant organs and possibly the bacterial communities in other parts of the body (Swann et al. 2011, Wikoff et al. 2009).

It is precisely these bacterial metabolites that have shown science that certain diseases and the microbiome not only have a certain statistical association, but that there is actually a causal relationship between bacteria and their metabolic products and specific clinical presentations (Koh and Bäckhed, 2020).

Studies have demonstrated an intimate connection between the intestinal microbiota and the development and function of our brain and consequently also our behavior and psyche (Mayer, 2011, Heijtz et al. 2011).

Our almost intimate interplay with our intestinal microbiome and its apparently central importance for health and disease has – in conjunction with an increase in non-communicable disease – led to numerous considerations regarding a possible decrease in the biodiversity of the microbiota and its causes. The fact that intestinal bacteria remained connected to their host over long evolutionary periods suggests a positive selection of this symbiotic relationship in the course of evolution, but also shows that disorders in this relationship after such a long time spent together can have potentially negative health consequences.

Effects of the human lifestyle on the microbiome – hunter & gatherer societies vs. modern urban societies.

About 12,000 years ago at the beginning of the Holocene, numerous human populations around the world gradually changed their lifestyle from a hunter-gatherer existence to a sedentary one at almost the same time. We refer to this period as the beginning of agriculture. This serious change in the human way of life, also known as the "neolithic revolution", subsequently triggered a longer period of even more rapid cultural developments, which accelerated even further, especially within the last 300 years in the course of industrial and post-industrial societies.

The latest scientific data suggest that the transition from the hunter-gatherer existence to the agricultural way of life initially led to a change in the frequency of different types of intestinal bacteria, but not necessarily to a change in bacterial diversity (so-called alpha diversity, *Table*

1). Comparisons of the intestinal microbiome of two populations living practically next to each other in Central Africa, one living as a hunter-gatherer (BaAka Pygman) and the other farming (Bantu), showed that hunters and gatherers have more types of intestinal bacteria that metabolize amino acids and vitamins, whereas in the rural Bantu, more types of intestinal bacteria are concerned with the metabolism of carbohydrates and xenobiotics. As far as general intestinal bacterial diversity is concerned, however, no significant differences could be found, there was only a difference in the species composition and functioning of the intestinal microbiota (Gomez et al. 2016). Similar results were found when comparing the gut microbiome of traditionally agricultural populations from Peru and Cameroon. Here, too, there were clear differences in the species composition, but no differences in terms of diversity (Obregon-Tito et al. 2015, Morton et al. 2015). These results show that there are differences in the species composition of the microbiome (but not necessarily in diversity) in the case of two neighboring population groups, who both pursue different but nonetheless traditional and "natural" ways of living and eating with close contact to the natural environment. These studies support the adaptability of the microbiome in response to changing ways of living and eating.

However, a comparison of the intestinal microbiome of Nigerian farmers (Bassa) who live in isolation, with daily intensive contact with their environment, with that of people from several Nigerian cities and other regions of the world, shows that specific microbiome properties are ultimately increasingly lost via an urbanization gradient (Ayeni et al. 2018). In agreement with this, we see a significant reduction in bacterial diversity within the gut microbiome in populations that lead an industrial or post-industrial lifestyle. For example, Americans have significantly fewer types of bacteria in their intestines than individuals with a traditional lifestyle, such as in Malawi, Peru, Venezuela, or Papua New Guinea (Yatsunenko et al. 2012, Clemente et al. 2015, Martínez et al. 2015). Likewise, the intestinal microbiome of Italians is less species-rich than the microbiome of the Hadza hunters and gatherers in Tanzania (Schnorr et al. 2014, Rampelli et al. 2015). People from rural Africa also have a higher variety of bacteria in their intestines than African Americans in the United States (Ou et al. 2013). Similar differences, especially with regard to functional metabolic capacity, can be found when comparing people from rural areas of Russia with their urban fellow citizens (Tyakht et al. 2013).

Stool samples from traditional populations, on the other hand, showed a significantly higher overall biodiversity than the microbiome of people with a modern, western lifestyle and contained families of bacteria that are rarely or not at all present in the intestines of people from industrialized nations (Smits et al. 2017). Analysis of stool samples from a Yanomami tribe that had never been in contact with the outside world showed that their microbiome is home to the highest diversity of bacteria and genetic functions ever recorded in a human population (Clemente et al. 2015).

Examinations of fossilized stools, so-called coproliths (*Table 1*), from archaeological sites that are 1,400 to 8,000 years old, reveal a similar picture. The microbiome composition from the archaeological finds is more similar to that of populations with a traditional lifestyle (and therefore close to nature and soil) than to that of people from industrialized societies (Tito et al. 2012). A recent study on up to 2,000 year old but well-preserved "paleostools" from the southwest of the USA and Mexico uncovered bacterial DNA sequences, 39% of which could be assigned to previously unknown types of bacteria (Wibowo et al. 2021).

Clearly, there seems to be a diversity decline of gut microbiota along an urbanization and civilization gradient.

What are the possible causes and drivers of this microbial species loss?

What could be the cause of the alarming loss of diversity in our gut microbiota? We should be concerned with this question simply because we now have numerous indications that a decrease in biodiversity in general, but in our intestines in particular, is associated with considerable instability and susceptibility to disturbances and, hence, to an array of modern diseases (e.g., Agus et al. 2021, Al Bander et al. 2020, Albhaisi et al. 2020, Verhaar et al. 2020, Gomaa, 2020).

Since the only access to our intestines is through the oral ingestion of matter from our environment (if we disregard the potential access through the other side), the solution to the all-important question will probably lie here. So, what do we absorb or not absorb that leads to the apparently ever faster decreasing biodiversity of our intestinal microbes?

Some scientists are convinced, that the widespread use of antibiotics over the last few decades has made a large contribution to the disturbances and the extinction of species observed in our intestines (Shane, 2014).

Another cause of the observed microbiome depletion in industrialized societies is very likely the significant increase in the consumption of highly processed foods with a simultaneous decrease in the fiber content in our food. Long-term experiments on mice have shown with worrying clarity that reducing dietary fiber can lead to the complete elimination of various types of bacteria. As a result, these types of bacteria could no longer be transmitted from the mother to the offspring. They were simply lost over generations (Sonnenburg et al. 2016).

However, the comparisons between traditional populations and modern societies made so far show one thing very clear: the less contact there is with nature in all its forms (soil, animals, vegetation, etc.), the more impoverished the bacterial biodiversity in people's intestines.

In this context the terms "hygiene hypothesis" and "old friends hypothesis" (*Table 1*) have been established, referring to a lack of exposure to beneficial environmental bacteria (the "old friends" in part as a consequence of too much hygiene in modern living environments (Rook et al. 2014, Rook, 2012).

The richest source of microbes: our environment

There is no doubt that humid environments such as soils and aqueous environments (groundwater, lakes, rivers and wastewater) are the natural habitats in which the largest and most diverse population of bacteria can be found globally (Tamames et al. 2010). These large microbial habitats are therefore an important source for the spread of bacteria and their genes into our intestinal microbiome (Vaz-Moreira et al. 2014). So far only a few groups of bacteria from these habitats have been associated with the human microbiome. However, there is reason to suggest that some bacteria from these habitats have a connection to humans and are involved in the direct or indirect transfer of metabolic properties, including resistance to antibiotics.

A recent comparison of microbiome data sets from global soil samples and different humans showed that taxonomic groups of bacteria found in the gut microbiome also match a number of bacterial taxa of the soil. Such findings suggest that soil bacteria may have played and continue to play an important role in the development of the human microbiome and immune system throughout evolution (Schnorr, 2020).

Linking soil biodiversity and human health

A search of the scientific literature, however, reveals relatively few studies that are dedicated to the soil as a biological link between the microbial environment and human health. One possible reason for this is that the microbial diversity of soils and our exposure to them are difficult to quantify scientifically. Especially not over long periods of time. In addition, the soil microbiome has so far not been one of the most pressing research questions in biomedical science, but rather has been dealt with by representatives of agricultural science and soil science. So far, because that has increasingly changed in recent years due to the blurring of specialist boundaries in the course of microbiome research. The question of the origin of our intestinal microbes is literally obvious when considering a holobiont (*Table 1*) like humans as a whole.

Route of exposure/mechanism of action of soil microbiota

Air surveys at different altitudes showed that viable bacteria make up on average about 20% of the total particles in the range from 0.25 to 1 μm in diameter (DeLeon-Rodriguez et al. 2013). Such studies clearly show that bacteria make up a significant and previously underestimated proportion of atmospheric aerosols. Although relatively little is known about the species composition and the spatial and temporal distribution of these “flying microbes” (*Table 1*), it can be assumed that they originate from a terrestrial source (ocean, soil, freshwater, etc.) and, in disregard of any human limits, can spread across the troposphere around the globe.

Both the “old friends’ hypothesis” and the “hygiene hypothesis” are based on the fact that microorganisms to which we humans have been exposed throughout evolution have assumed many important functions in our body over time. Our daily direct contact with a wide variety of environmental germs takes place at the interfaces between the organism and the environment (the so-called epithelial barriers) through ingestion and inhalation and through contact with the skin and mucous membranes. If there is an increasing lack of contact with these microorganisms, health problems appear to arise.

Is there any scientific evidence that microbes in the environment have beneficial effects on health?

As far as the possible effect of soil bacteria on our health is concerned, several interesting studies (as always mostly on mice) have shed light on this largely unexplored matter.

For example, a study on the effects of hygiene and living environment on the composition of the gut microbiota of mice and the development of the immune system showed, that exposure to soil, house dust and rotting plant material improved gut microbial diversity and innate immunity (Zhou et al. 2016).

A comparison of the intestinal microbiome and some immune parameters of mice that were either kept on a sterile bedding or with natural ground contact revealed clear differences in the composition of the intestinal microbiota between the two groups. The mice with ground contact also showed an altered gene expression, whereby exposure to soil microbes led to an alleviation of the inflammatory reaction that occurs in the context of allergic diseases (Ottman et al. 2019).

It has been criticized in part that the mice in these experiments were exposed to an unrealistically high bacterial load and excessive contact with the ground, so that the results could not be compared with the natural exposure of humans. Another research team therefore examined the question of whether different, "realistic" microbial concentrations in the air, with a gradient from low to high natural bacterial biodiversity, have different effects on the intestinal microbiome and fear behavior in mice. In a randomized controlled study, they exposed mice to air that had minimal traces of dust from soils with high and low bacterial biodiversity for seven weeks. The results of this study provided the first evidence that realistic exposure to soil dust containing bacteria in the lowest concentration can actually influence the intestinal microbiome with leads to a reduced anxiety-like behavior, at least in mice (Liddicoat et al. 2020). A suspect from the ranks of soil microbes that could be responsible for the anxiety-relieving effect in mice could also be identified: *Kineothrix alysoides* from the family Lachnospiraceae. The authors of the study assumed, that this "anxiety-relieving germ" could both be part of the normal intestinal flora and also be absorbed or supplemented through contact with a biodiverse, bacteria-containing environment. *K. alysoides* is one of the butyrate-producing bacteria and probably occurs primarily in soils with high biodiversity and abundant aboveground vegetation.

In rats it was also shown, that the intestinal symbiont *K. alysoides*, along with other bacteria, improved the intestinal barrier function and reduced the amount of endotoxins from intestinal bacteria such as lipopolysaccharides in the blood (Qiao et al. 2020). Both a disturbed intestinal barrier and bacterial endotoxins are factors that are associated with a disturbed intestinal microbiome and chronic low-grade inflammation, factors that are believed to play an important role in the development of numerous chronic diseases (e.g. Tilg et al. 2020).

So indeed, first convincing experimental evidence exists, that bacteria from a biodiverse environment can be transmitted via the air and thus even become part of the intestinal microbiota. However, harmless bacteria from the environment do not necessarily have to establish in the intestine in order to have a positive effect. In many cases it is probably sufficient if they come into contact with our intestinal immune system, as living or dead bacteria, in order to develop a positive immunomodulatory effect.

Although there are undoubtedly differences in the immune system of humans and mice or rats, experiments have shown, that there are potential connections between environmental bacteria and important physiological functions of vertebrates. The administration of living bacteria of the species *Mycobacterium vaccae* (a non-disease-causing soil bacterium) to mice, reduced, similar to *K. alysoides*, fear-like behaviors and improved their ability to learn (Matthews and Jenks, 2013). Even heat-killed bacteria of this type retained their immunomodulatory effects and induced an anti-inflammatory effect, which could prevent allergic asthma, intestinal inflammation and fear-like behavior in mice (Reber et al. 2016, Loupy et al. 2019). The repeated injection of killed bacteria of the species *M. vaccae* led to better fear extinction after traumatic events in male rats and, according to the authors of a study, could lead to novel interventions in post-traumatic stress disorder (Fox et al. 2017). Interestingly, 20 years ago scientists observed a substantial improvement in children with atopic dermatitis (neurodermitis) after they were subcutaneously injected with killed bacteria of the species *Mycobacterium vaccae* in what is probably not a very pleasant therapeutic procedure (Arkwright and David, 2001).

The farm as a source of knowledge for science

After the reunification of Germany, there was an opportunity to check whether factors that can be traced back to increased pollution in the environment play a significant role in the development of asthma and allergies. In various studies, children and adults from large western and eastern German cities were examined for the frequency of allergic diseases. Scientists initially assumed, that an increased allergy rate was to be expected in Eastern Germany due to greater environmental pollution. Surprisingly, the opposite was the case: both the number and diversity of allergic diseases were significantly lower in former Eastern than in Western Germany. Several studies confirmed that factors such as growing up under poor hygienic conditions, having several siblings, drinking unpasteurized milk and more intensive contact with nature and animals on a farm can result in lower rates of allergies. Children who grew up on farms had the lowest rates of allergies. On the other hand, the allergy rate increased with increasing distance from the farm. The protective effect of farms against allergic diseases and the high numbers of microbes there is now known in science as the “farm effect” (*Table 1*, Illi et al. 2012, Genuneit et al. 2013, Depner et al. 2020, Pfefferle et al. 2010)

Some circumstantial evidence was also gleaned from comparing the microbiome of Hutterite and Amish children who share similar genetic backgrounds, lifestyles, and diets. However, Hutterite children are raised on highly industrial farms where contact with animals and the environment is limited, while Amish children live on traditional farms, where bacterial endotoxin levels are 6.8 times higher than in Hutterite habitats. Inhalation of Amish house dust extracts alone was sufficient to protect mice from experimental asthma (Stein et al. 2016). In previous studies, contact of children with farm animals and dogs during the first year of life and the associated higher microbial exposure was associated with a reduced risk of asthma at the age of 6 (Fall et al. 2015).

North Karielia: allergic Finns and “healthy” Russians

The 700-kilometer border between Russia and Finland runs right through the sparsely populated region of Karelia in the far north of Europe. As the two sides are alike from a geoclimatic point of view, they are just as different from a socio-economic point of view. And another important difference was found: over the last few decades of the 20th century, the rate of hay fever in Finnish Karelia had risen to 27%, even among school children. In Russian Karelia, however, allergic reactions such as hay fever were virtually non-existent. The allergic reaction situation particularly affected Finns who were born after the late 1970s. For people born in the 1940s, there was no significant difference between Finnish Karelia and Russian Karelia (Haahtela et al. 2015). A follow-up investigation showed that the allergy rate in Finland continued to increase, while it was still barely existent on the Russian side (Laatikainen et al. 2011). The reasons for these enormous differences are likely to lie in the different living conditions of these two populations living side by side. Exposure to environmental microbes of all kinds is many times higher in Russian Karielia. For example, on the Russian side, the microbial contamination of the drinking water was 19 times higher, and the house dust had a significantly higher number of germs and a higher bacterial diversity.

The largest bacterial reservoir, however, is the soil. The civilized society of the 21st century has largely lost contact with soil, be it directly, through drinking water, through house dust containing bacteria or through animals on a farm. The abundant saprophytic bacterial species in the soil (they feed on dead, organic substances) such as some mycobacteria could be of central importance for a differentiated immune system (von Hertzen and Haahtela, 2006).

Similarly, the authors of the Karelia study concluded that the lost contact with a diverse microbe-rich environment as well as an overall more modern, non-natural lifestyle, especially during childhood, contributed to the Finnish epidemic of allergic children. With regard to the previously postulated hygiene hypothesis and the old friends hypothesis, they coined the term "biodiversity hypothesis" (*Table 1*). Detailed studies have shown that low biodiversity in everyday living is indeed a risk factor.

Compared to healthy individuals, people with various allergic diseases have a lower ecological biodiversity in the vicinity of their home and a significantly lower diversity of Gammaproteobacteria in their skin microbiome. Bacteria from this class, such as Acinetobacter, produce the anti-inflammatory messenger substance interleukin 10 in healthy people (Hanski et al. 2012).

Soil microbes and their propagation in the environment

The possible routes of exposure to harmless "old friends" are therefore mainly through intensive physical contact with nature (soil, vegetation, air, water ...) or through the daily intake of food, 95% of which comes from the soil. In fact, the food we eat every day is the ultimate link between nature (our food comes directly from nature since eons) and our body. The actual contact area is considerable due to our enormous intestinal surface of at least 40 square meters.

Due to intensive interpersonal contact, there is of course the possibility of the microbes being passed on from person to person. In the event of intimate contact with animals, the environmental germs are transmitted via the respective animal. Ultimately, however, the microbes always come from the biosphere, of which soil is a tremendously important habitat.

Preserving soil biodiversity – future (agricultural) soil management

Land use intensification in particular widely applied agricultural management practices, are a major threat to soil biodiversity (El Mujtar et al. 2019). The soil function "genetic resources" is hence lost with species and constitutes a significant problem, as soil microbes are a source with tremendous potential for new antibiotic discoveries (de Castro et al. 2014, von Bubnoff, 2006). As mentioned above, the excessive use of herbicide can further jeopardize this unique potential. It is hence of highest priority to preserve our ecosystems by a more sustainable management to ensure soil and human health. Conservation of natural ecosystems and the restoration of land that is severely affected / damaged by human activities is a process that often takes decades for recovery. Efficient ecosystem functioning requires biodiverse soils. Life on earth is dependent on ecosystem services such as plant production / provision of food and regulating resource use efficiencies, nutrients and water (El Mujtar et al. 2019). To tackle the challenges we face today, it is crucial to developing sustainable agricultural / food systems that are healthy and of sufficient nutritional values, to feed a growing population (Lehmann et al. 2020). Sustainable agricultural intensification should emphasize the following practices (i) flower strips / hedgerows, (ii) conservation agriculture / regenerative agricultural practices to increase SOC or avoid SOC losses and stimulating soil biodiversity, (iii) organic farming, (iv) breeding programs.

Landscape elements such as flower strips, hedgerows or field fringes provide a habitat for both above- and belowground organisms. With more diverse plants, insects, birds, mice, groundgame the larger variety in the food-web improves ecosystem functions (i.e. pollination, control of pest outbreaks by natural predators, degradation of agrochemicals) (Marshall and Moonen, 2002). More diverse systems are suggested to be more resistant against the spread of invasive species (i.e. ragweed, earthworms or insects) (Geisen et al. 2019).

Diversification within the arable production system is another important option to sustain and recover core ecosystem functions that can be obtained by "regenerative agriculture". The practices include a crop rotation, cover crops and organic amendments (manure or compost), minimized soil tillage, as well as integrated pest management, agroforestry and silvopasture (combining trees and grazing) (Lehmann et al. 2020). There is a major aim to increase soil SOC or to counteract SOC losses, via these management practices. In particular, reduction in tillage intensities or organic amendments may boost, soil microbes and their transformation and mineralization activities, to support plant nutrition and growth, and thereby add to crop health and resilience via improved biodiversity (Cesarano et al. 2017). A boost of mutualistic soil microbes via the soil organic amendment application reduces the vulnerability against plant pathogens (Bonanomi et al. 2018). Improved soil (micro)nutrient availability and crop nutritional content, is related with greater SOC, and varies with crop type, hence a vital soil essential to counteract dietary deficiency (Jacoby et al. 2017).

Organic agriculture has been reported to positively impact soil microbial biomass and enzyme activities on a global scale (Lori et al. 2017). However, strongly depended on soil type and land use, organic inputs, crop rotations and intercropping were mentioned as the most powerful implementations (Lehmann et al. 2020). Higher microbial richness, and lower evenness after long-term organic farming systems can be beneficial for sustaining ecosystem functions and to counteract productivity losses under more intense drought with changing climate (Hartmann et al. 2015). Reduced pesticide residues in organic fruits and vegetables is beneficial for consumers (Yu and Yang, 2017). Additionally, the risk for resistances in soil can be diminished, as it was reported that herbicides can induce a change in susceptibility to multiple antibiotics in potentially pathogenic bacteria (Kurenbach et al. 2017). Exposition of bacteria to herbicides and (manure derived) antibiotics can result in the evolution of mutations that show a higher level of resistance (Kurenbach et al. 2018). Breeding programs should emphasize on the new needs of plants to select for crop species with enhanced defense mechanism and pest resistance, more efficient water and nutrient usage. Wild ancestors of the crops may provide a valuable resource for reconstructing functional plant traits and the interaction between plant and rhizosphere microbial communities (Pérez-Jaramillo et al. 2018).

Conclusions

A strong linkage between soil and human health has been recently proposed (Blum et al. 2019). This fits to the concept of the "holobiont" which considers the host (i.e., human, plant) and the numerous species that inhabit the host as one ecological unit. The relationship between soil and the future of humans is often exclusively seen under the aspect of the use of soil products, e.g., the provision of food and water. But soil is also directly influencing human health because soil and the human intestine are the most important microbial pools on earth, which may have phylogenetically developed together. The direct solid and dust contact between the soil and humans is of outstanding significance for the adaptation and restoration of the human microbiome. Human health is increasingly influenced by adverse environmental impacts on soils, for example by intensive agriculture, using pesticides and antibiotics. In addition, intensive agricultural practices, causing soil compaction and erosion, is harming the soil microbial habitat. In view of the future of human health, the improvement of our environmental biodiversity, sustainable land management, and fostering the direct contact to the natural environment in our daily lives is of paramount importance.

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Chapter 2

Microbial biodiversity in Mediterranean soils, a challenge in a changing world

Fuensanta García-Orenes and Minerva García-Carmona

Introduction

Soils represent the most biologically diverse and important ecosystem on the planet (Roger-Estrade et al. 2010). Most of the biodiversity of ecosystems is found in the soil (Young and Crawford, 2004), about one gram of soil may typically contain one billion bacterial cells, that corresponds to about ten thousand different bacterial genomes, up to one million individual fungi, about one million cells of protists, and several hundred of nematodes (EU, 2010a). In this sense, soils are a key reservoir of global biodiversity, yet little is known about them as only 1% of soil microorganism have been identified (FAO et al. 2020) compared to 80% of plants (Jeffery et al. 2010). Soils are remarkably complex and dynamic environments and hence typically comprise a wide range of habitat types for organisms over a range of dimensions from micrometre to the landscape scale. The highly heterogeneous nature of the soil, particularly at the microhabitat level, is responsible for the considerable biodiversity (Jeffery et al. 2010).

The functions performed by soil biota have considerable direct and indirect effects on crop growth and quality, nutrient cycle, quality, and soil sustainability. Moreover, the biodiversity of soil is vital as it is the engine driving soil-based ecosystem services such as carbon sequestration, soil formation, decontamination and bioremediation of pollutants, control of pest outbreaks, and water purification (Dominati et al. 2010, Turbé et al. 2010). Soils with higher biodiversity express more resistance and resilience to perturbations, thus a loss in biodiversity can lead to lower resistance to a perturbation and reduced capacity to recover, affecting the ability of soil to function normally (Brussaard et al. 2007, Allison and Martiny, 2008).

The major threats for soil biodiversity loss include deforestation, urbanization, agricultural intensification, soil organic matter decline, soil compaction, surface sealing, soil acidification, nutrient imbalance, contamination, salinization, sodification, land degradation, fire, erosion, and landslides (FAO et al. 2020). Those threats are of major relevance under Mediterranean conditions based on a strongly seasonal climate with scarce and irregular rainfall and frequent drought periods, which can trigger these processes accelerating the loss of biodiversity.

There is not a specific European regulation or legislation about soil biodiversity conservation, but the European Commission acknowledged the importance of soil biodiversity in the role of ecosystem functioning, stating that “these functions are worthy of protection because their socio-economic as well environmental importance” (Stone et al. 2016). Moreover, “biodiversity pool, such as habitats, species and genes” soil functions are collected in the SDG number 15 of the UN “Sustainable Develop Goals (SDGs)” for the period 2015-2030, by relating the topics “ensure healthy lives and promote well-being for all at all ages” and protect, restore, and promote sustainable use of terrestrial ecosystem, sustainable manage forest, combat desertification, and halt and reverse land degradation and halt biodiversity loss (Keesstra et al. 2016).

The mediterranean characteristics

The Mediterranean-type climate regions are distributed over five continents: Africa, Australia, Europe, North America, and South America. The most representative is the Mediterranean basin, extended over 3800 km from east to west in south Europe, and whose environmental and climatic special factors contribute to exposing soils to adverse processes that make them very vulnerable and condition the microbial biodiversity. The long periods of droughts interrupted by heavy, occasionally torrential rainfall (200-700 mm), together with high annual mean temperatures (16º-19ºC) and high radiation (EU, 2010b), increases rates of erosion in soils prone to degradation.

Soil degradation would be exacerbated in the most vulnerable Mediterranean ecosystems according to the climatic predictions, the arid and semi-arid areas, which especially represented in SE Spain. In those regions, the most widely represented lithological substrates are carbonate rocks, quaternary sediments, and loams, altogether materials easily eroded. Moreover, agriculture has been practised in the semi-arid Mediterranean for ca. 10 000 years, thus centuries of ploughing, burning, and grazing have resulted in the clearing of natural potential vegetation. This combination results in large areas of low-quality soils with little plant cover. Furthermore, the low organic matter content of large extensions, a key factor describing the Mediterranean soils, determines the quality and fertility of soils.

The combination of all these factors implies Mediterranean soils are especially sensitive and vulnerable to soil erosion, and little changes could trigger several degradation processes that ultimately affect soil biodiversity. In this sense, keep the soil biodiversity in Mediterranean soil is a real challenge in a scenario of climate change, fire-recurrence increment, and land-use intensification in the Mediterranean basin.

Millennian Mediterranean agriculture

The Mediterranean basin has a millenary history of intensive and extensive land use, many cultures have evolved in the area generating socioeconomic and land-use changes. Agriculture has been practised for more than one millennium, transforming large extensions of landscapes in terraced slopes for agricultural purpose and natural vegetation to crops lands (Pausas et al. 2009).

Agricultural management influences soil microorganisms and soil microbial processes through changes in the quantity and quality of plant residues entering the soil, altering their spatial distribution and through physical changes (Christensen, 1996). It has been reported intensive arable farming causes a progressive decline in soil organic matter levels (Caravaca et al. 2002), which affects physical, chemical, biochemical and biological soil properties. Intensively used agricultural lands treated with soil tillage often receive high levels of mineral fertilizers and pesticides. The excessive use of herbicides can modify drastically the function and structure of soil microbial communities, thus altering the normal functioning of terrestrial ecosystems, which in turn has important implications for soil fertility and quality (Pampulha and Oliveira, 2006). Since many of the agricultural practices commonly used provoke negative impacts on microorganisms and their processes, the application of a conservation management system tries to minimize soil disturbance.

Agricultural land management is one of the most significant anthropogenic activities that greatly alter soil characteristics, including physical, chemical, and biological properties (Jangid et al. 2008, García-Orenes et al. 2010). This fact is particularly relevant in Mediterranean environments, where unsuitable land management along with climatic constraints (scarce and irregular rainfall and frequent drought periods) can contribute to increased rates of erosion and other soil degradation processes in agricultural land (Caravaca et al. 2002). These conditions can lead to a loss in soil fertility and a reduction in the abundance of biodiversity in soils. This is of especial importance in Europe, where more than 45% of the land is for agricultural production (EUROSTAT, 2019).

Differences in agricultural production systems, such as integrated, organic, or conventional, have also reported affecting the soil biota, both the overall biomass as well as biodiversity. Indeed, soils subjected to disturbance by tillage can be more susceptible to reductions in soil biodiversity due to desiccation, mechanical destruction, soil compaction, reduced pore volume, and disruption of access to food resources (Giller, 1996). Some organic fertilisers, such as manure and sewage sludge, promote the activities of soil microbial communities (Enwall et al. 2007); however, repeated application of manures may pose environmental hazards, as they introduce faecal microbial flora into the soil, and have the potential to alter the endogenous microbial structure (Soupir et al. 2006). In many cases, the agricultural activity implicates the use of water for irrigation without enough quality that contains a high amount of salt and organic pollutants, which ultimately has an important effect on soil biodiversity (Friedel et al. 2000, Mangkoedihardjo, 2006). A major threat is salinization, the accumulation of soluble salts of sodium-calcium, potassium and magnesium in soils causing deterioration or loss of one or more soil functions. Salinization of soils occurs either as a result of natural processes or as a consequence of mismanagement of irrigation practices or poor drainage conditions.

On the other hand, soil management implicates in many cases the loss of organic matter, especially relevant in Mediterranean soils that are submitted to semi-arid conditions (Novara et al. 2011, Laudicina et al. 2015). A reduction in soil organic matter is generally associated with a lower soil organism abundance and diversity. Soil biodiversity is intimately bound to soil organic matter content (García-Orenes et al. 2013), each type of soil organism occupies a different niche within the food web of life and favours a different substrate and nutrient source. Consequently, a large, varied source of organic matter will generally support a wider variety of organisms due to the greater range of substrates and nutrients content, for example, functional microorganisms related to N cycling. The use of various types of organic amendments in agricultural soils can lead to an improved potential for N capture and release and therefore contribute to the conservation of soil biodiversity in agricultural land (Pereg et al. 2018a). Organic agriculture management promotes soil structure and fertility as well as increases water infiltration and storage, of major importance in the degraded Mediterranean soils.

The Mediterranean wildfire

Forest fires are a common natural disturbance in the Mediterranean ecosystems, the marked seasonality with long droughts periods and frequent temperatures above 30ºC provides ideal conditions for fuel accumulation and the ignition and fire spread. However, anthropogenic impacts in the last decades in the Mediterranean basin such as changes in land use, agricultural abandonment, and fire suppression policies, have led to an increase in fire severity regimens aggravated in a context of climate change (Pausas and Keeley, 2009). Wildfires have become one of the most critical threats to the Mediterranean forests in Spain, in which the predicted climate change scenario, with expected rainfall decrease and drought risk increase and consequently forest wildfires, make them especially vulnerable. The loss of organic matter through fire and the diminution of vegetation cover contribute to increasing rates of erosion and other soil degradation processes, resulting in the loss of soil ecosystem functionality.

Fire effects on soil biodiversity can vary widely, depending on fire severity, the changes in some soil properties and post-fire environmental conditions. Each of these factors has a range of effects depending on the type of soil organism being considered (Certini et al. 2021). Despite the protective mechanisms, fire can lead to major shifts in the fire-adapted Mediterranean ecosystems. Fire can affect soil microbes directly through heating and indirectly by modifying soil properties. Microbes will also be affected by post-fire environmental factors and the re-establishment of vegetation. The most important factor affecting soil biodiversity seems to be the burn severity, which is controlled by such factors as fire intensity, duration, and soil properties, which normally causes a decrease in the numbers of microbes. The temperatures reached in the topsoil are often sufficient to affect soil microorganisms and other soil properties related to the post-fire microbial recolonization. In extreme cases, the topsoil can undergo complete sterilization. Fungi seem to be more sensitive to heat than bacteria, and a higher impact under wet soil conditions has been reported. In the case of fungi that form arbuscular mycorrhizas, almost all the studies show a negative influence resulting in a reduced number of propagules (Mataix-Solera et al. 2009).

The current level of knowledge sets that although burning strongly reduces belowground biomass and its functioning, it may recover within a few years after burning. However, previous and subsequent forest management practices to the wildfire are of major importance affecting fire impacts in soils and soil microorganisms (Lucas-Borja et al. 2019, Mediavilla et al. 2020). Especially, post-fire management on burned soils can be a key to promote soil biodiversity recovery or, on the other hand, a threat that hinders the restoration of the belowground functions. Several studies have reported salvage logging, the most common post-fire strategy carried out in fire-affected areas in Europe performed to recover timber values, could hamper the soil biodiversity recovery via soil compaction (Hartmann et al. 2014, García-Orenes et al. 2017, Pereg et al. 2018b). However, the effects of post-fire management on microbial community composition and structure and its consequences for ecosystem recovery remain partially unknown, being these of especial importance in Mediterranean forest where soils are prone to degradation. Moreover, the functional resilience and sustainability in fire-prone ecosystems are threatened by the increasing predictions of fire danger and burned areas, due mainly to global warming, hence in need to be addressed (FAO et al. 2020).

Examples of research work carried out in the Mediterranean area related to soil biodiversity

Since the late 1980s, soil biological parameters have been assessed in an increasing number of studies (Pulleman et al. 2012). The study of soil microbial structure, function, and biodiversity has been considered essential to understand the soil ecosystem and keep its quality and fertility in the last decade. In this sense, several experiments developed in the Mediterranean area under semiarid conditions have been selected as examples of the effect of different human activities on soil biodiversity.

Agricultural management effect on soil microbial properties

The different agricultural practices can produce important changes in microbial structure, functionality, and biodiversity. In general, sustainable agriculture practices enhance soil microbial biodiversity in contraposition to conventional practices. As an example, an experiment developed on erosion plots in an agricultural area of the Spanish Mediterranean (38º50´N; 0º42´W) discerned the effect of five different treatments on soil microbial structure (Garcia-Oerenes et al. 2013). The treatments tested were based on some of the most common practices used by farmers in the east of Spain, as residual herbicides, tillage, tillage with oats and oats straw mulching, practices which were evaluated against an abandoned land after farming and an adjacent long term wild forest coverage. The results showed a substantial level of differentiation in the microbial community structure in terms of management practices (*Figure 1*), which were highly correlated with the soil organic matter content. The application of oat straw to soil increased the organic carbon content and, as a result, a significant increase of the microbial biomass was registered. The total PLFAs were highly correlated with the microbial biomass carbon, determined by the fumigation-extraction method. The correlation obtained in this research between these parameters (r= 0.80) was similar to the reported in previous works (Zornoza et al. 2009, Hackl et al. 2005). These results corroborate the notion the microbial community structure is a good indicator of soil quality, perturbations, and the effects of different management practices (Zornoza et al. 2009) since the microorganisms respond against changes in soil management more rapidly than chemical or physical soil properties.

In the same area, a field experiment in a vineyard was performed after 10 years to assess the medium-term effect of mineral fertilizer and two organic fertilization systems with different nitrogen sources on the soil microbial community biomass, structure, and composition (phospholipid fatty acids, pattern, and abundance), and microbial activity (basal respiration, dehydrogenase, protease, urease, β-glucosidase, and phosphomonoesterase activity) (*Figure 2*) (García-Orenes et al. 2016). The three fertilization systems assayed were: inorganic fertilization, the addition of grapevine pruning with sheep manure (OPM), and addition of grapevine pruning with a legume cover crop (OPL). The organic fertilization systems increased microbial biomass, shifted the structure and composition of the soil microbial community, and stimulated microbial activity, compared with inorganic fertilization (García-Orenes et al. 2013, Zhang et al. 2005). The abundances of fungi and Gram+ bacteria were increased by treatments OPM and OPL. The total replacement of inorganic by organic fertilization in a semiarid agroecosystem had a significant medium-term effect on the biomass, composition, and function of the microbial community, which may be attributed to the organic carbonaceous substrates and nutrients introduced into the soil. In particular, the addition of grapevine pruning combined with sheep manure, or a legume cover crop promoted the proliferation of fungal and Gram+ bacterial populations in the soil microbial community. Shifts in soil microbial populations and microbial processes related to nutrient cycling promoted by the use of organic fertilizers improved soil fertility, maintaining crop yields at levels similar to those of the inorganic fertilization system. The effectiveness of the organic fertilization systems, for promoting the sustainability and soil biological and chemical fertility of an agroecosystem under semiarid conditions, was dependent on the organic N source.

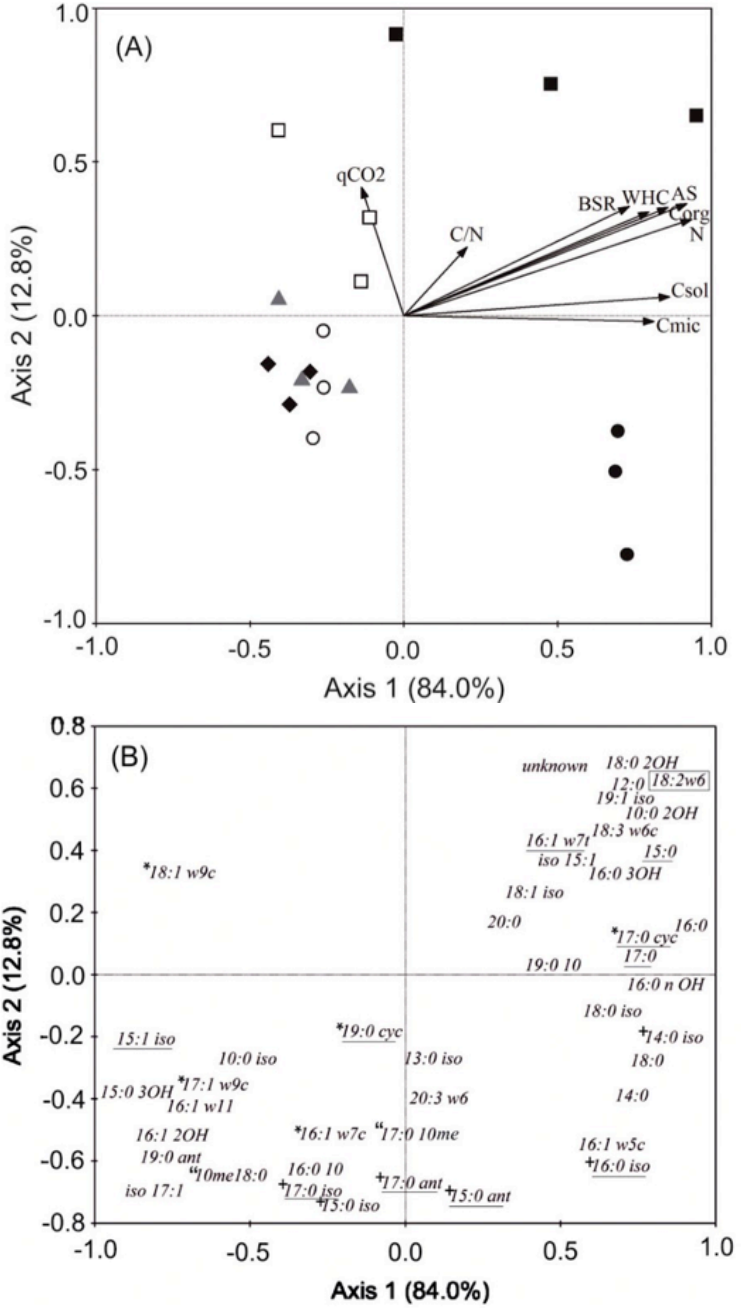


Figure 1. Samples and soil characteristics biplots (A) and loadings plots (B) from RDA performed on the relative concentration of PLFAs in all management practices: residual herbicide (♦), tillage (▲), oats + tillage (○), oats straw (●), land abandonment (□) and wild forest coverage (■). PLFAs used for microbial groups designation are marked as: underlined (bacteria), framed (fungi), * (Gbacteria), + (G+ bacteria) and (actinobacteria). Corg: soil organic carbon; N: total nitrogen; Csol: soluble carbon; WHC: water holding capacity; AS: aggregate stability; Cmic: microbial biomass C; BSR: basal soil respiration; qCO2: BSR/Cmic. (García-Orenes et al. 2013).

Figure 2. Vineyard crop with organic fertilization situated on Mediterranean area under semiarid conditions (39º49´24”N; 0º48´17”W).

In this agricultural area, the effect of these treatments on the nitrogen cycling microbial community of soil was investigated by quantification of genes involved in key pathways, in particular nitrogen fixation, denitrification and nitrification (Pereg et al. 2018a). The assimilation of inorganic N by soil microorganisms is critical for N retention in soil, thus reducing the loss of N fertilizer to the environment (Tahovská et al. 2013), particularly important in degraded agricultural lands. In addition, N is often applied in forms that are less efficient or unavailable for uptake by plants, such as urea (Witte, 2011) or organically bound N, making microbial N cycling a key process required for N transformation in soils (He et al. 2010). Nitrification and denitrification are major pathways in the soil N cycle, involving ammonia oxidation and nitrate and nitrite reduction to N_2O and N_2 (reviewed by Teixeira et al. 2012), respectively. Elucidating the impact of N application practices on microbial diversity and community structure in general, and on N cyclers in particular, is pivotal to identify agricultural practices that enhance the soil potential for microbial immobilization and transformation of fertilizer N. The results of the study showed the abundances of bacterial *nif*H, *nos*Z, *nir*S and *nir*K genes, these were significantly increased under a decade of organic fertilization when compared to inorganic fertilization (*Figure 3*), linked to an increase in soil organic carbon. The abundance of *nif*H was lower where fertilizers rich in ammonia and nitrate were used, and its increase under organic fertilization was more related to the availability of organic carbon than to the nature of the organic amendment. Our results indicate that soil microbial communities involved in biological nitrogen cycling, in particular nitrogen fixation and denitrification, are more abundant under management practices that include organic fertilization compared to traditional agricultural practices. A decade of organic amendments using grapevine pruning, combined with sheep manure or legume cover crop, resulted in an enhanced grapevine soil potential for N fixation and denitrification, two microbial processes that are essential for input and output of N into the biosphere. The raise in diazotrophic and denitrifying communities, shown by increased *nif*H, *nir*S, *nir*K and *nos*Z gene copy numbers under organic management, comes hand in hand with a significant improvement in biological and physical soil properties. It may therefore be recommended that organic fertilization should be utilized in agricultural systems to assist with sustaining healthy soils.

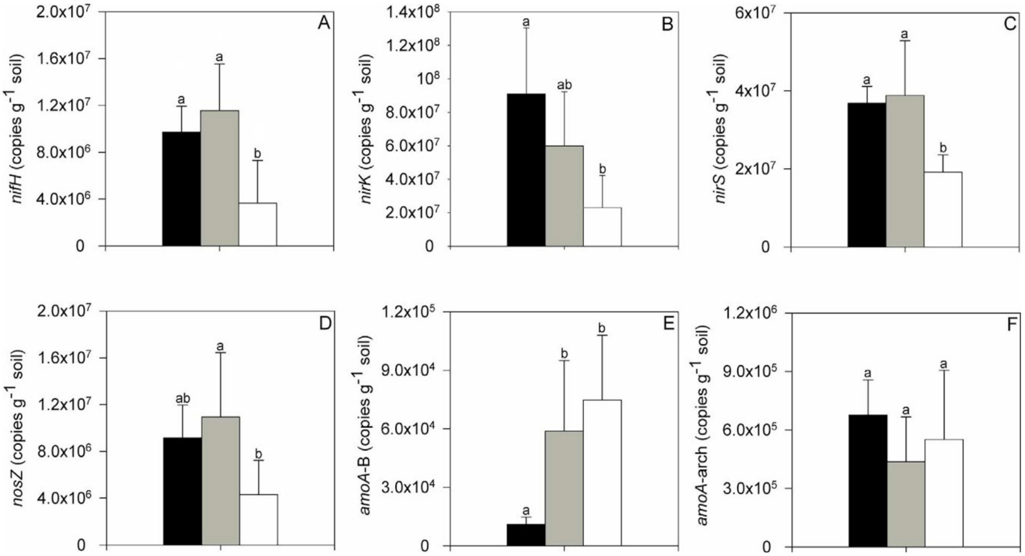


Figure 3. Mean values (±standard deviation) of nitrogen cycle genes in grapevine soil. Different letters indicate significant differences (one-way ANOVA, $P < 0.05$) between treatments. Bars: white (IF), black (OPM) and grey (OPL). (Pereg et al. 2018).

Water shortage is a major problem faced by agricultural industries in the Mediterranean area. With currently available water levels below 300 mm per year, this situation is progressively worsening as a result of climate change (IPCC, 2013). The scarcity of water is problematic in many areas of southern Spain, where groundwater is used for irrigation in 27% of the irrigated agricultural soils and the rest of irrigation is provided by surface water (Lidón et al. 2013). In southern Spain, there is 158,859 ha of citrus orchards, representing 53.5% of the total citrus orchards in Spain, of which about 70,162 ha are sweet orange. The most common irrigation system in sweet orange orchards in this region is drip irrigation (64% is drip irrigation versus 36% flood irrigation) (MAPAMA, 2017).

As a necessary issue to address, it was evaluated how different soil moisture levels, dependent on distance from drip irrigation points, impact the biological, properties of citrus soil under organic and inorganic fertilization, soil microbial community structure (phospholipid fatty acid assay), and abundance of microbial nitrogen cyclers (by quantitative PCR). A field experiment was performed in an orange orchard (Citrus sinensis) (Morugán-Coronado et al. 2019) in southeast Spain. 30 and 18 soil samples were taken from each plot to compare the impacts of soil moisture: near (wet, w) or away (dry, d) from drip-irrigation points, in plots with inorganic fertilizers under intensive ploughing (PI) or organic fertilization (OA) (*Figure 4*).

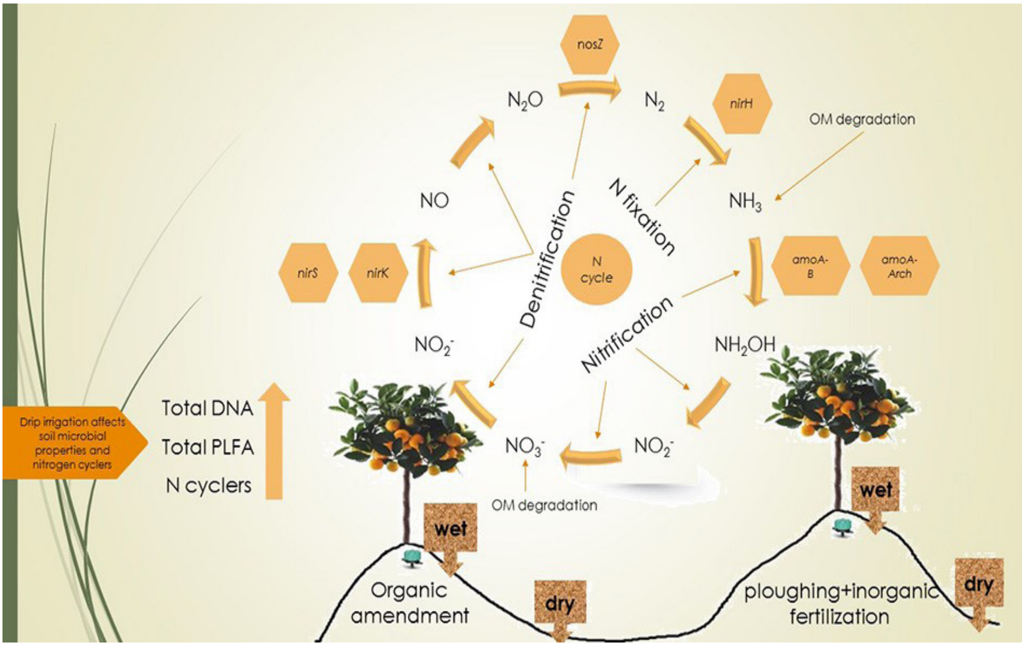


Figure 4. Diagramming of the experiment to evaluate the effect of moisture on soil microbial properties and nitrogen cyclers in Mediterranean sweet orange orchards under organic and inorganic fertilization. (Morugán-Coronado et al. 2019).

The results showed that changes in microbial properties and soil microbial indexes were strongly associated with soil moisture content under both organic and inorganic fertilization, and with organic carbon content. Soil moisture influenced bacterial and fungal load (PLFAs) (*Table 1*), and the abundances of bacterial N cycling genes, including nifH (nitrogen fixation) *nir*S/K and *nos*Z genes (denitrification) and amoA-B (bacterial nitrification) (*Table 2*). The potential for N fixation and denitrification, two microbial processes that are crucial for determining the amount of N in the soil, were improved by increased soil moisture in the proximity of the drip irrigation. Soil organic carbon and total N, which are usually higher under organic fertilization than under inorganic fertilization, were also highly correlated with the abundance of the N cycling genes. By controlling irrigation doses and applying organic amendments, it may be possible to increase the microbial abundance and function in soil, thus supporting greater fertility of soils.

	OAd	OAw	Pld	Plw
Total PLFA (nmol g^{-1})	71.1±2.0^{a}	260±4.5^{b}	22.5±0.1^{c}	29.2±2.1^{d}
Gram+ PLFA (nmol g^{-1})	12.2±2.3^{a}	26.6±1.7^{b}	bdl	0.4±0.1^{d}
Gram- PLFA (nmol g^{-1})	31.9±2.90^{a}	166.8±2.3^{b}	3.60±0.3^{c}	15.0±2.1^{d}
Fungi PLFA (nmol g^{-1})	25.5±0.9^{a}	55.0±2.4^{b}	18.8±0.4^{c}	13.4±0.5^{d}
Actinobacteria PLFA (nmol g^{-1})	1.4±0.4^{a}	11.7±2.4^{b}	bdl	0.4±0.1^{d}
Fungi/bacteria	0.6±0.1^{a}	0.3±0.1^{b}	5.2±0.6^{c}	0.9±0.1^{d}

Values are mean ± standard deviation (n=36)

A one-way ANOVA ($P<0.05$) was used to compare differences between treatments. Values in rows sharing the same letters do not differ significantly

bdl: below detected levels (<0.1 nmol g^{-1})

Table 1. Total fatty acid content and abundance of signature phospholipids fatty acids of different treatments (**OAd**: organic amendment management without irrigation; **OAw**: organic amendment management with irrigation; **Pld**: ploughing and inorganic fertilization without irrigation; **Plw**: ploughing and inorganic fertilization with irrigation) (n=36) (Morugán-Coronado et al. 2019).

Code	*nif*H gene copies g^{-1} soil (x10^{5})	*nir*S gene copies g^{-1} soil (x10^{6})	*nos*Z gene copies g^{-1} soil (x10^{6})	*nir*K gene copies g^{-1} soil (x10^{7})	*amo*A-B gene copies g^{-1} soil (x10^{4})	*amo*A-arch gene copies g^{-1} soil (x10^{4})	16S rRNA copies g-1 soil (x107)
OAd	72.7±8.9^{a}	29.5±3.5^{a}	29.0±2.6^{a}	14.5±6.1^{a}	8.5±0.3^{a}	30.0±10.4^{a}	0.1±0.1^{a}
OAw	223.1±20.7^{b}	78.4±9.0^{b}	40.1±3.9^{b}	34.6±12.3^{b}	34.4±1.0^{b}	26.4±9.5^{a}	1.1±8.2^{a}
Pld	3.1±0.9^{C}	2.4±0.4^{c}	1.8±0.2^{c}	0.6±0.3^{c}	3.95±0.7^{c}	3.2±2.2^{b}	1.4±1.9^{a}
Plw	5.5±0.2^{C}	4.9±0.4^{c}	5.1±0.4^{d}	2.8±1.3^{d}	15.1±1.6^{d}	5.4±3.1^{b}	2.9±2.4^{a}

Values are mean ± standard deviation (n=36)

A one-way ANOVA ($P<0.05$) was used to compare differences between treatments. Values in rows sharing the same letters do not differ significantly.

Table 2. Copy numbers of genes related to the nitrogen cycle in soils under different agricultural managements (**OAd**: organic amendment management without irrigation; **OAw**: organic amendment management with irrigation; **Pld**: ploughing and inorganic fertilization without irrigation; **Plw**: ploughing and inorganic fertilization with irrigation) (n = 36) (Morugán-Coronado et al. 2019).

Post-fire management effects on soil microbial properties

Wildfires can impact soil productivity of burnt areas (Robichaud, 2009), especially when are affected by huge and high-intensity fires such as those that are frequent in summer seasons under extreme weather conditions in the Mediterranean area. The negative impacts can be exacerbated after the post-fire management, which in some cases are even more severe than the fire itself. Salvage logging (SL), one of the most common management techniques in fire-affected areas, which comprise the extraction of the burnt wood and in many cases using heavy machinery and dragging the trunks over the soil, is an example of a strategy that can lead to an increase in soil vulnerability to erosion and degradation (Mataix-Solera et al. 2016).

After a wildfire in "Sierra de Mariola Natural Park" in Alicante (Spain), a study was developed to evaluate the impact of post-fire salvage logging on soils. The implementation of the management triggered severe soil physic-chemical degradation, with a critical decrease in the nutrient content and soil structure deterioration, and a decrease in the microbial biomass and activity (García-Orenes et al. 2017). In addition, impacts of SL on soil microbial communities were assessed, specifically on the abundance of nitrogen cycles and, thus, the potential of the soil for microbial nitrogen cycling (Pereg et al. 2018b). It was demonstrated that salvage logging reduced bacterial load compared to tree retention control, resulting in significant changes to the abundance of functional bacteria involved in nitrogen cycling (*Figures 5 and 6*). Microbial gene pools involved in various stages of the nitrogen cycle were larger in control soil than in soil subjected to post-fire salvage logging and were significantly correlated with organic matter, available phosphorous, nitrogen and aggregate stability (*Table 3*). The impact of post-fire management strategies on soil microbial communities needs to be considered in relation to maintaining ecosystem productivity, resilience, and potential impact on climate.

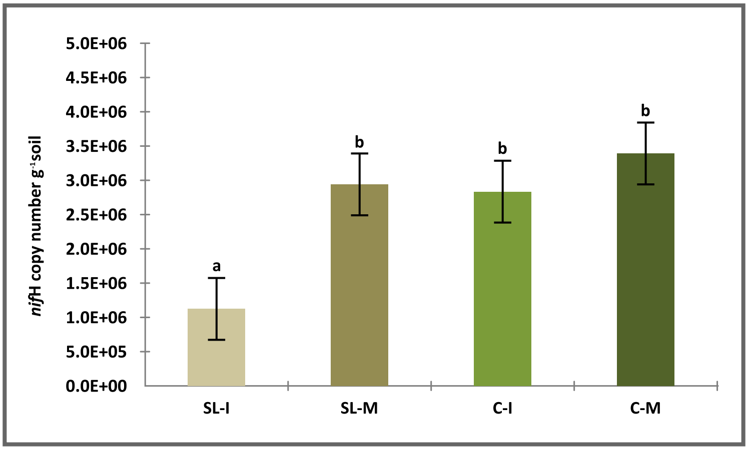


Figure 5. Abundance of nifH gene (mean ±standard deviation) in soil with different treatment after one-way ANOVA. SL-I: soil under salvage logging treatment sieved at 2mm; SL-M: soil under salvage logging treatment sieved between 63 and 250 µm; C-I: control soil with tree retention sieved at 2mm; C-I: control soil with tree retention sieved between 63 and 250 µm. Different letters above the bars indicate significant differences (Pereg et al, 2018b).

Parameter[a]	OM	N	AP	AS	MBS	BSR	*nif*H	*nos*Z	*nir*S	*nir*K	amoA-Arch	amoA-B	16rDNA	Total DNA
OM	1	0.95**	0.91**	0.77**	0.75**	0.78**	0.70**	0.47*	ns	ns	ns	ns	0.76**	0.84**
N		1	0.88**	0.81**	0.80**	0.70**	0.68**	ns	ns	ns	ns	ns	0.79**	0.83**
AP			1	0.78**	0.66**	0.75**	0.74**	0.49*	ns	ns	ns	ns	0.63**	0.70**
AS				1	0.69**	0.54*	0.64**	0.56*	ns	ns	ns	ns	0.76**	0.63**
MBS					1	0.51*	ns	ns	ns	ns	ns	0.62**	0.72**	0.59**
BSR						1	0.53*	0.66**	ns	ns	ns	ns	0.53*	0.71**
*nif*H							1	0.67**	ns	ns	ns	ns	ns	0.67**
*nos*Z								1	ns	ns	0.56*	ns	ns	0.59*
*nir*S									1	ns	0.56*	ns	0.65**	0.66**
*nir*K										1	ns	ns	ns	ns
amoA-Arch											1	ns	ns	ns
amoA-B												1	ns	ns
16rDNA													1	0.77**
Total DNA														1

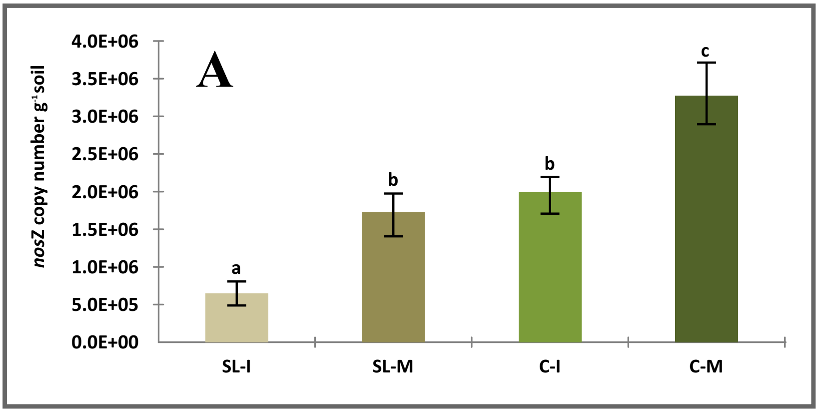
ns: not significant; 16S rDNA: 16SrRNA gene. The results were confirmed using a non-linear method (Spearman) a similar value was obtained.

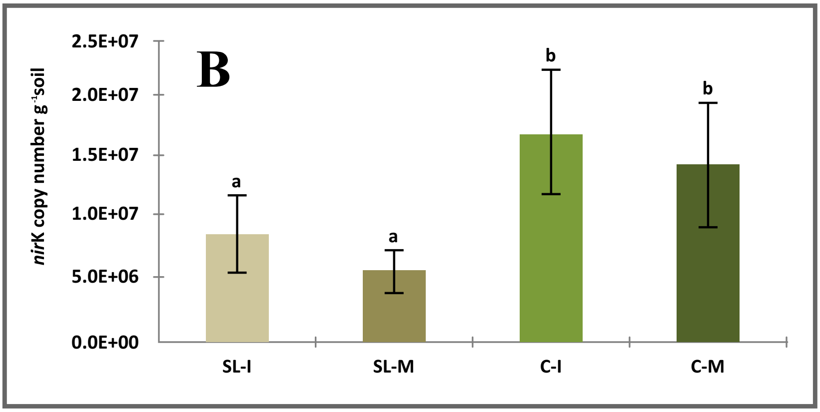
a OM: soil organic matter; N: Kjeldahl nitrogen; AP: available phosphorus; AS: aggregates stability; MBC: microbial biomass carbon; BSR: basal soil respiration.

* Significant at $P<0.05$

**Significant at $P<0.01$

Table 3. Correlation coefficients (r values) for relationships between the different physico-chemical soil properties determined and genes of soil (n=18).





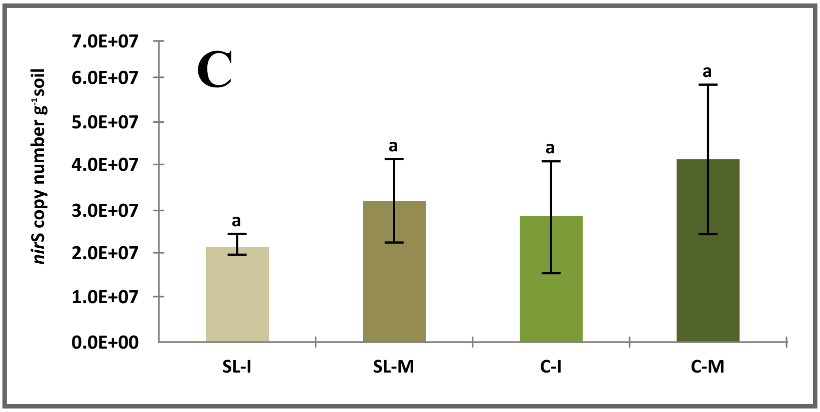


Figure 6. Abundance of *nos*Z gene (A), *nir*K gene (B) and *nir*S gene (C) (mean ± standard deviation) in soil with different treatment after one-way ANOVA. SL-I: soil under salvage logging treatment sieved at 2 mm; SL-M: soil under salvage logging treatment sieved between 63 and 250 µm; C-I: control soil with tree retention sieved at 2 mm; C-I: control soil with tree retention sieved between 63 and 250 µm. Different letters above the bars indicate significant differences. (Pereg et al. 2018b)

The degradation of soil structure- promoted by the decrease in organic matter content and reduced plant development after the wildfire and the post-fire salvage logging- resulted in the main driver in the community composition and structure shifts of the bacterial and fungi in a study of amplicon sequencing (García-Carmona et al. 2021). Soil degradation induced by logging operations resulted in new niches related to anoxic habitats, being Proteobacteria and Firmicutes families capable of anaerobic respiration found in high levels in the affected soils (*Figure 7*). In addition, the depletion in C and N nutrients as a consequence of the soil erosion reduced the microbial populations sensible to substrates availability, e.g., Actinomycetales. Ascomycota increased proportionally in managed soils, which might be due to the removal of host plants dependent on ectomycorrhizal fungi. The study demonstrated that physical soil disturbance related to post-fire management could seriously hamper the resilience of soil microorganisms after a fire perturbation.

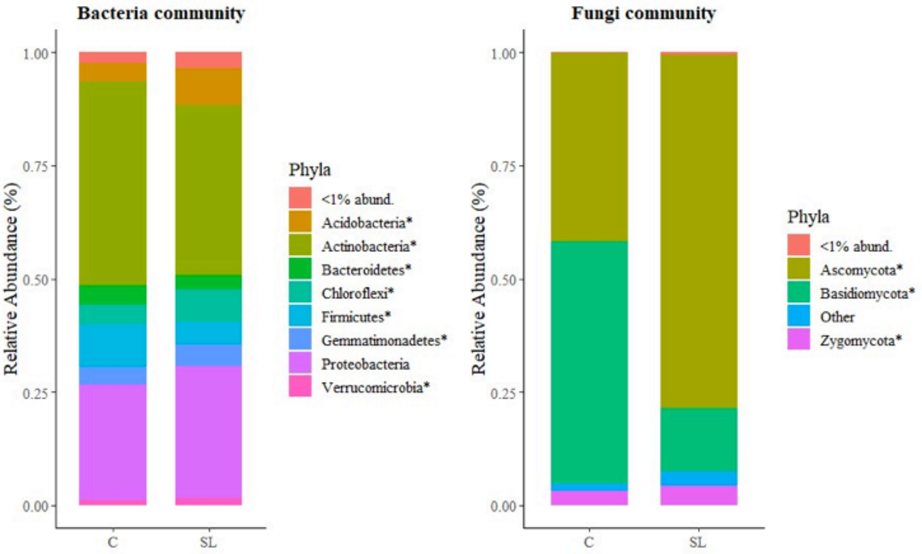


Figure 7. Relative abundances of the dominant bacterial and fungal phyla of control (C) and salvage logging (SL) soils. For each phylum, significant differences were assessed by the t-test calculated at $p<0.05$ and indicated by an asterisk. (García-Carmona et al. 2021).

The early post-fire vegetation colonizing has special relevance for the Mediterranean soils, the absence of vegetation cover exposes soils to rainfall events, runoff, and erosion processes. Ruderal mosses, forming part of the biological soil crust, have been pointed as early successional species colonizing burned soils in Mediterranean ecosystems, thus revealing important roles such as soil stabilization. A study in the same study area revealed that the presence of mosses played a significant role in soil fertility improvement and microbial activity six years after the wildfire (García-Carmona et al. 2020). In addition, SL management negatively affected the percentage of soil covered by mosses, decreasing 22% in SL soils. Considering the presence of mosses in the post-fire management was reveal of major importance given their role in Mediterranean ecosystem functioning.

Conclusions

The soils in the Mediterranean area extremely sensible to agro-forest management due to their particular environmental conditions. Therefore, agricultural practices or post-fire action can modify the equilibrium of microbial populations that can be reflected in a loss of biodiversity. In the Mediterranean area, it is essential to look for the best sustainable management practices of the soil to keep the fertility, quality, and microbial biodiversity of the same.

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Chapter 3

Microbial biodiversity and soil functions restoration

Rosalia Garcia Teijeiro and Andreas Bernreiter

> *“The entire range of living matter on Earth from whales to viruses and from oaks to algae could be regarded as constituting a single living entity; capable of maintaining the Earth’s atmosphere to suit its overall needs and, endowed with faculties and powers far beyond those of its constituent parts.”* – Gaia: A New Look at Life on Earth, 1979. James Lovelock

Soil genesis, function, and ecosystem services

Soils emerge from long evolutionary processes and are shaped by biological, chemical, climate, and topological forces operating over the Earth’s rocky crust. Needing more than 3000 years for one cm to emerge, soils are considered non-renewable resources (Ponomarenko, 2015). Soil scientists describe the soil as a matrix of solid mineral structures combined with gases, water, and a diversity of organic compounds (Baldock and Skjemstad, 2000). They also describe another soil characteristic: the arrangement in layers or horizons (Coleman et al. 2004). Depending on the age and location of the soil is how well defined the horizons will be. Typical soil horizons are: O of organic matter from biota, and A horizon makes the topsoil. Topsoils are often rich in organic matter, allowing an extraordinary variety of biological activity. Then, follow the subsoil B with less organic matter than A and more minerals content. Finally, between the B horizon and the parent bedrock lies the C horizon born from the weathered bedrock R and possesses little life.

The soil matrix provides an immense variety of habitats for enormous unknown biodiversity, organized in different levels within the ecosystem. However, little is known about how soil functions as a habitat and how organisms adapt to live in them, especially at a microscopic scale. Organisms living in soils foster a wide span of specific adaptive strategies and specific biological traits (Bardgett, 2010). If we aim to restore damaged ecosystems, it is pivotal to understand these adaptive strategies in a holistic approach, linking chemical, physical, biological, and societal processes.

The soil and the biota make up a complex living system with emerging properties (Ponge, 2005). Emerging properties are critical functions of soil ecosystems that result from the unique metabolic interaction, synergism, and trophic relations of the total biota. If we modify the number and species composition in the soil ecosystem, inevitably, there will be a modification in the metabolic interactions, and consequently, we will observe a change in soil´s emerging functions (Vogel et al. 2019). We can now describe a few soil functions upon which life on Earth depends. One soil function is habitat provision to support biodiversity both above ground and below ground. When we study the biodiversity of a spoon full of soil rich in organic matter, it will contain millions or billions of microorganisms: bacteria, microalgae, archaea, viruses, fungus; it will have thousands of mesofauna (e.g., nematodes, rotifers) and macrofauna (e.g., earthworms). These organisms are all metabolically connected in the soil habitat, forming trophic networks with feedbacks at different scales. The trophic networks are the drivers of matter cycling, including carbon sequestration and release to the atmosphere. It also holds and releases other gases such as oxygen and nitrogenate gases, thus resulting in soil ecosystems as climate regulators. Last but not least, the natural function of the soil is to filter and buffer the water, consequently interacting with the Earth's hydrological system.

The ecosystem services emerge from those soil functions relevant to human societies (Stavi et al. 2016). Traditionally we have classified soil ecosystem services as crop production for fiber and food, pathogens control, water purification, habitat, cultural and recreational sources. Therefore, we assess the soil's capacity to provide these ecosystem services when we evaluate soil quality. However, little attention is given to the fact that soil microorganisms are the main drivers of soil functions.

The reason microbes are considered an essential element providing soil ecosystem function and services lies in their extraordinary diversity, which brings an enormous genetic diversity. Microbes are the first manifestation of life on Earth, and the eons-old heritage allows them to thrive in almost all known environments. Why? Because they have strength in the versatility and diversity of their genetic information. We can visualize the microbial genetic diversity when aligning the genome of two strains of the same bacteria species, such as the case of *Variovorax paradoxus* (Garcia Teijeiro et al. 2020) and *Rhodopseudomonas palustris* (Lo et al. 2018). Both bacteria species are plant growth promoters, but they can exhibit great strains differences in their phenotypic characteristics. Therefore, *V. paradoxus* strains (EPS and S110) show differences in motility, organic matter metabolism, and antibiotic resistance. *R. palustris* (PS3 and YSC3) strains show differences in the synthesis of signaling molecules for interaction with plants.

The complexity of microbial composition in soils challenges the scientific understanding of the connection between microbial (genetic) diversity and ecosystem functions. Several models adopt microbial classification based on their life-history traits (Martiny et al. 2015). This form of classification may aid scientists in understanding the connection of "effects" traits with ecosystems functioning. One typical trait utilized for categorizing microbes is the capacity of a microbial population to transform different nutrients and energy, called the copiotrophic-oligotrophic continuum. Copiotrophic microorganisms thrive on nutrient-rich mediums, but they use the resources with low efficiency. Oligotrophic microbes grow slowly, but they can live in nutrient-deficit niches and use the resources more efficiently. Among other differences, oligotrophic microbes can metabolize a wide range of chemical elements. The variety and availability of nutrients characterize different soil qualities. Thus, in copiotrophic-oligotrophic continuums, the soil offers a possibility of metabolic dynamics that can ultimately affect the nutrient cycles.

The soil matrix and plants as habitats for microbes

The soil matrix contains microniches organized within soil particles and relative to plants' root space (Hartmann et al. 2008, Berg et al. 2014). The soil's physicochemical characteristics, water accessibility, plant types, and distribution will shape microbial communities. Plants' exudation

through the roots accounts for 10 to 40 % of net photosynthesis compounds, consisting of organic acids, amino acids, peptides, nucleotides, flavonoids (Newman, 1985). Thus, the plant exudates serve as a chemo-attractant or chemo-repellent for the surrounding bulk soil microbes. The dynamic area around the root, where root exudates shape the microbial communities, is called the rhizosphere (Berg et al. 2014). The microbial biomass of the rhizosphere is higher than the bulk soil. However, the relative taxonomic composition between both niches varies depending on the plant and the soil conditions. Some studies showed a shift between bulk and rhizosphere bacteria diversity, with more diversity in the bulk community (García-Salamanca et al. 2013). This observation was in corn roots growing in organic-rich soil. The researchers noticed a higher OTUs (Operational taxonomic unic) percentage in bulk soil than in the rhizosphere of the typical oligotrophic *Acidobacteria, Verrucomicrobia, and Actinobacteria.* In addition, they observe in bulk soil bacteria in the copiotrophic-oligotrophic continuum such as *Bacteroidetes, Cyanobacteria, Chloroflexi, Betaproteobacteria, Deltaproteobacteria* and*, Planctomycetes.*
However, the rhizosphere had higher OTUs from the copiotrophic *Alphaproteobacteria, Gammaproteobacteria*, and *Firmicutes* than bulk soil. This result shows a bacterial population's distribution relative to the nutrient quality and an organic-rich bulk soil acting as a reservoir of microbes for plant root colonization.

On the other hand, Chen and colleagues (2021) compared nutrient utilization traits within the microbial communities of the rhizosphere and adjacent bulk soil in arid habitats. In this case, soil water content and heat tolerance also play a role. The researcher's findings reveal a resemblance of the distribution of oligotrophic bacteria in the bulk soil and copiotrophic bacteria in the rhizospheric zone. In arid environments, the microbes dwelling in the bulk soil are under the ecological pressure of high temperatures and dryness. Thus, resistant microbes such as *Firmicutes* and *Actinomyces* are abundant.

In addition, the rhizospheric microbial community can display diversity in other kinds of trophic strategies like saprophytes, commensals, symbionts, and pathogens. Among rhizospheric diversity, a particular group exhibits a metabolic dynamic with the plant, promoting the plant's health and fitness (Fitzpatrick et al. 2019). They are called plant growth-promoting rhizobacteria (PGPR). PGPR plays a role in increasing the nutrient availability and absorption for the plant; they also produce plant hormones for modulation of the plant root-shoot signaling of plant response biotic/abiotic stresses (Garcia Teijeiro et al. 2020). In addition, rhizosphere inhabitants produce chemicals for defense and competition, thus modulating the community composition. Among these chemicals are antibiotics, fungicides, insecticides, and herbicides.

The internal root space can be a microbe niche called the endosphere (Compant et al. 2021). The diversity of the endosphere community depends on that of the rhizosphere and the soil quality. In addition, the endosphere diversity is smaller than the rhizosphere because not all the microbes have the machinery to enter the plant tissue. Examples of the endosphere of corn are *Proteobacteria, Firmicutes,* and *Bacteroidetes* (Correa-Galeote et al. 2018). Interestingly, we can also find the mycosphere, bacterial communities colonizing and interacting with root and fungus hyphae.

We have a reductionist approach to all the ecosystems and particularly from the soil system. Consequently, our activities unleash a state away from natural Earth homeostasis (Bahram et al. 2018). We can no longer refer to soil as dirt or mere physical support for our crops or a substrate to overflow with fertilizers to achieve crop yield. Nevertheless, within the natural soil system, we can find the solution to impact human activities since soil microorganisms can utilize soil microorganisms as biofertilizers, biocontrol agents, and bioremediation to achieve soil quality.

Our agricultural heritage and origin of soil degradation

Looking back at the past, we can find enough evidence supporting soil quality as a fundamental factor in the rise and, in many cases, the fall of ancient civilizations (Herrera and Garcia-Bertrand, 2018). The first human settlements were in geographical regions with high inherent soil productivity, and these soils were used primarily for agriculture: food, fiber production, and shelter. However, a typical pattern in many societies was that the inherent regional soil productivity of the settlements diminished after years of use. Scholars have found evidence of soil degradation by erosion, nutrient depletion, and salinization as a devastating factor among diverse cultures (Plieninger, 2008). Such was the case of the cradle of human civilization, "the Fertile Crescent," a vast region that lies at the mouth of the Euphrates, Tigris, and Nile rivers (Hole, 2006). Here, the rivers deposited their rich sediments and, combined with clouds of dust derived from the Sahara Desert, a thick layer of fertile soil developed over thousands of years (Amit et al. 2020). Once rich and productive, it has become a barren and unproductive land. The growing population had high demands on food production and habitable space (Wuepper et al. 2020). The demand triggered the shift of agricultural activities to marginal land and modification of the rivers by dams and irrigation systems. Thus, the land eventually became saline and desertified.

Land degradation by agricultural practices

In ancient times, the domestication and cultivation of plants happened according to a particular ecosystem or geographical position (Herrera and Garcia-Bertrand, 2018). However, once soil quality started to fail, some societies shifted their unsustainable agricultural demands to other lands. Then agricultural practices were adapted to the new local environmental conditions. Nevertheless, it is interesting to note that we are still dependent on a small number of domesticated plant species and animals for food. Equally, we are still heavily reliant on crop yields´ soil/water systems. Consequently, farmers need to actively change the natural ecosystem where they are living (Edwards et al. 2019). These operational changes on Earth began, among others, via deforestation, soil allocation, irrigation systems, and plowing.

The global agricultural system now leaves a footprint on the topsoil where the critical soil functions occur via microbial diversity. Today's major land degradation processes are desertification, soil erosion, loss of soil organic matter, and loss of plant coverage (Prăvălie et al. 2021). The soil microbes tend to occupy different niches within the soil particles, determining microorganisms' dispersal during wind erosion events (Gardner et al. 2012). *α-,β-Proteobacteria,* and *Acidobacteria* are found in coarse soil particles while copiotrophic *Firmicutes* and *Bacteroidetes* occupy the fine soil particles. The oligotrophic *α-,β-Proteobacteria,* and *Acidobacteria* have an active implication on the C and N cycles, implying an impact on the parent soil function when losing these groups. *Firmicutes* and *Bacteroidetes* are related to resistance to harsh conditions, such as drought or soil salinity. The distribution in different size particles will determine the extent of relocation that will depend on the event's strength.

A significant example of a soil erosion event was the "Dust Bowl" in the 1930s in vast areas of North America (Lee and Gill, 2015). The combination of changes from natural drought-resistant plant communities to vast monoculture crops and drought events lead to crop losses, thus leaving soils unprotected. The bare soil generates high temperatures in the topsoil originating soil dust aerosol and increases air circulation. As a result, topsoil was carried out in vast areas generating anthropogenic land degradation.

Often fertile topsoil losses are accelerated by human agricultural activities, low vegetation coverage, tillage, soil plowing. The topsoil possesses plant nutrients such as organic matter (carbon), nitrogen, phosphates, and microorganisms dwelling on particles in the soil system. When they are lost, the allocation of these elements in the water bodies will cause pollution, developing water quality degradation, air pollution, and a general disturbance of the ecosystem homeostasis (Wuepper et al. 2020) (*Figure 1*).

Figure 1. Agricultural intensive land showing topsoil erosion. The white arrows indicate topsoil allocation due to the slope and inadequate plant coverage. Picture made by Sofia Kommer Garcia.

Soil organic matter and soil erosion

The giant carbon pool in terrestrial ecosystems is the soil, and half of all the carbon is stored in the topsoil (Heimann and Reichstein, 2008). The carbon component strongly influences all three aspects of soil quality: biological, physical/structural, and chemical. The soil organic matter (SOM) is a nutrient source and a structural component implicated in the formation of soil pores. The carbon in the soil appears as inorganic or organic. The origin of organic carbon is cellular decomposition producing DNA, proteins, lipids, and carbohydrates.

In contrast, inorganic carbon originates from cellular activities such as osmoregulation products like calcium carbonate or respiration byproducts: CO_2, H_2CO_3, HCO^{3-}, CO^{2-} (Fierer et al. 2012). Microbial regulation is critical for both producing and degrading soil organic matter.

The microbial carbon use efficiency is the proportion between incorporation (organic matter) and respiration of carbon (CO_2) (Page et al. 2020). Soil erosion accelerates the increment of CO_2 in the atmosphere via microbial degradation of organic matter and inhibition of CO_2 sequestration. Soil carbon components are particular and mineral-associated organic matter (**POM**; **MAOM**) (Nunes et al. 2012). Both have different origins and vulnerabilities to climate change and land degradation. POM indicates plant origin, is recalcitrant, and has a lower turnover than MAOM, holding communities with higher proportions of oligotrophic microbes (Trivedi et al. 2018). POM has fast removal in events of soil erosion. Relatively more removal of POM will increase the release of CO_2 to the atmosphere. MAOM is a regular indicator of microbial activities and debris, and it is persistent in the soil since it binds to mineral particles and forms smaller aggregates than POM. It also has a more relative content of nitrogen; hence the relation C to N is low. MAOM can hold more copiotrophic microbes.

CUE determines the fate of MAOM in soils; when soil pH is low, some bacterial activity is inhibited, impacting the organic matter accumulation. When soil acidity is lower than 6.2, the net microbial growth efficiency is low (Malik et al. 2018). Equally, when soil texture is dominant in silt and clay, the net microbial efficiency is low. The microbial communities and the mineral types of the soils are changing across the different areas; this will influence the soil organic dynamics. In a climate change scenario where the temperatures and the water content are disturbed, soil warming likely influences microbial soil organic matter turnover (Kallenbach et al. 2016, García-Palacios et al. 2021). Implementing soil conserving strategies through the incrementing soil organic matter needs the consideration of the type of soil and ecosystems characteristics, such as the relative content of MAOM and POM in the SOM (Cotrufo et al. 2019). Considering the type of soil and ecosystems characteristics is especially important when tackling soil carbon sequestration (*Figure 2*). In addition, essential plant nutrients such as nitrogen and phosphorus are in equilibrium with SOM. Therefore, soil degradation triggers not only the loss of SOM but also other nutrients related to it.

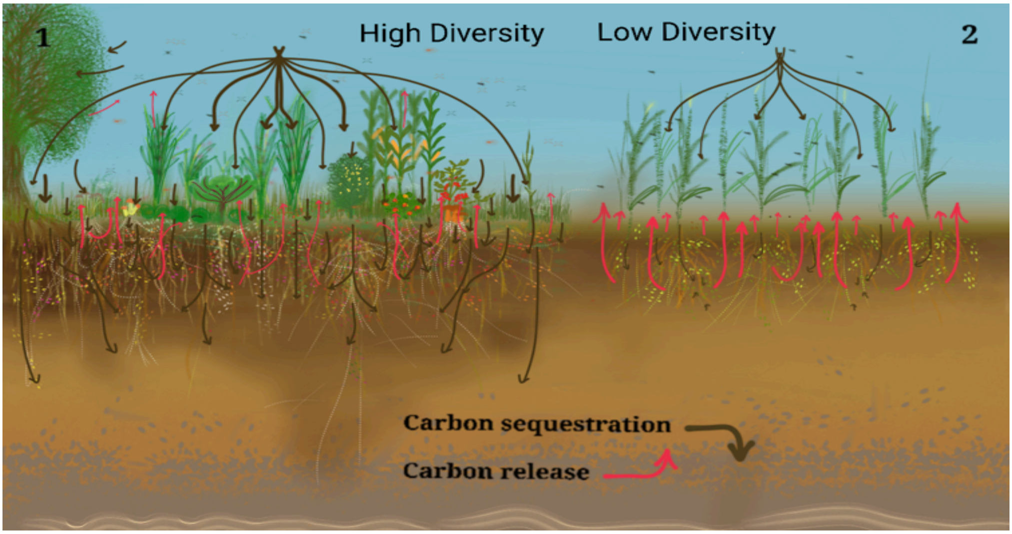

Figure 2. Contrasting Carbon cycle fluxes in two different ecosystems. 1 High Diversity indicates an agricultural set production mimicking natural ecosystem, while 2 Low Diversity indicates intensive agriculture production. The brown arrows show the carbon sequestration, and red lines indicate carbon release in the form of gases (i.e., CO_2, CH_4).

Soil organic matter and plant nutrients

Earth history is linked to nutrient cycling, and this cycling helps maintain the nutrient homeostasis required for primary productivity. The nutrient accessibility in the soil is connected to the SOM pool supplied through plant-animal-soil-atmosphere interactions (Alvarez and Alvarez, 2000). There are 17 essential basic elements required for plant growth. The lack of any one of these essential nutrients can result in a severe limitation of crop yield. Complex nutrient cycles (biogeochemical cycles) incorporate various physical-chemical and, most importantly, biological processes. Agricultural activities modify biogeochemical cycles in the soil, and the impact will depend on the agricultural practice. Monocultures and extensive agriculture degrade microbial communities that provide nutrients from the soil to the plant (Yang et al. 2020). Shifts of microbial communities determine changes in soil chemistry and mechanical properties. Microbes' populations in the soil environment are dynamic and drivers of the mineral cycle feedbacks, including carbon, nitrogen, and phosphorus.

Hence, inadequate and intensive managed agricultural activities will affect soil properties to the extent of causing soil fatigue (Wolińska et al. 2018). In this situation, the farmer perceives that the crops can no longer incorporate nutrients due to a loss of soil fertility. Therefore, to maintain soil fertility for sufficient crop yields, farmers apply more soil amendments to enhance soil fertility, including inorganic chemical fertilizers and organic sources of a nutrient, such as manure or compost, often resulting in surplus quantities of primary macronutrients (Smith and Siciliano, 2015). However, the amount of fertilizer application is not always optimized for the levels used by the crop, and excess nutrients, especially nitrogen and phosphorus, can be transported via surface runoff or leaking through agricultural fields and pollute surface and groundwater.

The amount of fertilizer applied on the fields is considering only to cover the needs of the crops. However, the critical elements for the plant nutrient availability are the soil microbiological communities in charge of the nutrient cycles (McDaniel et al. 2014, Sun et al. 2020). The soil microbes maintain ecosystem functions, and if we could master the nutrient biogeochemistry (processes catalyzed by microbes in the soil matrix), we could avoid the commercial hunt for these valuable elements. Many social conflicts derive from the capitalistic exploitation of nature and labor to control limited natural resources (Verde and Matusso, 2014). For example, under the control of Morocco and Spain, the disputes of the Sub-Saharan phosphates' mines forced the Subsaharian people to live in refugee camps. These people suffer displacement and are obligated to live outside their land. Apart from this conflict, phosphorous resources will only last a few more years. Therefore, we need to learn to manage this nutrient wisely. The overuse of fertilizers is unnecessary, and the excess will runoff or leak to body water, leading to eutrophication of both soils and waters (causing the excessive growth of some microorganisms and the extinction of freshwater species). Exploration of efficient processes for preventing runoff or removing excess nutrients from these sources is urgently needed.

Research is also needed to improve the nutrient use efficiency and biogeochemical cycles (especially N and P), which are now relatively poor. For example, N fertilizers in agriculture contribute 70+% of all anthropogenic emissions of nitrous oxide, the third most important greenhouse gas. Additional topics needing urgent attention include the influence of land use and cropping practices on N_2O emissions and the design of engineered rhizosphere microorganisms that disfavor N_2O and funnel N-cycle intermediates toward nutrients.

Agricultural practices for soil recovery

Due to climate change and an exponential increase in the human population, food insecurity is a critical issue (Abbas et al. 2020). Following population increment with better income and without ecological awareness, an increase in animal protein consumption becomes the new scenario (Edwards et al. 2019). Conventional agriculture is characterized by monoculture crops, with slight rotation, fertilizers, and other agrochemicals applications and tillage. These intensive practices reduce the soil richness and evenness of the microbial communities. Monocultures always have the same selection pressure within the bulk microbial populations to form the rhizosphere (Orwin et al. 2021). In this scenario, and with synthetic fertilizers, there will be another selective pressure within the rhizosphere for those microbes that thrive in high concentrations of nutrients. The soil ecosystem services are mesmerized because of the uneven and low diverse rhizosphere. The formation of a diversity of SOM will be affected, also shifting other nutrient cycles. Chances will be that an invasive pathogen can find its way to the plant since the low biodiversity cannot control pathogens.

On the other hand, tillage impacts the soil microorganism by destroying the soil structure, soil exposure to changes in temperature, gases, and reduction of water content. Furthermore, it is predicted to increase SOC losses due to anthropogenic climate warming, making tillage effects more severe. Additionally, tillage reduces the soil pH and changes the SOM and nitrogen cycles (Behnke et al. 2020). Therefore, feeding the human population will require revolutionary ideas that imply taming conventional agriculture to mimic natural ecosystems functions as much as possible.

One of the options is to use microbial biomass to produce products rich in proteins and supplementary foods. This form of production is called “single-cell protein.” In this model, air and water use solar energy to produce microbial proteins through photovoltaic energy for human consumption. Production efficiency can be ten times higher than any crop (Leger et al. 2021). In addition, this model can be introduced in urban settings, saving land for ecosystem recovery.

Another opportunity for cleaner agriculture is to create more resilient and biodiverse systems. If we consider the trophic system within the rhizosphere, the more diverse, the greater will adapt to changes, and a diverse rhizosphere comes with plant diversity. Ecosystem restoration from intensive agriculture can be achieved by reintroducing native plants and additional strategic crop management. In restoration experiments with plant management, the soils gradually return to a native state. In the example, prairie soils were recovered, and two microbes’ genus were naturally reintroduced during the recovery process: *Verrucomicrobia* and *Acidobacteria* (Barber et al. 2017). Likewise, the plant diversity of an agroecosystem will increase adaptability in the face of disturbances. The ability of agriculture to adapt to climatic changes was not a critical point in the middle of the XX century, but since 2020, it began to have importance. Instead of considering sustainability as dependent on a set of variables, such as soil type or climate change, it is now considered holistic, embedded in a complex system.

Sustainable agriculture seeks to use natural resources strategically, minimizing the impacts in existing ecosystems (Brodt et al. 2011). One way to minimize these impacts is by converting intensive agriculture into soil conservation or focusing on soil conservation. Conservatives’ practices maximize incorporating the crucial soil functions into the agroecosystems (Kremen, 2005). Natural plant residues were eliminated from the soils in conventional agriculture, while these residues are left at the maximum in conservative agriculture. In this way, soil microbes will recreate the natural incorporation of plant residues into the cycle of the nutrients. Conservation of soils means to return the productivity of those soils degraded by intensive agriculture, then essential functions of the original natural ecosystems are incorporated again.

Maintaining the functions of the soil is necessary to keep or increase the soil's organic matter (Page et al. 2020) (*Figure 2*). SOM is an essential source of macro and micronutrients for the microbes or other soil inhabitants. In addition, plants with their root exudates maintain microbiota activity. The activity of this microbiota will provide a buffer for fluctuations in the soil that may occur due to disturbances. This activity can collaborate with the decrease of the impact of climate change. In soil systems where plant richness is increased, the MOAM turnover time decreased, thus pointing to an increment of microbial activity and diversity (Prommer et al. 2020) (*Figure 3*).

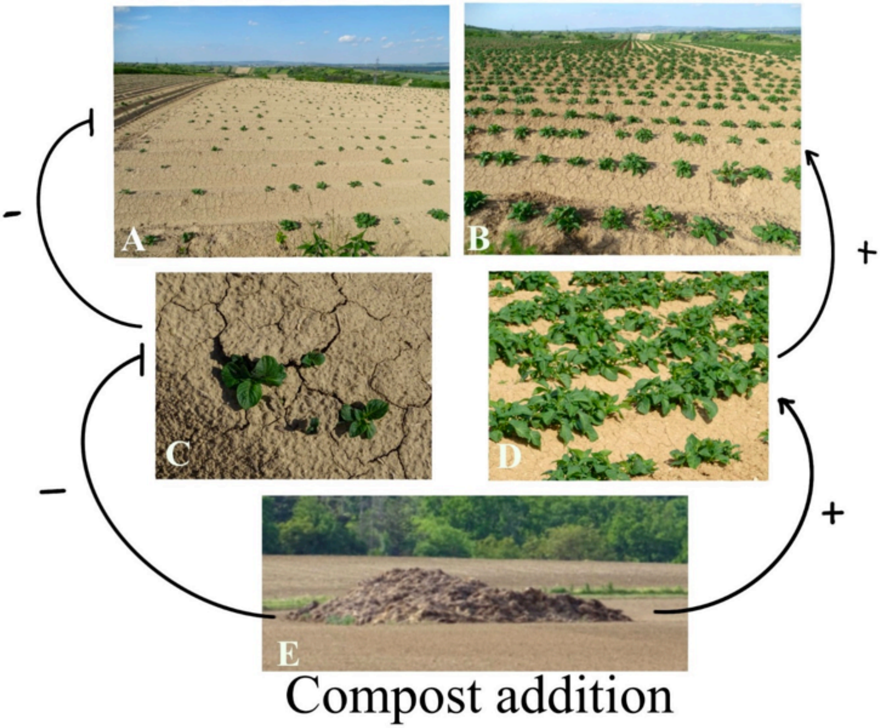

Figure 3. Effect of compost addition on potato fields. A (field setting) and C (plant view) show fields without the addition of compost, B (field setting) and D (plant view) show adjacent fields with compost addition. Picture made by Sofia Kommer Garcia.

Past cultures have taught us what happened when overpopulation demands on food put pressure on the soil systems. However, we can find good soil management practices examples, such as the "Terra Preta do Indio," a soil of human origin (biochar). This soil is dark and rich, located in different small areas within the unproductive oxisoils from the Amazonian Forest (Briones, 2012). These soils can regenerate their biodiversity after biodiversity changes by agricultural uses. This attribute is thought to be due to the structure acting as a niche reservoir for soil microbes. Biochar also enhances the nutrient cycles, and it can regulate the pH. Nevertheless, "Terra Preta do Indio" is not applicable for every region of the world; thus, it will depend on the particular characteristics of the site.

Another example of alternative agricultural practices is the "Milpas" used by the Aztecs, which consist of cultures (still used today) of mixed crops, typically corn, pumpkin, and beans (Moreno-Espíndola et al. 2018) (*Figure 4*). In this setting, the mixed crop benefits from the intertwined root microbes from each plant. There will be nitrogen fixation, microbes solubilizing phosphates, and other microbes delivering water to the crops. Thus, Milpas is an excellent example of an enhancement in soil ecology properties.

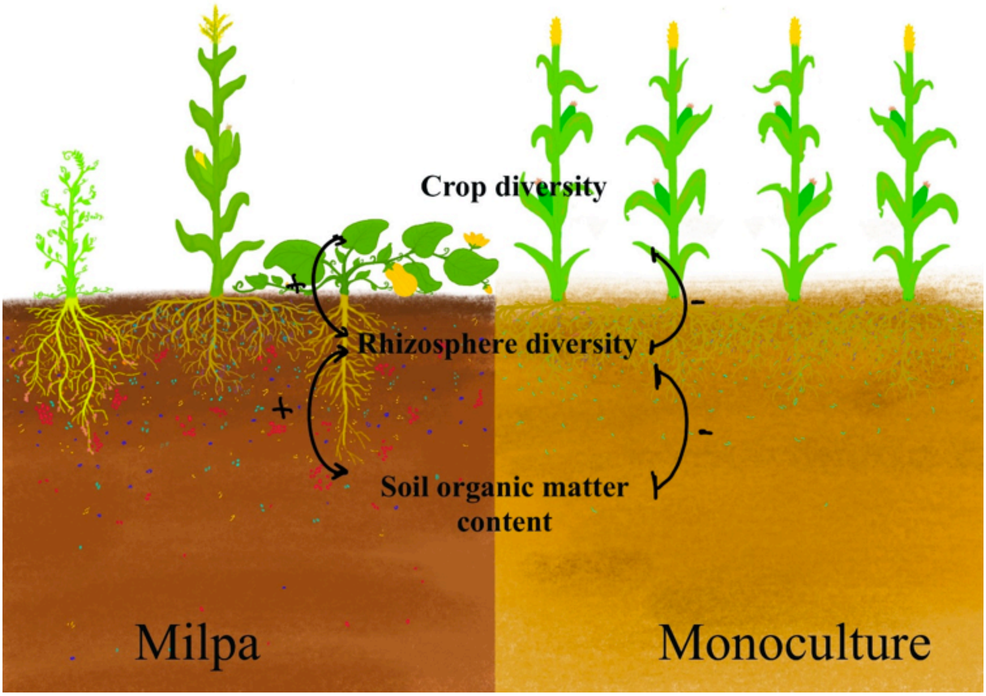

Figure 4. Milpa combines three crops enhancing rhizosphere diversity and soil organic matter in contrast with Monoculture (artistic drawing made by Sofia Kommer Garcia).

Implications of soil microbial communities in soil recovery

Soil microbiota contribute to nutrient cycling, SOM formation, decomposition, and interaction inter and intra-kingdom. Furthermore, the interactions between the soil biota are dynamic in space and time, bringing another level of complexity (Wassermann et al. 2019). It is then necessary to explore the diversity of natural soil and utilize it to explore all possible strategies for manipulating the soil microbiota under the pressure of agricultural disturbances (*Figure 2*). Microorganisms are providers of different metabolic substances that can benefit both the growth of plants and animals. For example, in agriculture, these compounds can be used for the plant's growth to protect against abiotic and biotic stress (Shade et al. 2012).

It is well known that only a tiny percentage of the total microbial biomass can be isolated in the laboratory (Garcia-Teijeiro et al. 2009). Recent advancements of next-generation sequencing technologies (NGS) allow us to use NGS as an instrument for practical applications in soil microbiology. NGS can be used for comparative genomics and phylogenetic studies from a community. Genomic sequencing applications are helpful when we need a complete genome sequence belonging to one isolated microbe either for new species characterization or for complex genomic regions studies (Bernreiter et al. 2016). Protocols for long-read sequencing from 10 to 50 kb are well developed using Illumina, Ion Torrent, PacBio, and Nanopore systems (Loman and Pallen, 2015). The selection of the technology will depend on the sequence coverage requirements versus costs per sample.

Metagenomics is the sequencing of all the genomes belonging to an environmental sample. We obtain the information of individual genomes within a community and inspect the genetic diversity that can contribute to the soil functions. The protocol used for metagenomics is random shotgun sequencing for complete genomes within the community (Deurenberg et al. 2017, Sekse et al. 2017, Slatko et al. 2018). Sometimes metagenomic is confused with metabarcoding, that is, PCR amplification of specific genes and further sequencing of the amplicons. Particularly, metabarcoding applies to phylogenetic studies within a community. For example, 16sRNA gene is applied for Prokaryotes, and 18sRNA and internal transcribed spacer gene (ITS) for Eukaryotes. NGS generates massive sequencing information that is further processed with bioinformatics pipelines.

With next-generation sequencing, it is possible to know the composition of the soil microbiota when the soil is degraded and compare it with the composition of the soil microbes after implementing the restoration. Studies on the impact of agriculture in grasslands were conducted applying next-generation sequencing technology. The researchers used patches of untouched graveyard soil placed in former grasslands and contrasted the soil microbial composition with adjacent grassland soil under intensive agricultural pressure (Fierer et al. 2013). They found that one bacterium genus was lost in the agricultural manipulated grasslands, the *Verrucomicrobia* (Kielak et al. 2010, Bergmann et al. 2011). *Verrucomicrobia* is challenging to isolate; thus, little is known about its ecological attributes. This bacterium allowed the native plants to grow in grasslands under specific stress conditions of climate and water availability. Prairie ecosystem restoration by reintroducing native plants brings a concomitant introduction of *Acidobacteria* and *Verrucomicrobia* (Barber et al. 2017). Theoretically, with these classic oligotrophic bacteria genus reintroducing, we will also shift the carbon dynamics towards soil sequestration, as explained above.

Using next generation sequencing to monitor changes in microbial activity and community structure helps identify good agricultural techniques for soil recovery. Especially interesting is to monitor long-term activities such as application of plant debris, alone or in combination with plant growth-promoting microbes, compost applications, biochar, and soil microbiome recruitment techniques with different compost. Different studies show that composts and PGPR can effectively recover soil fertility (Averill et al. 2021). In addition, omic' technologies application in soil microbial research generates an enormous amount of data, which can be used to predict microbial communities with environmental variables.

Other strategies are the field inoculation of characterized PGPR microbes alone or in combination as an artificial community (Garcia Teijeiro et al. 2020) (*Figure 5*). These methods are targeting advantageous modification of the rhizosphere microbiota. However, for the application of this amendment, there should be an effort of extensive site-specific characterization that matches the appropriate inoculum in the soil site. Important field characteristics to monitor are soil temperature, pH, water content, field history, and the target crop. This selection is needed because many PGPRs are sensitive to temperature, pH, osmotic stress and have a specific pattern of colonization that determines plant species. In field conditions, a significant number of variables may cause inconsistencies, impediments, and failures associated with soil microbiota disruption (Gu et al. 2020). Therefore, comprehensive knowledge of the microbiome changes is required under the light of microbiome-supported approaches

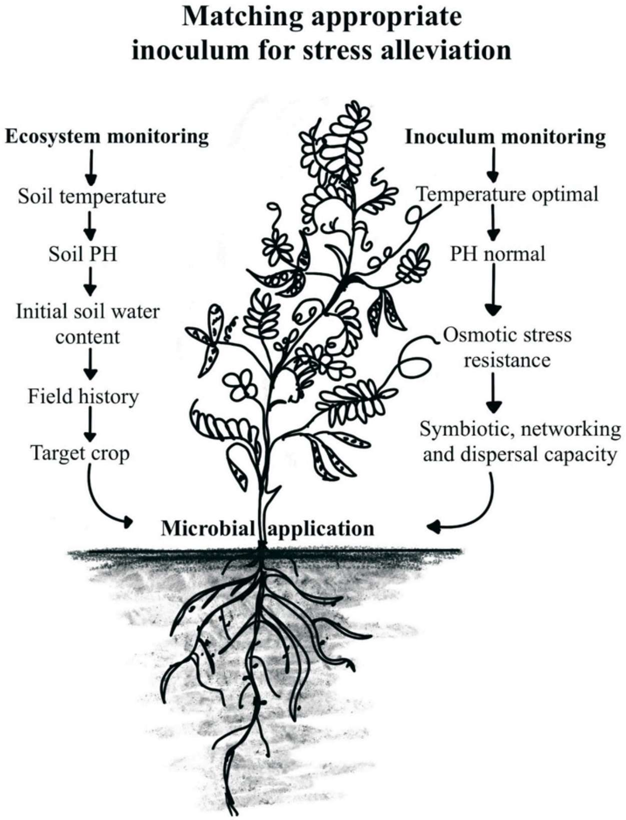

Figure 5. Roadmap for the application of plant growth-promoting rhizobacteria. Figure modified from Garcia Teijeiro et al. 2020.

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Chapter 4

Fungal biodiversity and forest soil health ecosystems

Andreas Bernreiter and Rosalia Garcia Teijeiro

Introduction

Since humans are utilizing land for farming, the global forest area has reduced significantly from the past. It is estimated that currently, around one-quarter of terrestrial land and herein one third (38%) of habitable land area is occupied by forests (Williams, 2003). Other significant habitable land areas are covered by glaciers (10%) and barren land (19%; deserts, dry salt flats, beaches, sand dunes, and exposed rocks). Today farming is making use of half of the habitable land, and mainly, the area of livestock farming is equal in area to the world's forest area. In Europe, 40% of the territory is covered by forests, and these have multitudes of benefits for the human population. Forests positively influence climate regulation, water supply, timber production, energy, erosion control, and more. Though forests are also rich in biodiversity, they still face problems due to anthropogenic-driven stress. The change in climate affects tree species composition and leads to a reduction in biodiversity. This reduction in biodiversity indirectly affects the reduced resilience against infectious diseases (Goberville et al. 2016).

Forest ecosystem functioning and plant growth and health are driven by interaction of microorganisms such as bacteria, archaea, and fungi in the rhizosphere with the plant host. Fungi in particular form symbiosis with plants, but also perform other essential roles, such as decomposition of organic material. To date, there are many studies indicating and pointing out that the diversity of soil organisms is of particular importance to withstand the disturbances caused by global climate change (Tedersoo et al. 2020, Langenheder et al. 2012, Bardgett and van der Putten, 2014). Microbial diversity helps to maintain ecosystem services and provides resilience to disturbances, particularly to the stress caused by global climate change. Not surprisingly, several studies revealed that soil properties primarily determine the fungal community composition. Particularly soil pH, but in general, soil chemistry is the primary driver of the species composition in soil fungal communities (Glassman et al. 2017, Fierer et al. 2009, Tedersoo et al. 2020).

People usually define forests as areas dominated by trees (see *Table 1*). However, today we do not see plants as single units but as an organism existing in complex systems with microbial networks, fungi, bacteria, archaea, viruses, protists, and algae, as well as with nematodes, arthropods, and protozoa (Ingham et al. 1985). Trees directly interact with an uncountable number of microbes on their roots, stems, and leaves. The microbial community is essential for biogeochemical processes making essential elements, particularly phosphorus and nitrogen, available for plant growth and survival. Ecologists assume that a high biodiversity community has greater functional redundancy than a community dominated by specialized species (Isobe et al. 2019). Functional redundancy means that various microbial taxa present have the same function in the biochemical cycling of nutrients. If one taxon is lost in a changing environment, others can take over the function, which gives the ecosystem resilience and stability

FOREST	Land spanning more than 0.5 hectares with trees higher than 5 meters and a canopy cover of more than 10 percent, or trees able to reach these thresholds in situ. It does not include land that is predominantly under agricultural or urban land use.
FOREST EXPANSION	Expansion of forest on land that, until then, was under a different land use, implies a transformation of land use from non-forest to forest.
AFFORESTATION	(Sub-category of FOREST EXPANSION) Establishment of forest through planting and/or deliberate seeding on land that, until then, was under a different land use, implies a transformation of land use form non-forest to forest.
NATURAL EXPANSION OF FOREST	(Sub-category of FOREST EXPANSION) Expansion of forest through natural succession on land that, until then, was under a different land use, implies a transformation of land use form non-forest to forest (e.g., forest succession on land previously used for agriculture).
DEFORESTATION	The conversion of forest to other land uses (regardless of whether it is human induced).

Table 1. Forest and Forest Management Definitions from FAO (2020).

Fungal biodiversity

The recent advances in technologies such as NGS to study microbial communities without culturing methods (Bernreiter, 2017) revealed that the microbial structure significantly changes by season and by soil properties (Yuste et al. 2011). Furthermore, studies show that fungal communities can adapt better to changing environments (moisture, temperature, plant activity) than bacteria (Yuste et a. 2011). Therefore, it is hypothesized that fungal-dominated soil communities might be more resilient to climate change (higher temperature; drier conditions) than bacterial-dominated communities. Soil fungi are usually classified into three functional groups based on their strategies to gain energy:

Saprophytic fungi

Decomposers or saprophytic fungi are essential for immobilizing and cycling nutrients in the soil. They break down complex organic compounds such as proteins, carbohydrates and fats, eventually mineralising the most basic elements. They play essential roles in the global carbon cycle by decomposing the plants carbon to generate fungal biomass and release extracellular substances. Part of the carbon is also released as carbon dioxide through cellular respiration to gain energy. Many fungi are specialized in decomposing cellulose and lignin in wood.

Mycorrhizal fungi

Mycorrhizal fungi colonize plant roots and make symbiosis with plants. Fungi solubilize soil nutrients and bring these nutrients (phosphorus, nitrogen, micronutrients) and water to the plant in exchange for carbon from the plant. There are two significant groups of mycorrhizae, the ectomycorrhizae, and the endomycorrhizae (see *Table 2*). Based on recent phylogeny and morphological data, researchers sometimes also classify Mycorrhiza in four principal types: arbuscular Mycorrhiza (AM), ectomycorrhiza (ECM), ericoid Mycorrhiza (ERM), and orchid Mycorrhiza (OM) (Brundrett and Tedersoo, 2018; Smith and Read, 2008; Tedersoo and Bahram, 2019).

Pathogenic fungi

Take their organic requirements from plants, and in contrary to symbiotic fungi, the plant does not benefit but is harmed, and this can cause reduced production or even death of the plant. Numerous pathogenic fungi cause huge economic losses in forestry and wood industry. Phytophthora ramorum (Oomycota) is responsible for the sudden oak death, Hymenoscyphus fraxineus (Ascomycota) is killing ash trees (*Fraxinus excelsior*) and several species of Mycosphaerella (Ascomycota) are associated with leaf spot diseases on *Eucalyptus* trees. *Eucalyptus* is grown in plantations worldwide and provide an important source of hardwood for commercial use.

Figure 1. In temperate forests there is a high diversity of ectomycorrhizal, saprotrophic and pathogenic fungi. Here is a selection of fruiting bodies found in the district of Lower Austria in 2020 and 2021 in a forest dominated by oak and birch (*Betulo pendulae-Quercetum roboris*). **A**. *Lycoperdon perlatum* (Saprophyte) **B**. *Morchella esculenta* (Mycorrhiza) **C**. *Amanita muscaria* (Mycorrhiza) **D**. *Macrolepiota procera* (Saprophyte).

Mycorrhiza

Bernhard Frank recognized that fungi form a symbiosis with tree roots in the search for ways to produce truffle. He suspected that fungi were providing the trees with nutrients. Frank invented the word “Mycorrhiza”, which is derived from the two greek words “mykes” for fungi and “rhiza” for root. He published his work about the symbioses between fungi and plants with the title “Über die auf Wurzelsymbiose beruhende Ernährung gewisser Bäume durch unterirdische Pilze” (Frank, 1885). Frank speculated that he found an interaction that is beneficial for both the fungus and the tree. Today, Mycorrhiza is a topic for many research groups and companies, and there are journals focusing only on mycorrhizal studies.

Several book chapters and reviews discuss the latest findings on mycorrhizal research and their implications in soil health (Karliński, 2021; Gupta, 2020; Smith and Read, 2008).
Scientists distinguish between two types of Mycorrhizae, namely arbuscular Mycorrhiza and Ectomycorrhiza (*Table 2*), and both types, though acting different, give the plant limiting macronutrients in exchange for plant photosynthate.

Forest trees regularly form a symbiosis with ECM fungi to have better access to mineral nutrients and to water, hence resisting more stressful situations.

Researchers believe that around 8% of plants have no mycorrhizal partner (Brundrett and Tedersoo, 2018), and these plants are usually found in extremely poor soils, such as infertile habitats in Western Australia and South Africa (Lambers et al. 2010).

	Ectomycorrhizae	**Endomycorrhizae**
Hyphae	The hyphae form a sheath around the outside of the plant root and enhance the nutrient exchange between the plant cells and the fungi	The hyphae of the fungi enter the plant cells at the root for nutrient exchange
Frequency	Symbiosis with around 5% of plants, dominant with forest trees	more than 80% of terrestrial plants affected
Contact	Extracellular colonization	Intracellular colonization
Phyla	Basidiomycota, Ascomycota, Zygomycota	Glomeromycota
Abundance	~2% of plants species (mainly woody plants)	~80% of plant species
Characteristic	Forms Hartig net between the cells in the root cortex	Forms arbuscules, which are hyphae structures inside the root cortex cells
Types	Arbutoid	Arbuscular, Ericoid, Orchid

Table 2. Characteristics of the two main forms of Mycorrhiza.

Arbuscular Mycorrhiza (AM)

The first land plants appeared without roots and instead endophytic associations with fungi (Strullu-Derrien et al. 2018). These first bryophyte-like land plants evolved from aquatic algae around 400 million years ago, and its associated arbuscular mycorrhizal fungi (AMF) are believed to coexist since this time of early Devonian (Smith and Read, 2008). The mycorrhizal fungi provided inorganic nutrients and water to the first land plants and received carbohydrates as a benefit. This hypothesis is backed by palaeobotanical, morphological data and DNA based phylogenetic studies, respectively, and led the way to the development of plant roots (Feijen et al. 2018, Miyauchi et al. 2020, Strullu-Derrien et al. 2018). This transition came along with increased atmospheric oxygen levels and carbon fixation.

Ectomycorrhizal fungi (ECMF)

ECMF have evolved independently from various genera and different fungi phyla. It is estimated that ECMF are forming symbiosis with approximately 2% of vascular plants, preferably trees, and more than 250 genera of mainly ascomycetes and basidiomycetes are to date known to function as Mycorrhiza (Tedersoo and Smith 2013).

These symbiotic relationships provide the host trees with improved resistance to biotic and abiotic stresses. Thus, most forest trees are highly dependent on their fungal partners and could possibly not exist without them in areas of poor soil quality. Many studies on pine and other temperate forest trees show the increased survival rates when ECMF are present (Smith and Read, 1997).

This partnership is dominant in temperate and boreal forest ecosystems, which are characterized by the low availability of plant nutrients (Smith and Read, 2008). In forest ecosystems, where nutrients are the main limitation, ECMF increase plant productivity.

Dual-mycorrhizal plants

While most vascular plant species form a mycorrhizal symbiosis of only one type, AM-colonized plants may sometimes co-occur with ECM and ERM fungi (Smith and Read, 2008; Brundrett and Tedersoo, 2018), because the dual symbiotic habit provides complementary benefits in poor soil and harsh environments. Dual-mycorrhizal plants seem to be more frequent than speculated as discussed in a recent publication (Teste et al. 2019). The identification is based on observing arbuscules or coils for the presence of AM and Hartig net or similar structures for ECMF status within the same plant species. It is expected that the presence of dual mycorrhizal plants gives benefit to the plant, particularly under stress conditions, such as abiotic and biotic stress or soil moisture and nutrient deficiency. However, to date, more experimental data is needed to understand the relevance of dual mycorrhizal plants (Teste et al. 2019).

Studies on eucalyptus suggest that AM are always dominant in seedlings and young trees, and they are in older trees later replaced by ectomycorrhizae (Adjoud et al. 1996). Nevertheless, a recent publication (Adjoud-Sadadou and Halli-Hargas, 2017) suggests that the dual mycorrhizal status of trees can be more complex. Experiments revealed the arbuscular mycorrhizae persist in soils and show over ectomycorrhizae during the dry season, while the ECMF are more abundant in the wet season.

Biogeochemical cycling

The liveliness of most forests is determined by the availability of one or more nutrients. Nitrogen (N) and Phosphorus (P) are the most needed growth-limiting elements in temperate forest ecosystems (Masclaux-Daubresse et al. 2010, Vitousek et al. 2010), but other essential elements Ca^{2+}, Mg^{2+}, and K^+, and even micronutrients such as B, Mn, Zn, or Si, can be limiting as well (Adriano, 2001).

The term biogeochemical cycling is explained simply as a movement of elements through various forms, such as organic and inorganic, and their return to their original state. In textbooks, we can find that separate biogeochemical cycles exist for each chemical element, such as the nitrogen, phosphorus, and carbon cycles. However, recent research revealed that we should not see nitrogen, phosphorus, and carbon cycling as separate cycles but cycles dependent on each other. We understand now that a balance in N and P availability controls plant growth and soil processes (Güsewell, 2004). If one element is limited, it will affect the productivity of the forest cycling/decomposition of other nutrients, influence the structure and functions of microbial communities, and negatively affect carbon sequestration.

Nitrogen is needed to metabolize amino acids, proteins, coenzymes, ATP, nucleic acids, chlorophyll, and other secondary metabolites. Phosphorus is essential for generating macromolecules such as nucleic acids, phospholipids, and other phosphate-rich compounds.

Mycorrhizal fungi improve the plants' access to the two most needed mineral nutrients Phosphorus and Nitrogen, by effective absorption and translocation. More data is available to date studying the interactions between crop plants and symbiotic Mycorrhiza in terms of nutrient supply and stress resistance, meaning that we know more about AMF than ECMF, which are dominating forest trees.

Several studies in recent years have revealed clearly that AMF play a significant role in the P uptake of plants (Smith et al. 2011, Smith and Smith, 2012). In exchange, plants provide all organic carbon to the fungi. Hence the symbioses are mutualistic. Besides P, AMF provide other nutrients, such as nitrogen in exchange for plant carbon (Smith and Read, 2008; Smith et al. 2011, Tedersoo and Bahram 2019). AMF have been shown to enhance plants' tolerance to salinity by improved ion homeostasis (Estrada et al. 2013). Experiments showed that plants inoculated with various AMF showed a significant increase of K^+ ion and reduced Na^+ ion accumulation as compared to non-mycorrhizal plants, hence higher K^+/Na^+ ratios were measured in the plant tissues. Na^+ is causing toxicity because it competes with K^+ for binding sites at the plasma membrane (Kronzucker et al. 2013). Other studies show that AMF improves the plants' drought tolerance by increasing potassium availability from soil and uptake and translocation to the plant shoot (Zhang et al. 2017). Experiments also showed that drought stress increases the colonization rate of AMF to the plant root.

To maintain ecosystem stability in soils can avoid the excessive usage of nitrogen (N) fertilizer (Verzeaux et al. 2017). Agricultural practices mostly perform soil perturbances, which disrupt plant/mycorrhiza symbioses, and agricultural soils are therefore largely exhausted mainly because of bad agricultural practices. Human actions lead to disruption of soil ecosystems, limiting the establishment of AM/plant symbioses (Verzeaux et al. 2017). Agricultural practices should change how soil favours AMF / crop interaction, thus improving nitrogen use efficiency and reducing nitrogen fertilizer usage (Verzeaux et al. 2017). Knowledge from agricultural practices could also be applied to forest sites that suffer from climate change effects, such as temperature increase or drought.

Ectomycorrhizal fungi significantly impact on ecosystem functioning and biogeochemical cycling in temperate forests (Soudzilovskaia et al. 2019). Soudzilovskaia and colleagues (2019) explain that temperate forests store much higher soil carbon than tropical and subtropical forests, where carbon is mainly stored in the aboveground plant biomass. ECMF alters organic matter by releasing extracellular enzymes, such as glycoside hydrolases, class II fungal peroxidases, glyoxal oxidases, and phenol oxidases, which take part in organic degradation (Frey, 2019). For the uptake and utilization of organic nitrogen, Ectomycorrhizal fungi release exocellular hydrolytic enzymes (Chalot and Brun, 1998). Assimilated amino acids are then forwarded to plants in exchange for carbon from photosynthesis. Experiments with amino acids containing fluorescent nanoparticles proved that plants take up amino acids in the presence of Mycorrhiza much more efficient than plants without Mycorrhiza (Whiteside et al. 2012).

The availability of nitrogen limits plant production in forests and affects the microbial composition (Högberg et al. 2017). In forests with little nitrogen, microorganisms retain much of the available soil nitrogen for themselves, and ECMF transfer little to ECM trees. Only plants with less N demand can survive, such as trees in symbiosis with ECMF. If N supply increases, the soil microbial community composition changes from fungal dominance to bacterial dominance.

Common mycorrhizal network (CMN)

Simard and colleagues (1997) showed that different plant species transfer C bi-directionally via a CMN. This favours the hypothesis that plants in the forest live in community, share nutrients and water, and not compete. In laboratory experiments, Grime and colleagues (1987) earlier showed that AMF connect plants via a CMN, and C is transferred from plant to plant. Many studies highlight the importance of microbe diversity in the soil and its impact on plant health and productivity, and excellent publications are summarizing scientific studies (e.g., Van der Heijden et al. 2008, Bardgett and van der Putten, 2014). Many plants species are estimated to be utterly dependent on microbial symbionts for growth and survival. (Brooks et al. 2006) could show in experiments with deuterated water that if water is applied locally, the Mycorrhizal network (MN) can move water horizontally through the soil in an asymmetric pattern dependent on the formation of the MN. Similar studies showed that water taken up by deep taproots is spread via the MN horizontally in the soil (Egerton-Warburton et al. 2007). Beiler and colleagues mapped the belowground distribution of the fungi *Rhizopogon vesiculosus* and *Rhizopogon vinicolor* using DNA microsatellite analysis to determine the architecture of a mycorrhizal network in an interior Douglas-fir trees (*Pseudotsuga menziesii var. glauca*) forest, and they found that up to 19 trees were linked with the same fungi (Beiler et al. 2009). The study further indicates that large trees play a foundational role in serving as a hub for younger trees and stabilizing the ecosystem by providing younger trees in stress conditions with nutrients and water. Old trees transfer water, nitrogen, and carbon via fungal networks to tree germinants, which show significantly greater survival in the presence of other trees than in isolated positions (Teste et al. 2009). These adult trees that support the seedling growth have also been assigned as “mother trees” (Simard, 2017).

Studies show that soil biodiversity loss and simplification of soil microbial diversity due to human activities in agriculture/forestry impair multiple ecosystem functions, such as plant diversity, decomposition, nutrient retention, and nutrient cycling (Wagg et al. 2019). The experiments indicate that changes in soil communities and the loss of soil biodiversity threaten ecosystem multifunctionality and sustainability.

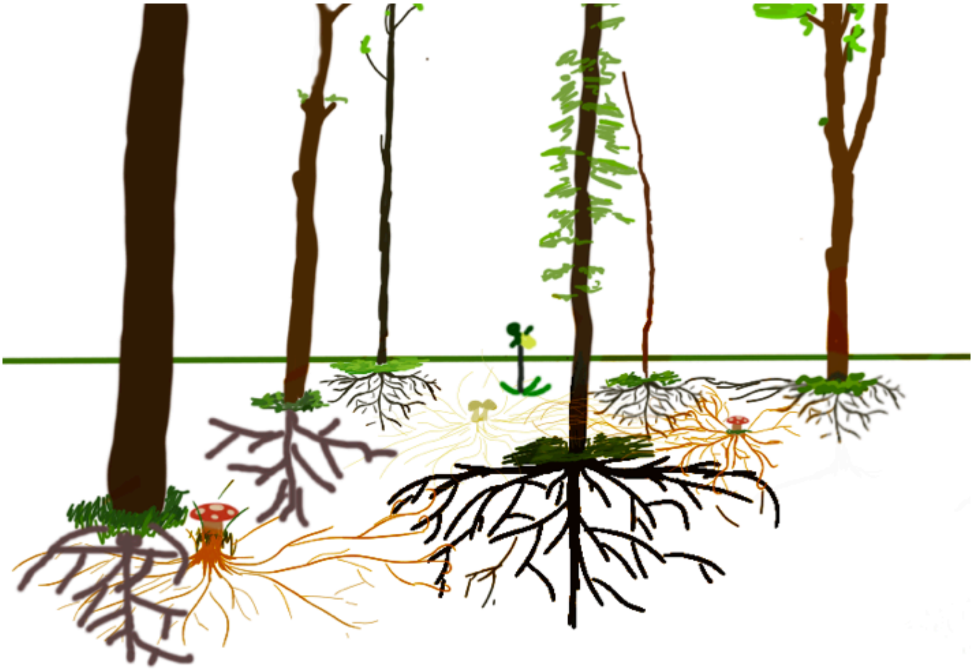

Figure 2. CMN. ECMF species cover the roots of trees and form networks. Orchids are only germinating in the presence of mycorrhizal fungi.

Forest management

Temperate forests, located in the most populated areas in the world in the middle latitudes, have been cut down for long to give human civilization and agriculture space. Seasonal changes throughout the year characterize temperate forests. These forests can be roughly classified into three types. There are coniferous evergreen forests, deciduous forests with trees shedding leaves every year, and mixtures of these two types. The cold winters mean a lower upper ground diversity in winter than in tropical or subtropical forests since many animals hibernate or migrate to warmer regions. The belowground biodiversity is highly complex and vital for a functioning forest ecosystem. It shows high flexibility due to seasonal changes in moisture, plant activity, temperature, and nutrient cycling. But the complexity of the microbial biodiversity has been disturbed by anthropic activities resulting in species loss due to soil erosion, changes in land use, fire, and invasions of non-native plants (DeFries et al. 2010).

Removal of all trees in a forest, or in other words, “clearcut harvesting”, reduces the presence of mycorrhizal fungal in forest soils and makes reforestation more difficult (Perry et al.1987). Symbiotic fungi will diminish in the absence of plants, because they lack photosynthetically derived carbon to grow a network for exploration of nutrients and water (Smith and Read, 2008). These reduced levels of mycorrhizal inoculum can be especially problematic for the survival and growth of seedlings for regenerating in clear-cut forests (Simard et al. 2021).

A recent review about fungal diversity concluded that the lower fungal biodiversity, the higher the intensity of forest management is (Tomao et al. 2020). Conversely, fungal biodiversity can be kept high when keeping the complexity of plant species high and promoting a higher abundance of deadwood. Studies show that in the case of clear-cut openings, the mycorrhizal abundance drops with increasing distance from trees along forest edges and is lowest in the centre of clearcut openings (Simard, 2017). In addition, tree seedling survival is much better within 5 m of residual trees and is dropping with increasing distance from adult trees.

Figure 3. Forest management strategies. Both pictures were taken in a temperate forest in the district of Lower Austria in summer 2021 in a radius of < 1km. **A**. clearcut forest **B**. thinning of forest.

Leaving these "mother trees" and trees to a degree of approximately 60% has little effect on ectomycorrhizal diversity, while clearcuttings will diminish ectomycorrhizal networks and promote the growth of pioneer plants and their symbiotic partners' arbuscular mycorrhizal fungi. In addition, forests store 80% of terrestrial carbon and are the most important terrestrial carbon sink. Therefore, conservation of the forests is therefore crucial for maintaining global carbon cycling and keeping the global ecosystem functional.

The forest carbon sink is based on the flow of carbon from plant uptake. CO_2 is fixed through photosynthesis and is transferred to biomass through plant growth. The carbon stored as biomass can persist for centuries in the wood of long-living trees. In the case of leaf tissues and dead wood, carbon is transferred into soil organic matter, where it can be stored for years before it is released back into the atmosphere by respiration, the mechanism which microbes, plants, and animals use to convert sugars into energy is breaking complex carbohydrate bonds formed through photosynthesis and releasing CO_2 into the atmosphere.

Because of forests' impact in storing carbon, loss of forests will lead to substantial loss of carbon to the atmosphere and recovering of clear-cuts is time-consuming. Studies revealed that in the case of forest clear-cuts, the ectomycorrhizal fungal covered around 6% of forest plants one year after clearcut of the forest, while before cutting around 80% of plants were associated with ECMF (Simard et al. 2021). Loss in an abundance of ECM fungal is explained by interruption of the carbon source through the host plant (Collado et al. 2020).

This loss of ECMF will reduce the survival rate of young tree seedlings, and eventually, a loss of species will occur. Furthermore, a slow recovering forest will not efficiently act as a carbon sink. Several studies indicate that the belowground fungal biomass is not reduced as long as at least 30% of host trees remain after the thinning a forest since the fungal networks are preserved (Simard et al. 2021, Sterkenburg et al. 2019). In contrast to ECMF, saprotrophic fungi will not response negatively when thinning forests since they are not dependent on photosynthetic C (Collado et al. 2020).

Though the belowground biomass may be diminished upon strong thinning of a forest, the reproductive structures (sporocarps) respond by increased production as a mechanism to scan the surrounding area for better growth conditions (Collado et al. 2020).

Climate change

The way we manage terrestrial ecosystems is critical to regulating our climate. By storing carbon in wood and soil, CO_2 is kept out of the atmosphere, contributing to climate change.

In temperate forests, belowground biomass, the soil, and dead organic matter can store more carbon than the aboveground biomass. Aboveground biomass consists of all living plant material above the soil, while belowground biomass consists of the living root systems and soil organic matter. Forest restoration can positively affect "sequestering" additional carbon each year.

To mitigate climate change, we have to consider studying the changes in soil organic carbon (SOC) changes, which refers to the carbon component of organic compounds. Since soil microbes are directly involved in soil organic carbon decomposition, microbial and fungal-rich soil can overcome ecosystem disturbances and can stabilize SOC (Yuste et al. 2011, Xu et al. 2021).

Nottingham et al. (2019) observed that tropical forests enhanced SOC loss due to shifts in microbial community composition toward taxa associated with higher temperatures. Besides this report, recent studies also provide information in temperate forests, which investigate the direct linkage between microbial diversity and the response of SOC decomposition to warming.

A recent study published in Nature Climate Change (Qin et al. 2021) found that in the last decade (2010 - 2019) the Amazon rainforest in Brazil released more carbon than it stored over that period. This is a consequence of deforestation, which increased in 2019 nearly four times compared to 2017, mainly because of weakened environmental protection since Brazil's government changed in 2019. However, the study also indicates that degradation is more significant in releasing carbon than deforestation. Degradation means that forests are damaged due to human activity, such as tree felling or and forest fires. Degradation of forests is also frequently observed in areas near deforestation zones. However, extreme weather conditions such as in 2015, when El Niño led to droughts and tree mortality the carbon losses were even worse than in 2019 with a substantial increase in deforestation. It is now expected that with climate change and the subsequent temperature increase and drought events, carbon emissions will be more significant than the storage capacity of carbon of the Brazilian rainforest.

Climate change is believed to be a direct consequence of rising CO_2 concentrations around the globe (Cox et al. 2000). A strategy to limit the rise of CO_2 concentrations across the globe could be to promote forest conservation and put programs to restore tree growth (Bastin et al. 2019). The authors highlight a vast potential area to transform croplands and urban areas into forest in their paper. In an effort to re-establish tree growth in areas that would naturally support woodlands and forests an increase of photosynthetic carbon capture of 205 gigatonnes could be reached.

Studies predict a decline in ECMF species richness due to climate change in North American Pinaceae forests (Steidinger et al. 2020) This will negatively affect the resilience of trees to abiotic and biotic stresses and on the forest ecosystems in general. Steidinger and colleagues are studying the effect of climate change on MN on Pinaceae because Pinaceae show a low level of host specificity and seem to be ideal for studying environment-community-function relationships (Ishida et al. 2007, Rusca et al. 2006). More than 60% of all trees are associated with ECMF, but the fungal biodiversity decrease with increasing temperature, and ECMF will be lost (Steidinger et al. 2020). The situation with AM is not well understood due to a lack of scientific studies, but data indicate that the diversity and function of AMF is likely to be affected in a changing climate (Cotton, 2018). To date, there are not many studies regarding this issue, also because working with AM in laboratory conditions is more complicated than with ECMF, but data indicate that AMF are essential for plants to resist climate change parameters such as elevated CO_2, temperature increase and drought stress (Compant et al. 2010). Therefore, losing the AM would make plants more susceptible to stress and more likely to get into plant pathogens.

Studies revealed that AMF communities are distributed worldwide and reflect the local environment conditions (Davison et al. 2015). A surprisingly high number of 34% AMF taxa were found on all continents. This is in contrast with the endemism of plants, which to a high percentage are found only locally or distributed in one continent. However, this hypothesis was criticized by Bruns and Taylor (2016), who argue that taxonomic classification of AMF is difficult because AMF are not growing in axenic cultures, and taxonomy is based on spore morphology and sequencing of ribosomal genes. They speculate that these AMF "species" in the phylum Glomeromycota are actually individuals of relatively distantly related taxa.

Potential of mycorrhiza in reforestation projects.

Forests have been converted for thousands of years mainly to use in agriculture. Today, because of climate change, there are efforts to perform reforestation. Afforestation and forest expansion are now accepted strategies to take atmospheric CO_2 and store it in forest vegetation and soil. However, afforestation is difficult due to the degradation and erosion of the complex forest soils. Young trees are particularly vulnerable to drought, heat, and nutrient starvation. Stresses. The knowledge in the beneficial plant/mycorrhiza symbiosis led to improving the growth and survival of young trees in poor and degraded soils using Mycorrhiza inoculums (Jeffries and Rhodes, 1987). One successful attempt to grow young trees with Mycorrhiza was established in growing Argan trees in Marocco (Sellal et al. 2017, Sellal et al. 2019). The argan tree (*Argania* *spinosa*) is endemic to Morocco, and its deep roots are perfectly adapted to the dry and desert-like ecosystem in Marocco. However, the tree is difficult to grow and establish outside its ecological niche in Marocco. Due to its importance in the food and cosmetic industry, there are attempts to regrow the tree in degraded (natural reproduction fails) or deforested areas (Nouaim et al. 2004). Sellal and colleagues (2019) describe a method in argan trees using seeds coated with composite endomycorrhizal inoculum for establishing mycorrhization and successfully establishing argan trees.

Rudawska and Leski (2021) found that the diversity of EMF species in symbiosis with Scots pine in nurseries increases with age. Since the presence of EMF is critical for the successful replantation in fields and forests, this knowledge is crucial for reforestation projects. Furthermore, numerous publications reported Eucalyptus species to benefit from ectomycorrhizal inoculation in nursery and field conditions (Adjoud et al. 1996, Adjoud-Sadadou and Halli-Hargas, 2017).

The potential to promote tree growth in deforested and degraded soil of the Amazon Forest was tested in experiments with the native tropical tree *Handroanthus chrysanthus* and native and non-native AMF (Schüßler et al. 2016). As a result, phylogenetically diverse AM fungi from Ecuador and a non-native AM fungus (*Rhizophagus irregularis*) enormously improved the seedling growth of native potential crop trees. The authors conclude from their studies that inoculation of tropical tree seedlings with AMF most likely improve tree growth and viability, but efficiency may depend on AMF species and plant and should be tested for each tree species.

Importance of healthy forest ecosystems

We observe increasing problems worldwide in maintaining healthy forests. Eventually, climate change has an impact by increased drought and temperature stress. This stress enhances the risk of forest fires and leads to changes in the biodiversity of plants, animals, and microbes. Hence the forest ecosystems are disturbed, and these disturbances lead to diminished resilience of forests and consequently pest and disease outbreaks. Diseases, such as sudden oak death (Brasier and Webber, 2010), ash dieback (Kowalski, 2006), or eucalyptus leaf spot diseases (Bernreiter et al. 2016), have, in consequence, substantial economic losses. To maintain healthy forest ecosystems for resilience, it is important to maintain genetic diversity in a forest by harvesting and reforestation, not only one tree species. Monotypic or reduced typic forests will lose ecosystem biodiversity and will be more vulnerable to diseases. Moreover, invasive species should be controlled since they provide no food value for the forest ecosystem and replace native food sources.

Desprez-Loustau and colleagues explain that homogenous plantations are susceptible to allowing fungal and fungal-like species to search for new ecological niches and adapt to new host plants while becoming more virulent (Desprez-Loustau et al. 2016). There are doubts that the breeding of resistant trees will lead to the disappearance of the disease. More likely, the fungal pathogen, which has a quicker evolution, adapts to the new breeding lines, and starts troubles again. Is the ash dieback problem a consequence of disturbed forest ecosystems, hence lack of fungal biodiversity? A recent publication revealed that the fungal microbiota on the leaf is crucial for the development of ash dieback disease (Becker et al. 2020). The ascomycete *Hymenoscyphus fraxineus*, which is the cause of the ash dieback disease (Kowalski, 2006), first colonizes and infects the tree via leaves. The fungal community structure is crucial to give tolerance to *H. fraxineus* infection (Becker et al. 2020). The authors found in leaves of tolerant ashes several filamentous fungi and yeast-like isolates that showed in cocultivation test potent inhibition of *H. fraxineus*. This effect still needs to be investigated *in planta* to show suitability as a biocontrol agent. Since long it has been known that the application of Mycorrhiza in forests is increasing the biomass (Haselwandter, 1996), but it needs further studies to show that the application of Mycorrhiza could also consequence a healthier forest ecosystem, especially in forests that were the target of bad forest management practices and show levels of degradation. Currently there are many projects studying the effect of Mycorrhiza on plant growth and more and more companies are offering products with Mycorrhiza for biomass enhancement. However, the effect on plant health in complex forest ecosystems is still unknown.

Future research goals will be to study the resilience of trees to tree pathogens in context of a healthier forest ecosystem. Another goal will be to define beneficial forest mycorrhiza species and find ways to grow them in laboratory conditions for future applications. Although AM fungi are not viable without the host plant, and some ectomycorrhizal fungi such as *Morchella* species could not be cultivated in artificial conditions to date, only outdoor cultivation has been successfully established (Liu et al. 2018).

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Chapter 5

Soil biodiversity in Chile: current scenario and future challenges

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Introduction

Chile is a country with considerable latitudinal and altitudinal gradients, which result in a great diversity of climates, ecosystems, and soils. The variation in factors of soil formation and associated processes originate four major soil zones in the Chilean territory, including the hyper-arid to semi-arid area (18-32° S), where the Atacama Desert is located, the Mediterranean zone (32-43° S), the rainy and Patagonian territory (43-56° S) and the insular and Antarctic zones. The edaphic diversity distributed in Chile represents an important reservoir of soil ecosystem services, most of which are highly associated with the edaphic biota. These benefits range from applications in the pharmaceutical industry, bioleaching and bioremediation processes, sustainable agriculture to carbon sequestration and ecological resilience. In Chile, despite the lack of systematic studies on the matter, it is anticipated that soil biota is affected mainly by degradation processes, including deforestation, erosion, deterioration of soil physicochemical conditions, in addition to the effect of climate change. Thus, to support belowground life and its benefits, it is critical to halt these threats in the national context. Considering the contribution of soil biodiversity to the Sustainable Development Goals declared by the United Nations, Chile has an immense opportunity through the support efforts in science, education, and public policy aiming to secure and protect soils and their biodiversity, not only for the current needs but also for the environmental, social, and economic well-being of future generations.

Environmental soil context in Chile

Chile is a country with significant latitudinal gradients, displaying a rich diversity of ecosystems (Luebert and Pliscoff, 2017, Arroyo et al. 2019), where soils develop within a great range of climatic, geological, and geomorphological conditions (Casanova et al. 2013). The continental territory expands on average 4,300 km from north to south, resulting in several climates with important variations in precipitation regimes and temperature gradients (Casanova et al. 2013, Luebert and Pliscoff, 2017). In the continent, Chile's typical geomorphological features are the result of volcanic and tectonic activity; these consist of the Coastal Range, the Central Valley, and the Andes Mountains, which run parallel to the coast and create an altitudinal gradient along with a narrow extension of 180 km approximately from west to east (Casanova et al. 2013, Luebert and Pliscoff, 2017). The formation process of most of the soils in the central and south-central zone of Chile (35-45° S) has been influenced by the physical and chemicals properties of volcanic ash, as well as by climate, characterized by high levels of precipitation (1,500-3,000 mm year^{-1}) and by a low-temperature regime (12-15°C) (Honorato, 2000; González et al. 2015).

These soils generally have a biological activity considered optimal, decreasing in depth, similar to organic matter (Shoji et al. 1993). The physicochemical properties of these soils interact with microbial activity, which stimulates in undisturbed soils the accumulation of organic matter with high levels of C, N, P, and S (Aguilera, 2000; Martínez et al. 2008). The variation in the factors of soil formation and associated processes result in four major soil zones in the Chilean territory, including the hyper-arid to semi-arid (18-32° S), Mediterranean (32-43° S), rainy and Patagonian (43-56° S) and the insular and Antarctic zones. These variations also explain the great diversity of soil orders distributed in Chile, which according to the US Soil Taxonomy system, correspond to: Alfisols, Andisols, Gelisols, Entisols, Inceptisols, Vertisols, Aridisols, Histosols, Spodosols, Mollisols, and Ultisols (Casanova et al. 2013), and based on the World Reference Base for Soil Resources (WRB) include Alisols, Andosols, Arenosols, Calcisols, Cambisols, Fluvisols, Gleysols, Histosols, Leptosols, Luvisols, Nitisols, Podsols, Regosols, Solonchaks, and Vertisols (Gardi et al. 2015).

State of knowledge of soil ecosystem benefits

Ecosystem services or benefits result from the different processes, or ecosystem functions, that occur in nature, which ultimately depends on the structure of ecosystems, that is, on their biotic and abiotic components (Wall et al. 2012). The edaphic diversity distributed in the Chilean territory represents a great reservoir of soil ecosystem services. In the north of Chile, the Atacama Desert, due to its unique and extreme conditions, is a reservoir of a particular microbial diversity with scientific, technological, and conservation relevance (Contador et al. 2020). For example, soils in the hyper-arid Atacama Desert are recognized and studied as sources of novel microbial species with potential applications in the pharmaceutical industry (Santhanam et al. 2012a, Santhanam et al. 2012b, Wichner et al. 2017) and in biotechnological processes including copper bioleaching and arsenic bioremediation (Azua-Bustos and González-Silva, 2014). Similarly, other studies conducted in high altitude locations in northern Chile have also highlighted the importance of microbial groups in the bioremediation of acid mine drainage and arsenic impacted environments (Anguita et al. 2018, Leiva-Aravena et al. 2019). Moreover, other reports conducted in the Atacama Desert have revealed the importance of these habitats in studying carbon dynamics (Azua-Bustos et al. 2017) and abiotic factors explaining ecological resilience under acute environmental conditions (Mandakovic et al. 2018), among others.

The central territory is mostly characterized by a Mediterranean ecosystem type, considered a biodiversity hotspot (Zachos and Habel, 2011). It extends from the southern Atacama Desert towards the Valdivian forests. Here the vegetation is characterized by sclerophyllous scrubs (matorral) and forests (Armesto et al. 2007) adapted to dry warm summers and cold rainy winters, where 2,900 native plant species have been described with 30% of them endemic (Rundel et al. 2016). Soils in the Mediterranean zone are the most productive in the country; thus, they are critical for food provision. In this context, studies performed in agricultural systems had reported positive effects of arbuscular mycorrhizal fungi (AMF) on soil aggregation and C contents when no-tillage was applied (Curaqueo et al. 2010, Curaqueo et al. 2011). This represents potential biotechnological alternatives to sustain the food supply and promote C sequestration, especially when conservation management is established. On the other hand, bacteria exhibiting several plant growth-promoting (PGPR) traits have been isolated from the rhizosphere of productive Alfisols and Andisols. Their effects have been studied in cereals crops (Jorquera et al. 2014, Inostroza et al. 2017, Mutumba et al. 2018), legumes (Sepúlveda-Caamaño et al. 2017, Cedeño-García et al. 2018), avocado (Barra et al. 2017) and tomatoes (Reyes-Castillo et al. 2019). Therefore, an important microbial heritage of native PGPR is stored in agricultural soils, with potential applicability in sustainable agriculture. The soils of southern Chile are mainly soils of volcanic origin, belonging to the order of the Andisols (CIREN, 1999). They are formed by the accumulation of volcanic ash and related volcanic material. They have special characteristics, such as the presence of non-crystalline clays such as allophone, high content of soil organic matter (SOM), low bulk density, high P, and water-binding capacity (Matus et al. 2014a). Soils from the southern territory represent the major reserve of soil organic carbon (SOC) in the country (INAP, 2019; Pfeiffer et al. 2020). Particularly, the greatest reservoirs are believed to be encountered within the Patagonian Andean region and fjords (INAP, 2019; Pfeiffer et al. 2020). In Chilean Patagonia, the potential for C sequestration in Andisols used by silvopastoral systems has been evaluated, determining that C sequestration rates are significantly higher than other soil orders (Dube et al. 2009, Dube and Stolpe, 2016).

Despite the wide variety of ecosystem benefits provided by soil microorganisms, few studies in Chile have specifically focused on this matter. Only 27 studies can be currently (June 2021) found in the Elsevier Scopus database when literature is explored by using keywords such as “soil*”, “Chile”, and “ecosystem services”. From these, 23 studies correspond to original research, from which only three studies are focused on the evaluation of soil biological properties (Castañeda et al. 2015, Castañeda and Barbosa, 2017, Rodríguez-Echeverría et al. 2021). For instance, Rodríguez-Echeverría et al. (2021) explored the biotic and abiotic drivers of soil microbial communities, showing that plant genus *Azorella*, along with aridity and UV-B radiation, were important drivers of the microbial community diversity. These studies show the key role of plants and soil microbial communities in ecosystem services such as biomass production and conservation biodiversity.

Causes of soil degradation and threats to soil species and functional diversity

Globally, the greatest threats to soil biota include the loss of vegetation cover, the introduction of invasive species, acid rain, pollution, agricultural practices, forest fires, desertification, and soil erosion, all this added to global climate change (Orgiazzi et al. 2016, FAO et al. 2020). In Chile, despite the lack of systematic studies on the matter, based on the information collected in the documents Biodiversity and Climate Change in Chile: scientific evidence for decision-making (Marquet et al. 2019) and the Report of the State of the Environment in Chile (INAP, 2019), it is anticipated that soil biota is affected by degradation processes including soil erosion, physical and chemical deterioration processes. Moreover, soils are degraded by deforestation, urbanization, and the effects of climate change, which exacerbates desertification processes, fire occurrence, and droughts (CONAF, 2016; Emanuelli et al. 2016, INAP, 2019, Marquet et al. 2019), all of which represent negative pressures that compromise the diversity and functionality of soil organisms.

Soil erosion and physical degradation

The leading cause of soil degradation in Chile is the erosion induced by anthropogenic pressures, with 37 million ha affected by some degree of soil loss, equivalent to nearly 50% of the territory (CIREN, 2010). Central Chilean regions, coinciding with the main agricultural and forest industries, show the highest erosion rates, which have produced both natural and anthropogenic processes (Armesto et al. 2010). The response of microbial communities and multiple functions due to the erosion impact is still unclear (Qiu et al. 2021). However, it is known that erosion and compaction affect soil microbial communities that directly contribute to the aggregation and formation of the soil structure (Rahman et al. 2017), an important condition to promote C sequestration and mitigate greenhouse gas (GHG) emissions (Sanz-Cobena et al. 2014).

Chilean soils are also degraded by other physical processes such as compaction, subsidence, and anoxia, which have been associated with land use change (INAP, 2019). In this sense, studies evaluating physical properties under different land covers in the Mediterranean zone of central Chile have reported increases in bulk density, with particular reduction of macropores, in soils under exotic forest plantations (Soto et al. 2018). Moreover, other studies conducted in volcanic soils in southern Chile have demonstrated the impact of grazing and irrigation on soil mechanical strength over time (Dec et al. 2021). Despite these advances, the impact of physical deterioration in soil biodiversity has not been deeply evaluated at the national context.

Chemical soil degradation

Anthropogenic activities have increased the contents of potential contaminants in soils (González et al. 2014, MMA, 2018). Probably, the most noticeable example of chemical degradation in Chilean soils is associated with mining activities that increased contents of metals and metalloids [metal(loid)s]. Mining in Chile is concentrated in the north and central regions, where other potential contaminating activities such as thermoelectric, smelting, and cement production also occur (MMA, 2012).

Most of the studies related to soil contamination in Chile have focused on soil physicochemical properties with lower or no consideration of biological attributes, which are crucial in maintaining ecological processes and ecosystem services (Bünemann et al. 2018). One of the first studies attempting to understand biological conditions in metal(loid) contaminated soils, evaluated changes in plant abundance at a long-term contaminated area (Ginocchio, 2000). Later studies assessed AMF in plants that naturally occurred at this metal(loid) contaminated soils (Cornejo et al. 2008). This opened a biotechnological field based on the ability of AMF to alleviate the stress of plants or even promote phytoremediation (Meier et al. 2011, Meier et al. 2012). Recent approaches have evaluated the response of soil microbial communities in metal(loid) contaminated soils (Meier et al. 2017, Moore et al. 2018), demonstrating positive effects of phytoremediation assisted by organic amendments in the microbial community structure (Meier et al. 2021). However, detailed studies about the soil microbial functioning in metal(loid) contaminated soils related to biogeochemical cycles and ecosystem functioning are incipient. Recently though, soil enzyme activities and the community level physiological profile (CLPP) have been used as indicators of soil microbial functioning in metal(loid) contaminated soils (Aponte et al. 2020, Aponte et al. 2021).

Land use change

The transformation of native vegetation into grasslands/scrublands, extensive agriculture, exotic tree plantations, and urban growth have been identified as the main drivers of landscape change in the Chilean territory (Aronson et al. 1998, Armesto et al. 2007, Hernández et al. 2016), causing diversity loss and fragmentation of habitats in native forests (Rodríguez-Echeverry et al. 2018). Over the past 40 years, the central-south Chilean territory has experienced changes in land use, especially due to the conversion of native forests to exotic forests ((CR)^2, 2015). Moreover, most of Chile's population and agricultural production is concentrated in central Chile, causing important changes in land use and land cover over the past decades, which have negatively impacted the provision of the ecosystem services that are important for soil productivity (Oyarzún et al. 2005, Lara et al. 2009).

Although the effects of change in land use from native vegetation to exotic plantations in soil biodiversity is an emerging area of study in Chile, it is expected this would alter belowground biodiversity as it is known that plant species composition and soil communities are tightly related (Schmid et al. 2021). In this sense, recent studies evaluating soil bacterial and fungal communities inhabiting native forests and vineyards have shown similar bacterial communities but contrasting fungal communities in these habitats, suggesting that the latter are more sensitive to land changes than bacteria in these ecosystems (Castañeda et al. 2015). Another study showed contrasting microbial metabolic functions between forest and vineyard soils, suggesting that surrounding native forests act as microbial reservoirs buffering land conversion (Castañeda and Barbosa, 2017).

The effect of land use legacy and modern vegetation patterns in soil microbial biodiversity has also been studied in the national context (Almasia et al. 2016). This study reported different bacterial communities from long-lasting forests compared to more recent established shrubs and concluded that current soil abiotic conditions and not a legacy of past forests explain the structure of these communities. In addition, other studies in ecosystems of central Chile have reported that the conversion of native forest to exotic forest plantations has compromised soil function and soil invertebrates (Cifuentes-Croquevielle et al. 2020). In terms of C storage upon land conversion, studies in Chilean soils have reflected increases in SOC mineralization (Parfitt et al. 2003), promotion of GHG emissions from soils derived from volcanic ash in agricultural and forest ecosystems (Merino et al. 2020), and mobilization of labile and stable C fractions to rivers where these sources are mineralized, further inducing CO_2 release to the atmosphere (Matus et al. 2014b).

Climate change, droughts, and fires

Scientific research related to the effects of climate change on ecosystems and biodiversity are emergent in Chile. Still, current evidence suggests this phenomenon is already affecting the territory (Arroyo et al. 2019). This is the case, for example, of central Chile, which is facing an extensive megadrought occurring since 2010, with mean rainfall deficits of 20–40% ((CR)^2, 2015, Garreaud et al. 2019). This is considered the worst megadrought event occurring within the last 70 years in central Chile, and a quarter of the precipitation deficit associated with this event has been attributed to climate change ((CR)^2, 2015; Arroyo et al. 2019). Although sclerophyllous Mediterranean vegetation, typical of this zone, is well adapted and resistant to summer droughts

(Bussotti et al. 2014), foliage browning and senescence observed in recent summer seasons have been attributed to these events (Arroyo et al. 2019). In terms of soils, Bown et al. (2014) modeled annual soil respiration values across a land degradation gradient in sclerophyllous ecosystems in central Chile, under current climate and future climate change scenarios, indicating that more disturbed soils would be greatly susceptible to the effects of climate change.

Climate change and the conversion of land use can also induce fires (González et al. 2018, Arroyo et al. 2019). Although in Chile, over 90% of fires are provoked by human activities, change in precipitation patterns, elevated temperatures induced by climate change, and the introduction of exotic forest species have exacerbated the occurrence of these events (González et al. 2018, Arroyo et al. 2019). Only in the summer season 2016-2017 about 600.000 ha were affected by a mega-fire event in south-central Chile (de la Barrera et al. 2018). The negative effect of fires in soil properties is well recognized globally, and the greater sensitivity of soil biological properties to disturbances (compared to abiotic factors) is also established (Hart et al. 2005; Mataix-Solera et al. 2009). Despite this, the consequences of land burning on soil biota are less understood than those on soil physicochemical properties (Yeager et al. 2005). This knowledge gap is not an exception in Chile, where studies are gradually increasing in this matter. One example is a survey conducted in temperate ecosystems (*Araucaria araucana* forests) in southern Chile, where microbial activity and diversity were altered one year after fire occurrences, as burned soils had greater values than unburned soils (Fuentes-Ramirez et al. 2018). This study also reported that the increase in microbial diversity does not necessarily relate to aboveground biodiversity recovery after fires. Other studies in central Chile, in soils under Mediterranean sclerophyllous forests, have also evidenced the effect of fires in soil microbial diversity and C storage capacity (Marín and Rojas, 2020; García-Carmona et al. 2021). These have evidenced a significant decrease in microbial counts following six months after fire occurrence (Marín and Rojas, 2020), and an increased basal respiration eight months post fires, which is reflected in lower C contents in burned soils (García-Carmona et al. 2021). Thus, in the studied ecosystem, microbial dynamics and C storage capacity have been disturbed by land burning.

Future challenges to preserve soil biodiversity contribution to the environmental, social, and economic well-being

In the Chilean context, the contribution of soil biodiversity to the Sustainable Development Goals (SDG) declared by the United Nations in 2015 (UNDESA, 2016) has remarkable potential. This is particularly true regarding *SDG 2: End hunger achieve food security and improve nutrition and promote sustainable agriculture and SDG 15: Protect, restore, and promote sustainable use of terrestrial ecosystems, sustainably manage forests, combat desertification, and halt and reverse land degradation and halt biodiversity loss,* as soil biodiversity and functions play key roles in supporting aboveground life. As previously mentioned, there have been important reports confirming the significance of soil biodiversity in food production under sustainable soil management practices and a great variety of agricultural crops in Chile. In addition, the contribution of native soil biodiversity to ecosystem services and land restoration has also been highlighted. Soil biodiversity is also extremely relevant to accomplish *SDG 13: Take urgent action to combat climate change and its impact*, as soil biological activity induced by soil management can contribute to the soil to serve as a reservoir or a source of GHGs. In addition, soil biodiversity contributes to the *SDG 3: Ensure healthy lives and promote well-being for all* *at all ages* by their potential as pharmaceutical reservoirs, by soil biological-based remediation following pollution, and their contribution to the human immune system (Brevik et al. 2020), among others. Other sustainable development goals relaying on proper soil functioning, where soil biodiversity is a key component, include *SDG 6: Ensure availability and sustainable management of water and sanitation for all*, here the biological nutrient cycling, improvement of soil porosity, and the formation of organic complexes between SOM and pollutants are of special relevance; *SDG 7: Ensure access to affordable, reliable, sustainable and clean energy for all*, in this context the contribution of soil biodiversity to the production of biomass for renewable energy is of interest; *SDG 8: Promote sustained, inclusive and sustainable economic growth, full and productive employment and decent work for all*, as previously highlighted, soil biodiversity can contribute not only to the food production and timber industry but also to the pharmaceutical and renewable energy sector, among others, which can have an enormous impact on the implementation of a sustainable economy.

In the national context, the support of soil biodiversity to the SDGs is not granted, and much needs to be done to preserve the functions and services related to this important biotic edaphic component. Advances in current knowledge of soil distribution and characteristics, robust overarching legislation, initiatives to protect edaphic ecosystems and the environment, along with centralized and free data access, among others, are needed to benefit from soils to fully accomplish these goals. Currently, efforts are required to expand the present soil survey system to lands beyond those used for food and timber production and include systematic field studies to complement remote sensing data (INAP, 2019; Pfeiffer et al. 2020). Moreover, Chile lacks a legislative system that protects soils, and most of what is nowadays applied —at least to regulate soil contamination— is of international origins (Neaman et al. 2020). Fortunately, a joint effort among academia, non-governmental agencies, and scientific societies is currently taking place and working on a proposal on soil legislation to be presented to the Chilean Congress (Senado de Chile, 2021). Another institutional initiative in Chile that is directly related to the protection of soils is *the Incentive System Program for Agro-environmental Sustainability of Agricultural Soils* (SIRSD-S, Spanish abbreviation), whose main purpose is to restore the sustainable, productive potential of agricultural soils by supporting the implementation of restoration and conservation practices. This program is a promotion instrument of the Ministry of Agriculture created in 2010, for a period of 12 years, established by Law N° 20,412. Under the current scenario, a new sustainable soil management instrument is required, strengthening the environmental component of soils in the context of climate change, conservation of biodiversity, and provision of soil ecosystem services (Rodrigo Osorio, National Coordinator SIRSD-S Program, personal communication). The National Plan of Land Restoration by the Ministry of the Environment and the Ministry of Agriculture (2020-2031) is another initiative that can significantly protect soil biodiversity and its contribution to SDGs. The Plan is part of the Chile's Climate action to accomplish the Nationally Determined Contribution (2020 NDC), in line with the international commitments, including The Bonn Challenge, The Convention on Biological Diversity (CBD), UN 2030 Agenda, and the national commitments related to the restoration of degraded lands within The Bonn Challenge and Initiative 20 x 20. This public policy aims to promote a comprehensive restoration approach at the landscape level to recover the functionality of ecosystems and degraded soils protecting ecosystem services, the resilience of territories and communities to the impacts of climate change (CONAF, 2019). Chile has also set the goal of having a robust legislative framework regarding climate change. In January of 2020, the Climate Change Bill (PLMCC, Spanish abbreviation) began the process in the Chilean

Congress, which among other goals, aims for carbon neutrality target by 2050 (Observatorio Ley Cambio Climático para Chile, 2021).

Finally, it is worth acknowledging the progress that is currently being conducted in the Chilean Congress to create the Biodiversity and Protected Areas Service (SBAP, Spanish abbreviation), a public service, under the Ministry of the Environment that will oversee the conservation of biodiversity by the administration of a national system of protected areas (MMA, 2021).

Thus, Chile has an immense opportunity to preserve soil biodiversity and secure soil ecosystem services, particularly under current and future challenges imposed by climate change. To support the accomplishment of SDGs related to soil biodiversity, in Chile it is imperative to halt soil degradation, particularly soil erosion and contamination, and support public and private initiatives towards soil restoration and conservation. In this line, is it also necessary to encourage research across soil science disciplines and beyond this, to enhance the current scientific evidence and knowledge related to the effects of climate change—and related phenomena, and soil degradation processes (particularly erosion, physical deterioration, and metal contamination) in soil biological conditions and the implications of these for the correct functioning of soils within ecosystems. Moreover, in terms of regulations it is urgent to articulate a strong legislation system that regulate and prioritize the use of soils in the Chilean territory to protect and conserve ecosystem services. The advances in conservation, knowledge, and awareness of soil biodiversity will deeply contribute to support the UN 2030 Agenda for Sustainable Development, not only for the current needs but also for the environmental, social, and economic well-being of future generations.

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Chapter 6

The state of the study of soil biodiversity in Mexico with emphasis on soil fauna

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Introduction

Mexico is considered a megadiverse country by the United Nations Environmental Program (UNEP), occupying fourth place amongst the greatest biological diversities in the world, surpassed only by Brazil, Colombia, and Indonesia. Also, Mexico ranks third in the diversity of terrestrial and aquatic ecosystems and has a remarkable cultural richness. It is one of the few countries in which almost all types of natural environments known on the planet occur. Most of the national territory is covered by deserts (37%), followed by coniferous and oak forests (19%) and dry tropical forests (14%). It is estimated that its territory has between 10 and 12% of the world's species (Llorente-Bousquets and Ocegueda, 2008, Sarukhán et al. 2017, Koleff et al. 2018), and most importantly, a large part of them is endemic to this country. According to the data of the national inventory of species, in the country around 100,000 species of all taxonomic groups have been described in more than 200 years of collection and it is estimated that this corresponds to 30 to 50% of all species known to exist. In the country, however, there are taxonomic groups practically unknown in Mexico, such as viruses and other microorganisms and even some families of insects and other invertebrates or families of plants that have rarely been collected (Christenhusz, and Byng, 2016; Llorente-Bousquets and Oceguera, 2008). Martínez-Meyer et al. (2014) carried out an analysis of 56 biological groups and reported on 94,412 species in Mexico which represents 8.6% of the world's total of the groups examined, with average endemism of 40%. The authors conclude that on average, Mexico's biota may be 3.3 times larger than what is now known. For example, within the invertebrate group, there are at least 10 phyla of which little or nothing is known about their presence in Mexico and even in the case of other relatively well-known groups such as molluscs and crustaceans, there are also entire orders and families that have not been studied in Mexico. In these estimates, the numbers of species of amoeba, protists, fungi, parasitic nematodes and collembola stand out, where it would be necessary to identify many more species than those that have been described to date (Martínez-Meyer et al. 2014). Most of these groups live in the soil or at least for part of their lives.

In part, the enormous biodiversity of Mexico is mainly due to three factors: two biogeographic regions coincide in its territory (Nearctic and Neotropical), it has a complex orography, with the presence of mountains, which rise over 4500 meters above sea level, and complex geological history. These characteristics make Mexico a highly heterogeneous territory with mosaics of different climates and soils. Mexico has 25 types of soils, which consist of 78% of the worldwide types. The 3 main types of soils are Leptosol, Regosol, and Calcisol that occupy more than 50% of the territory, and other 19 types that occupy less than 2% each (Semarnat, 2016).

A very important problem is that this great biological diversity is being lost with an increase in the rate of extinction of species, which orders of magnitude greater than the natural rate (Ceballos and Ortega-Baes, 2011). The factors that have been identified as threats to biodiversity and direct causes of the extinction of species vary according to the taxonomic group, but among them are: the destruction of their habitats (particularly in tropical forests, cloud forest, desert and aquatic areas of both fresh and marine waters), pollution, overexploitation, the introduction of exotic species, and more recently, climate change (Sarukhán, 2017). Around 40% of the Mexican soils are reported as degraded in their physicochemical properties (Semarnat, 2016), however, these accounts do not contemplate their biological attributes and biodiversity. Because soil biodiversity provides and promotes many ecosystem services, such as organic matter humification and carbon sequestration, nutrient cycling, nitrogen fixation, pest control, soil aggregation, water filtration and drainage (Lavelle et al. 2016), it is of the utmost importance to concentrate efforts in the study of soil properties and biodiversity that allow us to generate information useful for decision making at multiple scales across the diverse ecosystems in the country.

In Mexico, the study of soil biodiversity started as such in the early 2000s and the first compendium made was a special issue of the journal Acta Zoologica (Fragoso and Reyes, 2001). Later on, Álvarez-Sánchez and Naranjo-García (2003) put together different topics on soil ecology and biodiversity in a book from the tropical humid forest as did Barois et al. (2009). Afterwards, a general book on soil fauna (Estrada Venegas, 2008) and a key to identify the soil fauna (Palacios-Vargas et al. 2014) were published. Recently a collection of information on soil microorganisms was put together in a volume (Álvarez-Sánchez and Rodríguez, 2019).

The world's soil biodiversity according to FAO (2020) is estimated to be 25 % of the total biodiversity of the planet, while Orgiazzi et al. (2016) estimated that 40% of the earth's total species inhabit the soil. The same FAO report indicates that Mexico's soil biodiversity corresponds to 160,000 records and 4700 soil species for the country. An initial search to CONABIO's database regarding soil organisms indicated a total of 8,899 species, but these species were only from three natural protected areas. This number of species at this moment represents approximately 8% of the total number of recorded species in Mexico. Thus, we require further research and analysis using databases to clarify these discrepancies and understand what has been published on soil biodiversity in Mexico.

As soil biodiversity is so extensive it has been divided according to its body size for its study, into microorganisms (bacteria, fungi, protozoa and some nematodes…), mesofauna organisms between 0,2 and 2 mm (Acari, Collembola, some Coleoptera and ants…), macrofauna organisms between 2 and 20 cm (earthworms, Coleoptera, ants, Myriapoda, Orthoptera….) and megafauna (reptiles, and small mammals…) (Swift et al. 1979, Briones, 2014).

At present, there is an effort to see what is known worldwide on soil biodiversity and whether some patterns can be found. This was carried out on specific groups like earthworms (Phillips et al. 2019), nematodes (van den Hoogen et al. 2019) and macrofauna (Lavelle et al. 2022) in order to identify general macroecological patterns related to the distribution and functioning of soil organisms to show their importance in the ecosystem and include them in conservation policies (Cameron et al. 2019, Eisenhauer et al. 2020, Guerra et al. 2020).

In Mexico, not much research has been done regarding the study of soil biodiversity. One of the most important efforts to compile and systematize information on Mexican biodiversity corresponds to the Mexican National Commission for the Knowledge and Use of Biodiversity (CONABIO), which was founded in 1992 and since then a large effort has been made to promote the study of Mexican biodiversity and to systematize its information in the databases. Then, some efforts from different researchers and academic institutions had been made along these lines of research. A recent example is from Dáttilo et al. (2020), who made an assessment on the state of knowledge of ant diversity in the country. In this bibliometric study, they have compiled quite a large set of information which also is reflected in this chapter.

The aim of the chapter is to understand the state of knowledge of soil biodiversity in Mexico and set a baseline. Our main approach considers bibliometric and database searches to get to know how much of soil biodiversity has been sampled in the country and use this information to detect spatial and taxonomic sampling biases and gaps, using the ecoregions to frame for ecosystem diversity. Also, we intend to show to what extent the databases are complementary and how much soil biodiversity is underestimated since many organisms that only spend one stage of their life in the soil are often not considered as part of soil biodiversity. Using accumulation curves we show how much effort it takes to reach an empirical species richness.

Bibliometric search

A search was carried out with the keywords SOIL, BIODIVERSITY and MEXICO in the bibliographic database of Web of Science (WOS) all the years available up to 2020. We found 651 publications written mostly by Mexican scientists; 50% of these works focused on the group of Arthropods (322 publications), particularly ants (27.8% of the total publications), followed by the Coleoptera, Fungi, Nematoda, and Earthworms (*Figure 1*).

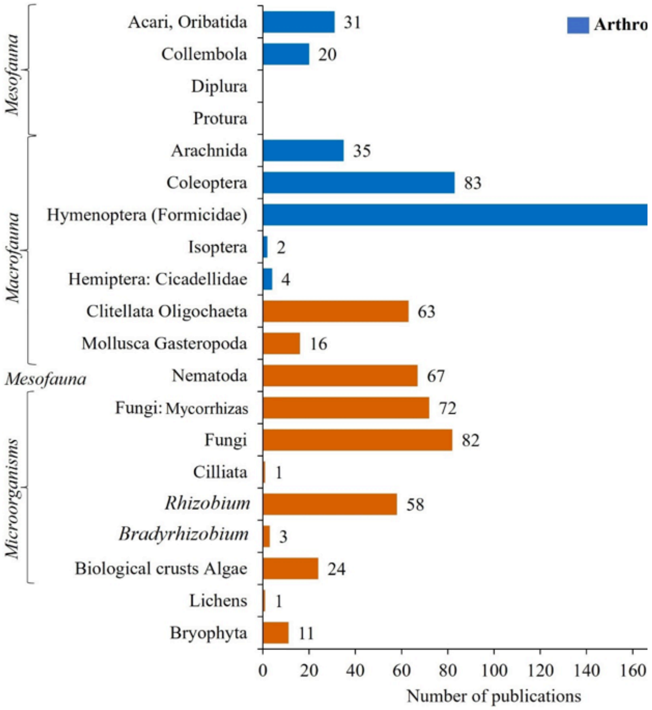

Figure 1. Which taxa and how many times they are studied in 611 references from the WOS (result of the search with the words SOIL BIODIVERSITY and MEXICO up to 2020).

Also, we determined from these publications in which states of Mexico this data was collected (*Figure 2*) and it was observed that the states where most work has been carried out are Veracruz, Puebla, Jalisco, and Tabasco, these being states from the centre and south of Mexico which also is reflected in *Figure 4*. Of the three states that have the highest above-ground biodiversity, Veracruz stands out as having the largest number of soil organisms, whilst the other 2 states only have 8 and 11 studies. Four states have no reports and the others between 4-13.

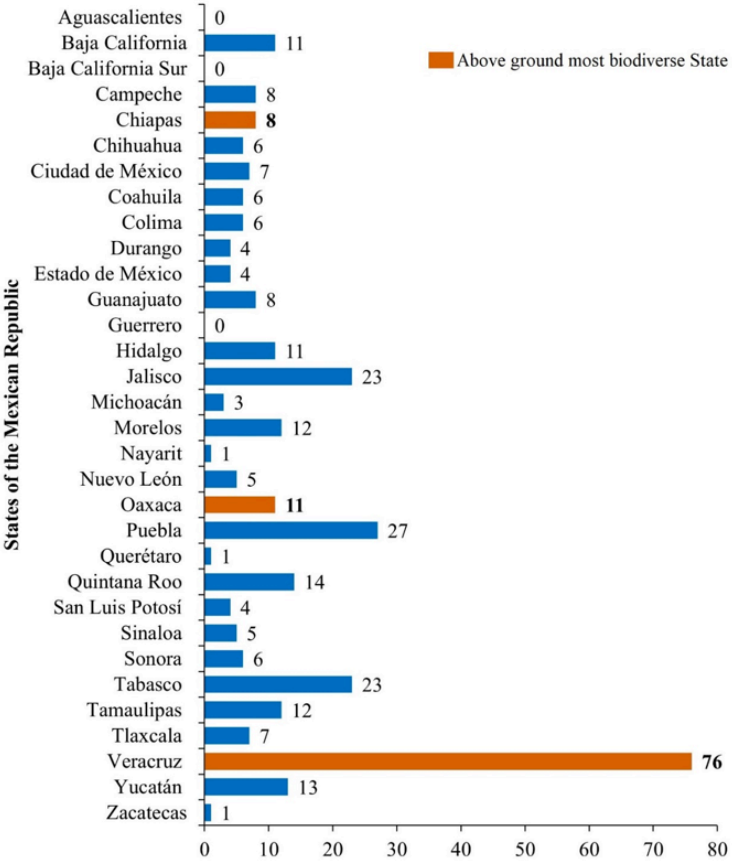

Figure 2. Number of publications from the WOS on soil biodiversity in each of 32 states of Mexico up to 2020..

Below Ground Biodiversity project (BGBD)

At the beginning of 2000, a pioneer project dedicated to the study of below-ground biodiversity called Conservation and Sustainable Management of Below Ground Biodiversity was launched by the Tropical Soil Biology and Fertility programme from CIAT, financed by GEF and implemented by UNEP (GEF, 2021). In Mexico, the work was done in the Biosphere Reserve of Los Tuxtlas, where soil organisms inventories at three different sites and four land uses (forest, fallow, pasture and maize) were carried out. The groups that were sampled at the same time at the same sites were Nitrogen-Fixing Bacteria (NFB), arbuscular mycorrhizal fungi, nematodes, Collembola, earthworms, Miryapoda, Isoptera, Coleoptera, Formicidae. This was a great opportunity to have many taxa sampled as well as basic physicochemical soil properties. The land use soil history was also been taken into account at the same time and it was noticed that there was less soil biodiversity in the maize and pasture fields than there is in the forest and fallow fields; although as a landscape it is a mosaic of land use, the total (gamma) species richness is larger (García-Pérez et al. 2017). This study also showed the sensitivity of some taxa to the land-use history. It was observed that the richness of Nematoda and plant symbionts (NFB and arbuscular mycorrhizal fungi), but not that of the Coleoptera increased with time (20-40 yr) under maize cultivation (Negrete-Yankelevich et al. 2020).

In the inventories, we noticed that the most diverse groups were the Acari, Coleoptera, and Formicidae; although Nematoda could be highly diverse since they were only identified at gender level. Many of the organisms were identified as morphospecies which is the result of the lack of taxonomists (*Figure 3*).

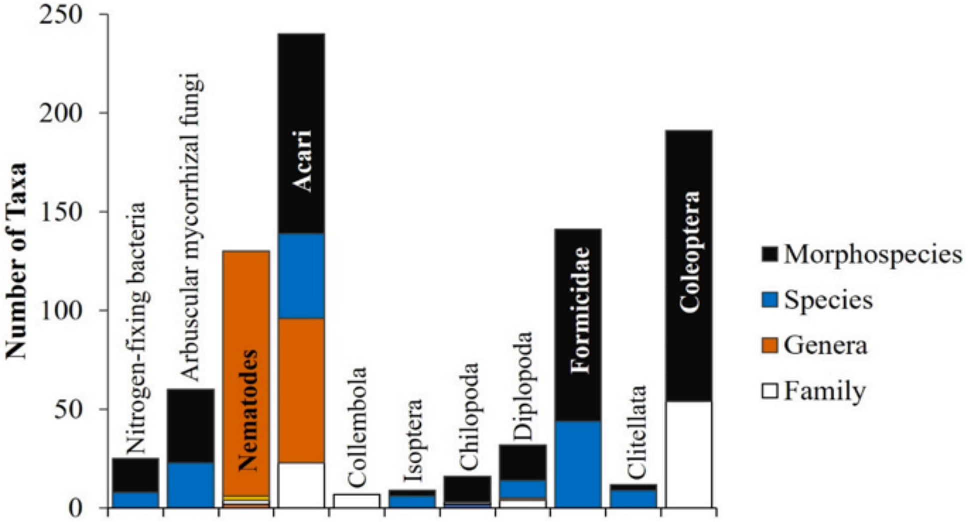

Figure 3. Taxa richness of soil organisms in Los Tuxtlas Biosphere Reserve.

Database search

How much of Mexico's soil has been sampled and how much can we learn from that? The databases used in our analysis were iNaturalist from GBIF and the National System on Biodiversity (SNIB) from CONABIO. This search is mainly focused on the fauna as microorganisms (bacteria and fungi) are not our field of expertise and they are difficult to filter from databases as we do not have the respective catalogues.

As many soil species only live part of their life cycles underground, understanding soil biodiversity can be hard, which is why most catalogues underestimate soil richness. The first step in our database filtering was done using two catalogues, one built by us using information from the literature (mainly from the articles from the WOS and the journal Revista Mexicana de Biodiversidad) and the other provided by CONABIO extracted from the SNIB. The whole data extraction transformation and load (ETL), as well as other processing and visualization creation, can be found in a Python 3 notebook at https://colab.research.google.com/drive/1kWNZk7akYRWy83OsYNBSaLRjFYCDaR8Q?usp=sharing. *Figure 4* shows how these catalogues and databases share information; it is evident that several taxa are unique to these sources of information, which speaks about the need to homogenize taxa and records across these different sources.

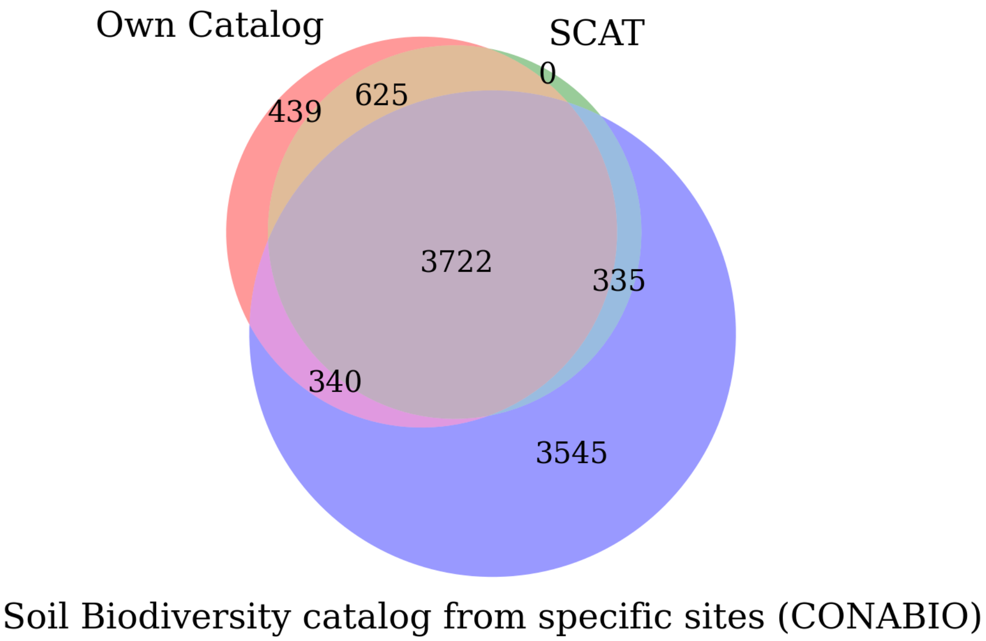

Figure 4. Venn diagram showing the origin of the soil biodiversity information according to the source of information (i.e., from our catalogue derived from the literature, and from CONABIO's databases SNIB and SCAT) showing the number of species or taxa registered (numbers within the circles).

Filtering the taxa from the catalogues can be better understood when species diversity is split according to three levels of taxonomic hierarchy (*Figure 5*). Even though some groups are missing, this can mean at least three things: that species have not been registered in the database, that species are not included in the catalogues, or that species taxonomy is not identical across databases and catalogues.

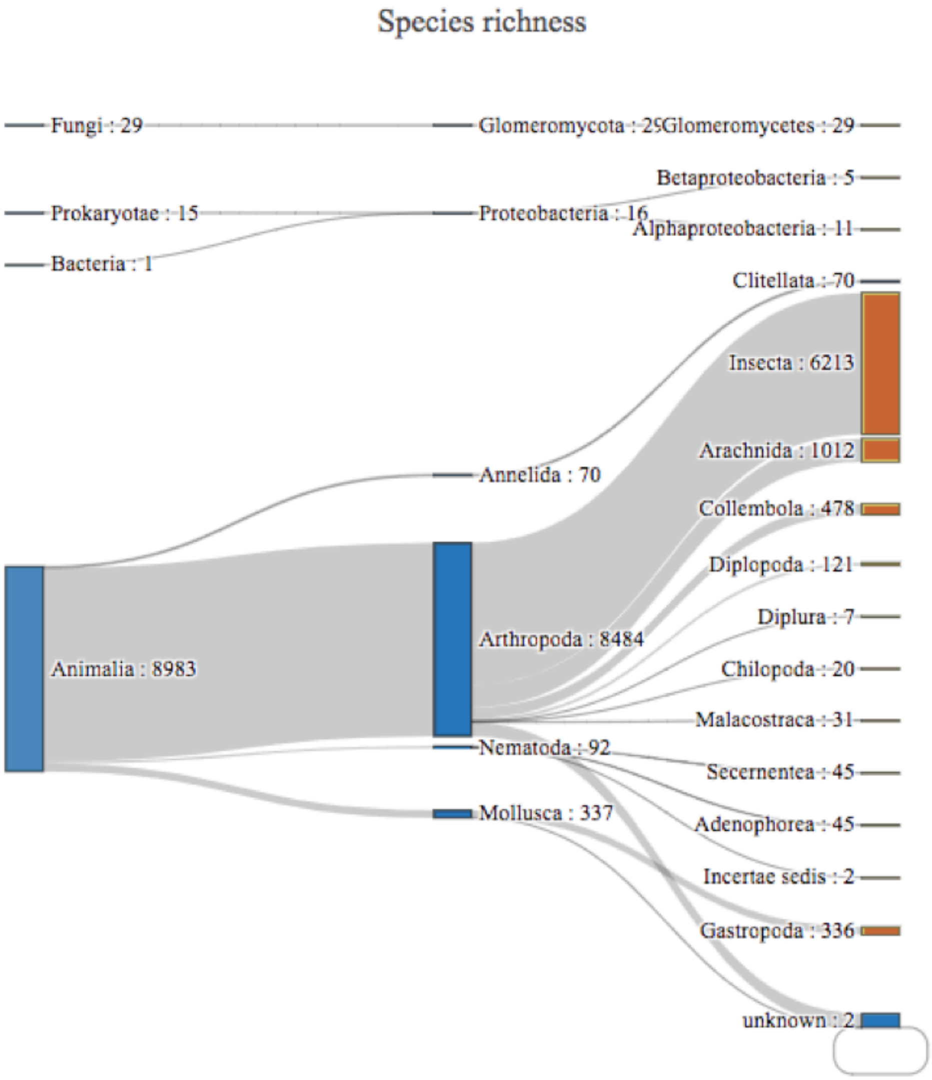

Figure 5. Observed species richness in Mexican soils according to three levels of taxonomic hierarchy (Kingdom, Phylum, Class).

When the databases are put together we can observe that there are 9,029 species, which is more than what has been previously indicated by FAO et al. (2020). The SNIB database has more records of soil organisms, while our catalogue for specific groups showed 439 species that had not been recorded previously in the largest database available today (i.e., CONABIO's; *Figure 4*).

Insecta is the group that has more unique species (*Figure 5*) and is followed by Arachnida and Collembola. Although Nematoda is underestimated as was mentioned above, the microorganisms (fungi and bacteria) are also underestimated, because only some NFB and mycorrhizal fungi were extracted from the database as we believe they come from the soil.

From this list of species, a map (*Figure 6*) was set up with a grid of cells (each cell has an area of 540 km^2) to see how many of them are in specific areas of Mexico to determine the empirical α diversity. This analysis showed there are very few sites with high species richness, being central and south Mexico the regions with the highest species richness (500-700 species per 540 km^2). The sites of the highest species richness belong to the Mexican Trans Volcanic Belt and temperate and tropical humid or dry forest areas. The hotspots detected are Biosphere reserves such as Los Tuxtlas and Tehuacán Cuicatlan or national protected areas as well as some areas near cities and roads which most likely indicate sampling bias (i.e., highly versus poorly sampled regions), and is a well-known pattern for many other taxa generated by the ease of accessibility and institutional and human capacity. In the north, the species richness is very low and even at some sites, there are places without any registration. The cells with no records occupy 34% of the country area (*Figures 6-7*). The ecoregion that has fewer records is the desert region (*Figure 8*).

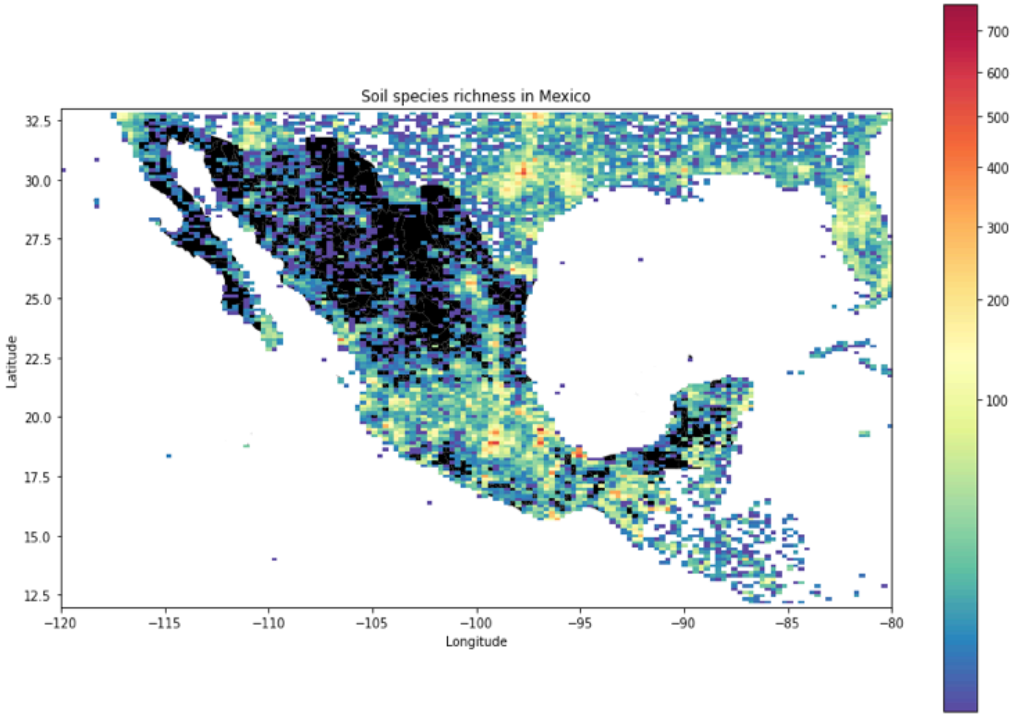

Figure 6. Species richness distribution in Mexico in a grid with cells of 540 km^2. Warmer colours indicate higher species richness while cooler colours indicate fewer species and black empty cells.

The distribution of species richness follows a common trend among inventories. In the hotspots, the cells can have more than 700 unique species but there are many cells with very fewspecies or that have not been sampled (*Figures 6-7*).

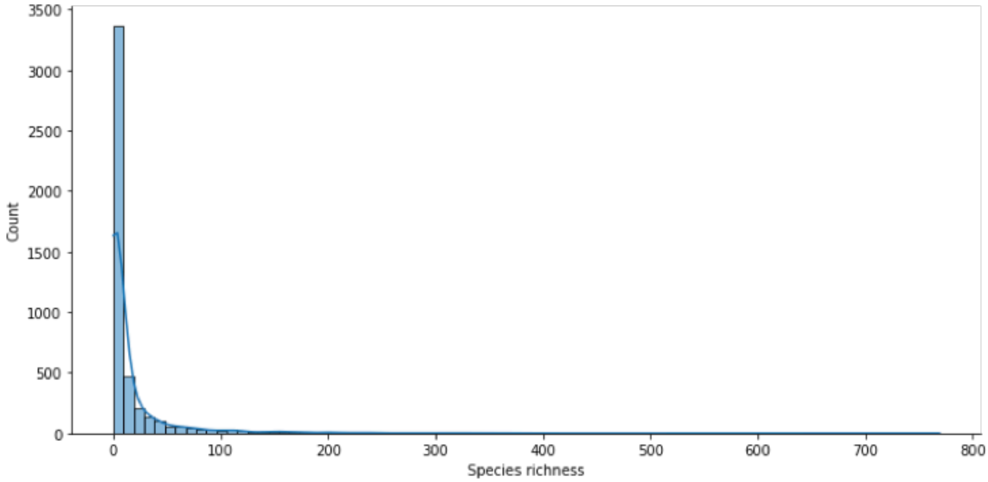

Figure 7. Histogram showing the count of cells from the map having a specific number of species or taxa of the soil, each cell is 540 km².

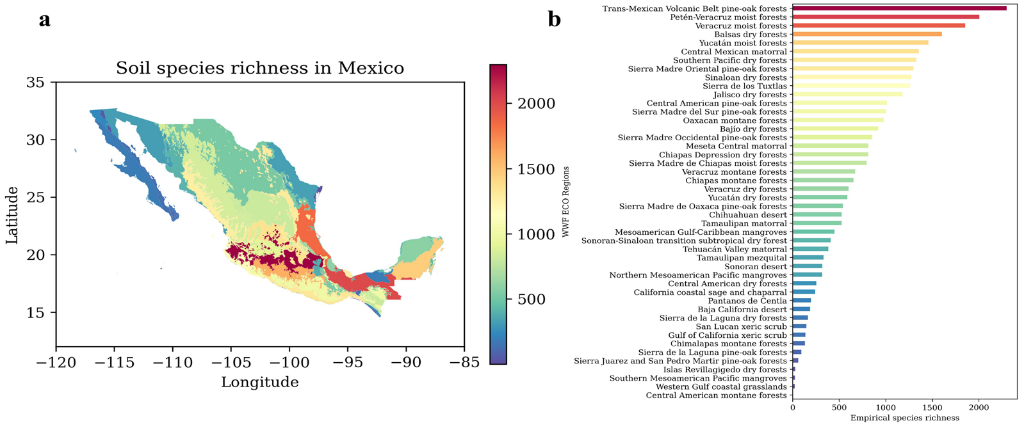

Figure 8. Geographical distributions of species richness by ecoregion as (a) map (World Wildlife Fund) and (b) list.

The spatial analysis of species richness per ecoregion (*Figure 8*) allows us to detect more clearly that the Mexican Trans Volcanic Belt pine-oak forest presents more species richness (> 2000), even more than the Peten and the Veracruz moist forest. This must be because this area has been intensively studied and besides it is an intersection from the Nearctic and Neotropical biogeographic regions. We also have a group of pine-oak forests with around 1200 to 1500 species or taxa, followed by the dry forest in general (1250-1000) and there is a mixture of vegetation from the dry forest to the chaparral (1000-750). Interestingly, the Chihuahuan desert shows more than 800 species and the Sonoran desert has 500 species and the Tehuacan valley matorral has less than 300. After these, there is a miscellaneous type of ecoregions that have low species richness which can be that per se they have poor biodiversity or that they have been under-sampled (*Figure 8 b*). Those regions in the northern and southern frontiers are undersampled because the region boundary might be cut by the national frontiers which show that ecological studies should aim to transcend them for a better understanding of the studied interactions. It is nevertheless intriguing that some deserts can have a medium soil biodiversity, but these regions may also be understudied overall.

The analysis by species accumulation curve over time shows that most studies of soil biodiversity started in 1960 when it started to grow constantly, although since 2010 the growth is almost vertical and we do not see when the plateau will be reached. The total number of species according to the Chao1 estimator suggests 9,594 species; however, we suggest this cypher might also be an underestimation because of the previously stated reasons and should further incentive standardization in global and public access catalogues and be used to prioritize fieldwork (*Figure 9*).

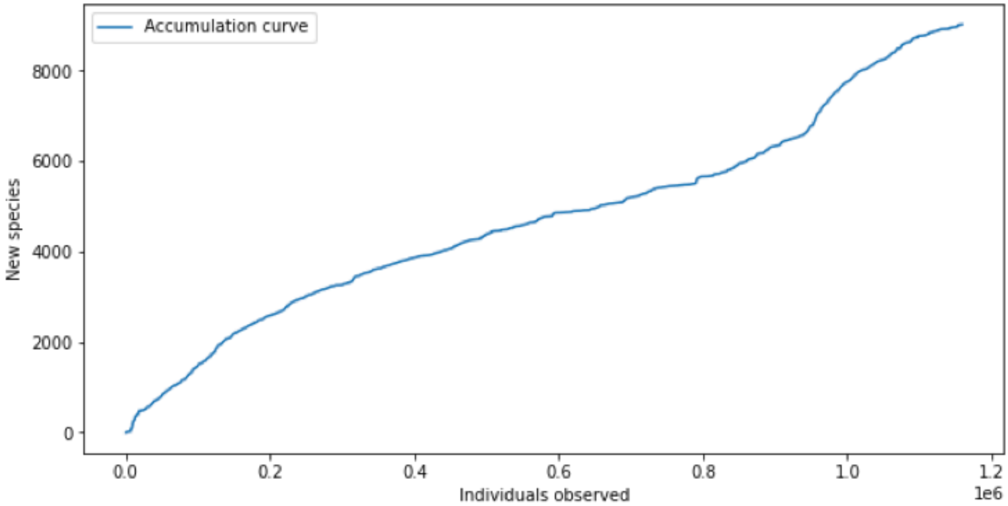

Figure 9. Total soil species accumulation curve from empirical observation across the databases used for this study (scientific notaation used in x axis with e^6 scale).

These empirical curves made by ecoregions show that the curve corresponding to the forests, humid temperate and dry forests will reach their plateau sooner. The curves corresponding to the deserts will require more sampling as the estimators show (Figure 10).

Discussions and conclusions

Many of the results we obtained were also observed by Guerra et al. (2020). The knowledge of soil biodiversity in Mexico is irregular throughout the country. Mainly the more charismatic or agronomically important groups are studied such as ants and Coleoptera or NFB and mycorrhizal fungi for the microorganisms.

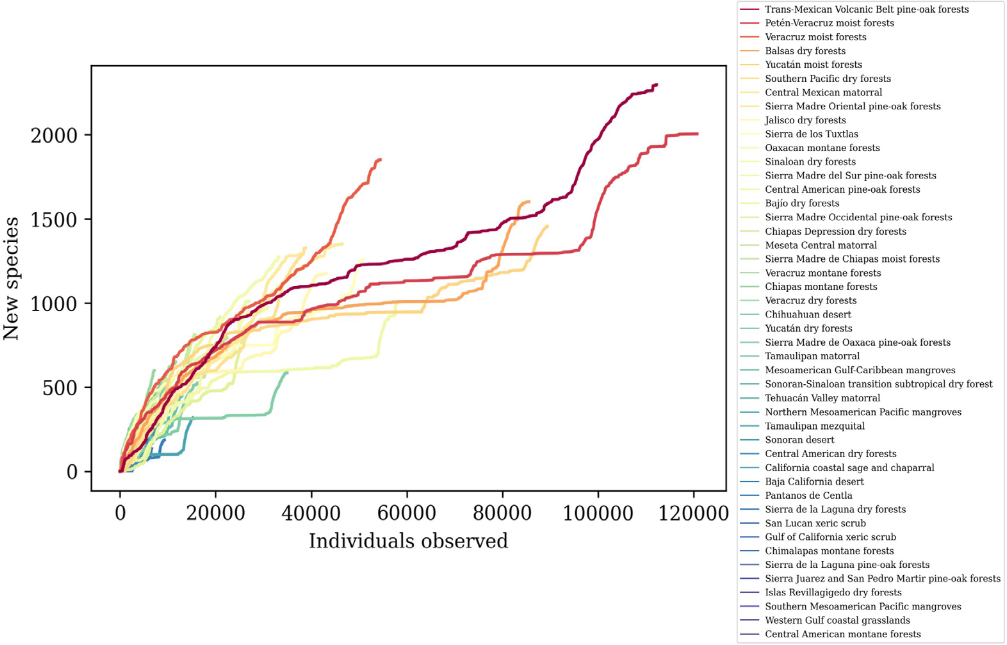

Figure 10. Accumulation curves of species richness by ecoregion.

Most observations were made in the neovolcanic axis and in pine-oak forests, dry and humid tropical forests which also had the highest taxa numbers. The Chihuahuan and Sonoran desert did present a large number of unique species (800-500 per cell) This was also noticed by Guerra et al. (2020) in his macroecological study; as many areas of northern Mexico have not been sampled and are deserts, we can suppose that many more species are going to be identified.

In the databases often species are not identified (Morphospecies) which shows that we do not have enough taxonomists and we require to encourage young people to become taxonomists (*see Figure 3*) or develop taxonomical tools and catalogues that can be easily built and supported by all the community.

All soil biodiversity samplings and catalogues should be publicly available, CONABIO should support these databases and catalogues to allow a data ecosystem that grows crowdsourced but supervised, stating which species spend their lives in the soil, even if it is only at some point in their lives. Systematic and standardized inventories throughout the country should be carried out including agricultural sources.

In addition, we need to make the link between soil biodiversity and function and soil characteristics and promote the coverage of all the soil taxa and in this way to promote the conservation of soil biodiversity and sustainable soil management, studies, where plants are included, would increase the understanding of the soil biodiversity interactions as most plant root systems develop underground.

Monitoring some specific sites across time would be interesting to assess how global change is impacting soil biodiversity and soil functions. It seems that there is no clear correlation between above and belowground biodiversity (Cameron et al. 2019), but countries that are megadiverse in aboveground diversity require special attention to the link between above and belowground biodiversity. Mexico, by being a megadiverse country, needs to study its soil biodiversity with more thoughtfulness to determine whether there is a functional link between above and belowground biodiversity.

A further study should include studying the analysis of the proportion of richness taxa (from different taxonomic levels) per ecoregion, noticing changes in the proportions of taxonomic diversities may be useful indicators to detect anomalous regions which may need more sampling effort.

Acknowledgements

To CONABIO for having provided their catalogue of soil biodiversity and SNIB database. To Laura Bertha Reyes for her invitation to be part of this book.

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Chapter 7

Macroinvertebrate communities: actors and indicators of soil quality

Patrick Lavelle and Elena Velásquez

Introduction

While soil degradation is now acknowledged worldwide as a major threat, a number of important multi authored publications show the magnitude and ecological importance of soil biodiversity (Orgiazzi et al. 2016). A unifying project, the Global Soil Biodiversity Initiative (GSBI, n.d.) is now operating and the World Decade for the restoration of ecosystems proposed by the UN for 2021-2030 (UNEP and FAO, 2020) should use these results. Soils, the basis of 16 of the 24 ecosystem services recognized by the Millennium Ecosystem Assessment (2005), are in great danger and the management options proposed to remedy this are calling out to scientists.

Soils are living entities that host an estimated 30% of Earth biodiversity. Much of it is still unknown and knowledge accumulated on their functions in soils, still uncomplete, is poorly integrated in general soil models and soil management techniques (Filser et al. 2016).

Macroinvertebrates, those invertebrates that can be seen at the naked eye, are present in all soils. They comprise a large number of taxonomical groups that may have numerous species with often large densities and important effects on the soil function. On the other hand, they are fairly well known to most farmers who may associate their presence with some important soil qualities.

This chapter describes the ways soil macroinvertebrates participate in soil function, explains how to build indicators of soil quality based on the composition and abundance of their communities and proposes a methodology to convert their observation by farmers in a tool to measure ecosystem services produced on farm.

A diverse community with a wide functional diversity

In most soils of non-desert areas, 17 different large taxonomic groups of macroinvertebrates can usually be found. Arthropoda comprise 15 of the 17 groups with Gasteropoda and Oligochaeta. To these 17 groups, minor groups are added locally: Embioptera, Onychophora, Nematoda (Mermithidae), Neuroptera larvae or Planaria among others.

These invertebrates are divided into three major functional groups: ecosystem engineers, litter transformers and predators which are not mutually exclusive since certain ecosystem engineers are also litter transformers or predators.

Ecosystem engineers

Ecosystem engineers are organisms that modify living conditions of other soil organisms through their physical effects (Jones et al. 1994) and also community management and biochemical engineering (Lavelle et al. 2016). The main ecosystem engineers are earthworms, termites, and ants although each organism may develop some kind of ecosystem engineering at its own scale. Through their digging and bioturbation activities, they often are the main architects, along with the roots, of the porous and aggregated soil structures. The aggregation of the soil in micro- (<250 microns) and macro- (> 250 microns) aggregates is a fundamental process which allows the soil to fulfill two essential functions, one hydric, the other climatic. The transformation of the soil into a porous structure dispersed among stable aggregates allows the infiltration, accumulation and transfer of water to the phreatic levels. When this structure is degraded or non-existent, water stagnates at the surface and surface runoff causes erosion of the soil.

Aggregates are complex structures the stability and internal structure of which are specific to the physical or biological process that created them (Lavelle et al. 2020). The simple compression of the soil by physical processes (wetting-desiccation cycles) or the enmeshment of soil particles in networks of fungal hyphae produce macroaggregates with relatively low stability, that may not last more than a few days or weeks. Conversely, the diversity of structures created by macroinvertebrates is remarkable (Decaëns et al. 2001, Hedde et al. 2005) and their internal structure sometimes surprising (Blanchart et al. 1997). In general, the macrostructure created by the engineer, for example an earthworm cast, strengthens rapidly by the formation inside them of very stable microaggregates created by microbial agents (Bossuyt et al. 2005).

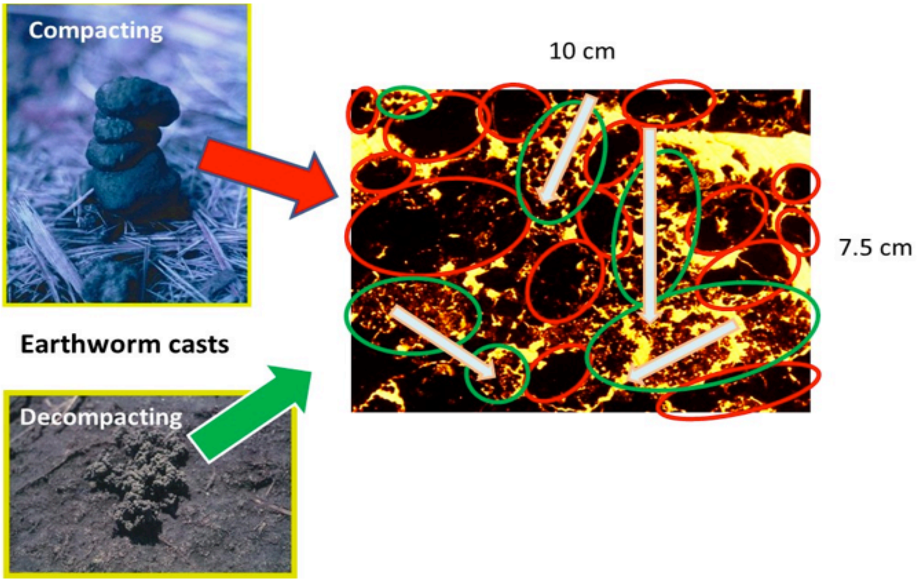


Figure 1. Soil thin section made in an oxisol of an Amazonian rainforest, showing the spatial array of compact macroaggregated structures, surrounded by a red ellipse, made by a compacting earthworm species, and the same structures, surrounded by a green ellipse, disaggregated by decompacting species (ants, termites or decompacting earthworm species), which avoids excessive soil compaction to occur. The white arrows represent the flow of air and water and the storage of the latter in pores of various sizes, allowing retention in a wide range of water potentials.

There are also co-construction dynamics of these aggregates between the earthworms and the roots which exploit the sources of assimilable nutrients present in the fresh castings, adding their exudates and their mycorrhizal networks (Zangerlé et al. 2011). The aggregative effect of some engineers classified as “compacting” may be so strong that some “decompacting” species specialize in the production of loose structures with low structural stability, such as ant deposits or the castings of certain earthworms that partially feed on compact structures (*Figure 1*). The coexistence of these two functional groups allows regulation of the porosity which would optimize the compromise between the different water, climatic and productive functions of the soil. Conversely, when decompacting species communities are depleted, soils may get highly compacted with negative consequences on plant survival (Chauvel et al. 1999).

Litter transformers

Litter transformers are a group of macroinvertebrates that live in the surface litter layer or in the plant rhizosphere where they feed on mainly dead roots. The main function of litter transformers is the natural composting of plant residues, dead leaves and wood on the soil surface, dead roots inside the soil. The most widespread are Diplopoda, Isopoda, earthworms of the epigeic ecological category, Blattaria or Diptera larvae depending on climatic conditions.

They play a major role in the fragmentation of plant debris (comminution) which increases the surface accessible to fungi and other microorganisms for their decomposition. Their digestion is often very incomplete because their enzymatic equipment is generally limited. The fecal pellets that they release into the decomposing litter are microbial incubators in which digestion takes place by the process called external rumen (Swift et al. 1979). Sometimes after, the same invertebrates, or others, re ingest the fecal pellets incubated, in poorly elucidated conditions. Who eats the fecal pellets from whom, after how long of incubation? There is no clear answer to these questions, and it is not clear how much biodiversity is needed to carry out this process and which specific function fulfills each component of the community (Anderson, 1975). The importance and diversity of litter transformers communities is often related to the general rate of decomposition since slow decomposition patterns allow the accumulation of a thick humus litter system with abundant food and diverse habitats for this type of fauna. Conversely, fast decomposition and/or efficient transfer of the litter to the above ground environment by anecic macroinvertebrates is generally associated with relatively low litter transformer communities (*Figure 2*).

Dynamics of decomposition are determined by a series of hierarchical - climatic, then edaphic and finally biological - determinants (Lavelle et al. 1993). Temperature and humidity are the main regulators of microbial activity responsible for over 95% of mineralizing activities (Wall et al. 2008). The quality of the soil, mainly the nature and abundance of its clay content and pH, are important local discriminants of the dynamics of decomposition. Then, the quality of the plant material produced intervenes, sometimes imposing significant restrictions under the laws of stoichiometry: the deficiency in certain mineral elements essential to the constitution of the microbial biomass can greatly slow down its activity (Sterner and Elser, 2002). Finally, the intervention of fauna can locally modulate the patterns of decomposition by creating, through their modes of spatial and temporal distribution, mosaics of spots with different dynamics (Bernier and Ponge, 1994; Rossi, 2003).

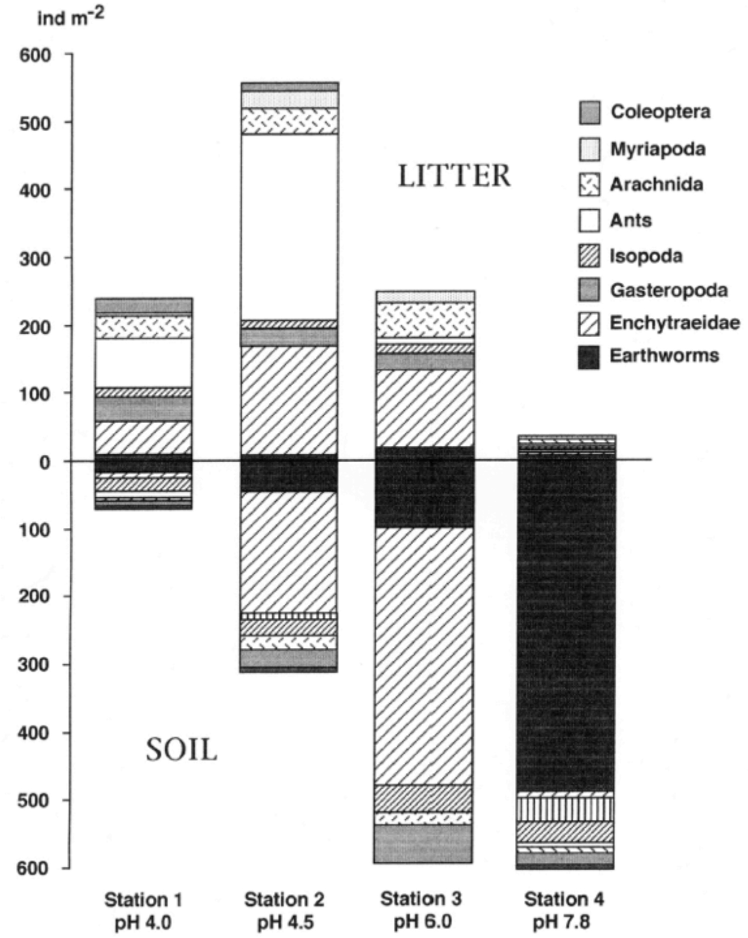


Figure 2. Distribution of major components of the litter and soil macrofauna along a pH gradient in a temperate forest of France (Lavelle and Spain, 2001).

Predators

The predators are distributed among Coleoptera, adults and larvae of certain families, the Myriapoda (Chilopoda), Arachnida, Hymenoptera (ants), Dermaptera and Gasteropoda (slugs) mainly. These predators are generally considered to regulate the various populations of primary and secondary consumers, ensuring a balance of functions. An important literature supports the idea that the functioning of the soil is related to the existence and to the functioning of the chains of micropredators organized in microfoodwebs which live on the microorganisms, the primary consumers of dead organic matter (Nielsen et al. 2015).

Without questioning this vision, we just believe that it fits into a larger soil model in which the ecosystem engineers that create the living conditions and the habitats of these organisms have a preponderant role. This hierarchical vision of soil functioning is consistent with the theory of self-organization, which we recommend applying to understand and manage soils holistically (*Figure 3*) (Lavelle et al. 2016).

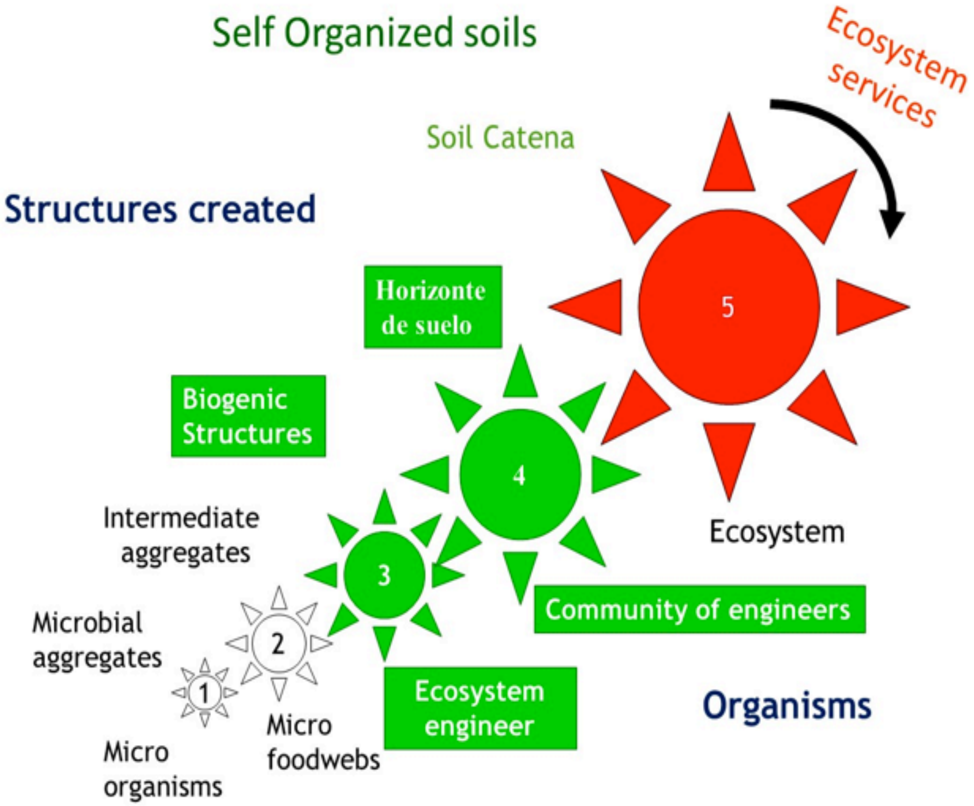


Figure 3. Soil as a self-organized system of biological entities. Organisms of increasing sizes, from microorganisms to micro invertebrates (microfoodwebs), ecosystem engineers and ecosystems (horizontal axis) create structures of different sizes (vertical axis) and organize soil at different nested scales from 1 (microbial aggregates inhabited by microbial communities) to 5 (landscape viewed as a mosaic of different ecosystems). Most soil-based ecosystem services are perceived and delivered at the landscape (5) scale (Lavelle et al. 2006).

Some rhizophagus groups feed on living roots which they consume directly (Coleoptera or Neuroptera larvae) or from which they suck sap (Heteroptera, order Hemiptera). Interestingly, roots are rarely consumed by the largest ecosystem engineers probably due to their chemical composition. The proportion of live root feeding in soils seems to be much lower that the proportion of above ground biomass consumed. However, still much research effort is needed to fully support this statement.

In addition to notable differences in their respective diets and habitats, macroinvertebrates differ for their affinity for very humid conditions (earthworms, Gasteropoda, Isopoda) or their resistance to drought (termites, ants), the thickness of the litter systems or sandy rather than clayey texture.

A standard ISO methodology and the macrofauna database

In the middle eighties, UNESCO launched a Decade of the Tropics initiative with several thematic programmes. Tropical Soil Biology and Fertility (TSBF) was one of them, aimed at applying soil ecology principles to agricultural and other soil management practices. TSBF proposed a set of either standard or innovative methods to do diagnostics of soil quality (Anderson and Ingram, 1993). A specific method was proposed to quantify soil macrofauna communities. Monoliths 25x25 cm in surface and 30 cm in depth arranged at regular distance on a rectilinear transect are extracted and sorted into 10 cm thick strata to extract the macrofauna. A minimum of 5 blocks is necessary to represent a site (Ruiz et al. 2011). A field manual (Ruiz et al. 2008) and a video (UNAL, 2020) explain the details of the method. The extracted invertebrates are identified in large taxonomic units and biomass may be further measured on individuals preserved in alcohol. This method was very successful very quickly because it quickly showed the very great sensitivity of macroinvertebrates to the natural conditions of their environment and to the changes brought about by human activities. An improved version of the technique adopted by ISO (2011) is now used as a reference. By the late 1990s, data collected from a wide variety of sites was merged into a database that now contains several thousand entries.

In an intermediate evaluation of ca. 4000 sites, an average soil macrofauna communities would comprise over 700 individuals per m^2 according to this sampling method. Ants are the major contributors (303 ± 29.3), followed by termites (187 ± 15.2) and earthworms (123 ± 3.8). Diplopoda (21 ± 2.0), Arachnida (17 ± 0.8), and Isopoda (14 ± 1.7) are other groups with over 10 individuals per m^2 on average (Lavelle et al. in revision).

A great sensitivity to soil conditions and management methods

The first studies comparing stands of macroinvertebrates living in soils with different plant cover and soil management methods, immediately showed the great sensitivity of these communities to the conditions of their environment. For example, in a primary forest of Peruvian Amazonia, the total biomass of macrofauna amounts to 53.9g fresh weight divided in a fairly equilibrated way among several major groups. It drops down to 3.1g in some intensive monocropping sites (Lavelle and Pashanasi, 1989). In contrast, pastures have a greatly increased biomass, but this is due to the arrival of an invasive earthworm species, *Pontoscolex corethrurus*, which feeds on the organic matter accumulated in the forest soil and will gradually eliminate the native earthworm species. In the fallow areas, there is a partial reconstitution of the initial forest community.

A global analysis done on a set of ca. 4000 sites shows great effects of both the latitudinal range and plant covers (Lavelle et al. in revision; *Table 1*). Macroinvertebrate densities are much higher on average in tropical (between 30°S and 30°N latitude) than temperate (30-60° latitude) areas. In natural savannas and tropical forests, average densities of ca. 1800 individuals per m^2 and taxonomic richness of 9.4 ± 0.2 are recorded, in the natural forest mainly due to high population densities of social insects. In colder areas, densities and taxonomic richness are far lower. Inside each of these broad latitudinal ranges, human management options globally have a detrimental effect on soil macroinvertebrate communities, although in all situations, the presence of tree components in cropping systems and fallows helps sustain larger populations. Overall, in tropical regions we observed a 54% average decrease in abundance and 36% in taxonomic richness between the highest values in natural ecosystems and annual crop systems without trees.

In temperate areas, corresponding decreases were 64% and 24.4% respectively. Behind these global figures lay a wide diversity of situations and differences in the response of individual taxonomic groups that will not be discussed here.

Finally, we observed that beyond the global and local effects of climate and vegetation cover, the same large groups are present in all soils except termites which are confined to the intertropical zone. These groups are of course represented by different families, genera, or species, but with similar ecological functions. Surprisingly, local conditions of soil, microclimate or management patterns can produce very similar communities in very different geographic areas, from the equator to the higher latitudes explored in this database (Lavelle et al. in revision).

Table 1. Average community density and number of large taxa present in macroinvertebrate communities according to plant cover and latitudinal location (Lavelle et al. in revision).

Plant cover	Average density m^{-2}	Average taxonomic richness
Tropical rain forest	1895 ± 17	9.4 ± 0.2
Savanna	1798 ± 6	6.5 ± 0.5
Tree associated crops and fallows	1188 ± 27	6.3 ± 0.1
Tropical Pastures	1083 ± 23	6.0 ± 0.1
Tropical transitory crops	755 ± 21	6.6 ± 0.2
Temperate grasslands	525 ± 17	4.5 ± 0.2
Dry forests and bushes	252 ± 9	4.2 ± 0.5
Home gardens	233 ± 5	5.4 ± 0.6
Annual temperate crops	232 ± 20	3.4 ± 0.1
Temperate deciduous and coniferous forests	225 ± 30	4.0 ± 0.1
Peat bogs	147 ± 2	4.0 ± 1.6
Urban soils	79 ± 1	3.9 ± 0.1

Macroinvertebrate communities as indicators of ecosystem services

The great diversity of ecological functions and adaptations to living conditions in the soils give macroinvertebrate communities an outstanding indicator value. Numerous studies show that ecosystem services in the soil are linked to each other and also linked to the types of macroinvertebrate communities that inhabit them. In a set of sites with different plant covers in Colombian Orinoquia, all relevant parameters of soil-based ecosystem services were measured and synthesized into indicators of support to plant production, hydrological services and soil macroinvertebrate biodiversity (*Figure 4*). Significant matrix correlation indices were calculated between soil macrofauna data tables established with the TSBF / ISO methodology and the set of data that described the physical functions of the soil, their support value for primary production or their contribution to climate regulation by storing organic matter and reducing greenhouse gas emissions.

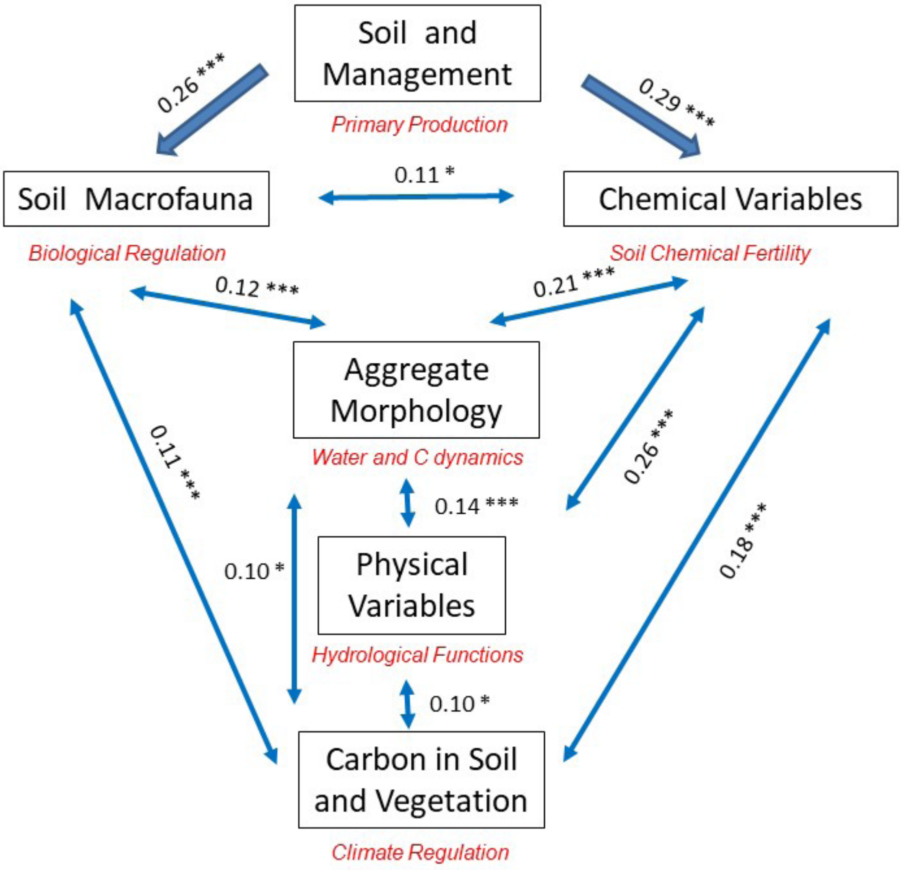


Figure 4. Coefficient of matrix correlation (Rv) among tables of data for the different indicators of ecosystem services and associated p values (*p<0.05; **p<0.01; ***<0.001) in a range of soils with different land use practices in Colombian Oriniquia (Lavelle et al. 2014).

This study and a number of others realized in various sites of tropical America confirms the interconnection of soil chemical, physical and biological functions in bundles of related ecosystem services (Grimaldi et al. 2014; Velasquez and Lavelle, 2019; Rodriguez et al. 2021). These correlations suggest that the composition and abundance of macroinvertebrate communities may be good indicators of the condition of other soil functions that are more complex or expensive to measure.

To this end, we can use the respective density of the various groups considered in the ISO / TSBF method, and a single indicator that summarizes the oberved pattern, usingy the method proposed by Velasquez et al. (2007) that ranks sites along a 0.1 to 1.0 range according to the abundance and diversity of macroinvertebrate communities.

Inviting farmers into the loop

Soil macroinvertebrate communities have shown a strong potential to indicate the various aspects of soil quality and the production of soil-based ecosystem services. We then sought to develop methodologies allowing to practically use this characteristic and putting at the disposal of field practitioners' tools adapted to their particular knowledge.

The approach used consists in creating and comparing two types of knowledge, the scientific knowledge that we know how to generate, and the knowledge of practitioners based on observations and practice in the field.

Scientific knowledge: indicator species

Four research projects conducted in tropical America have made it possible to develop a methodology to identify species indicator of production levels of different ecosystem services. On the one hand, ecosystem services are assessed using indicators that transform a series of relevant variables into a single indicator that ranks the various sites analyzed on a scale of 0.1 to 1.0 (*Table 2*). We can then use the IndVal method (Dufrêne and Legendre, 1997), which calculates an indicator, produced -for a class of determined values of the SE indicator- the fidelity (maximum when the species is always found in the category considered) and specificity, maximum when only this species is associated with the category considered.

Ecosystem Services	Type of data	List of variables	Methods
Biodiversity	Quantitative assessment of soil macroinvertebrate communities	Density per m^2 of 16 orders of soil macroinvertebrates	ISO 23611-5 sampling method
Plant production	Chemical Fertility	Macronutrients, pH, C, CEC, Al^{+3} saturation	Standard methods
Water related services	Physical quality	Bulk density, resistance to penetration, shear strength resistance, water content	Standard methods
Resistance to erosion	Macroaggregation	Relative proportions of biogenic, physical and root macroaggregates and non-aggregated soil	Velasquez et al. 2007b

Table 2. List of variables used to measure synthetic proxy indices of soil-based ecosystem services (Lavelle et al. 2021).

This research has been developed in a set of natural dry forests and derived cropping systems in Nicaragua, La Danta (Rousseau et al. 2013), rainforest remains, pastures and cropping systems in deforested areas of the Amazon in Colombia (Caqueta) and Peru (Loreto) (Duran-Bautista et al. 2020, Vasquez et al. unpublished data), and natural savannas and cropping systems in the plains of Colombian Orinoquia. (Meta) (Sanabria et al. 2014).

We have found that most macroinvertebrates can be significant indicators of either water functions, support of primary production by chemical fertility or indicator of overall soil macroinvertebrate biodiversity. In each of the sites studied, significant indicators were found, with only one exception (*Tables 3 and 4*). It is interesting to note that even if certain species of termites or genera of ants have turned out to be good indicators, the large units, at the level of families or orders are also effective which creates *a priori* favorable conditions for their use by soil practitioners whose taxonomic recognition level may be somewhat limited (Rousseau et al. 2013 and Vasquez et al. unpublished data).

	Sites	High	Medium	Low
Biodiversity	La Danta	6	1	0
	Llanos	5		0
	Caqueta		1	
	Loreto	6	0	0
	Total	17	2	0

	Sites	High	Medium	Low
Physical Quality	La Danta	0		0
	Llanos			
	Caqueta			1
	Loreto	2		
	Total	2		1

Chemical Fertility	La Danta	1		1
	Llanos	2		1
	Caqueta	1		
	Loreto			4
	Total	4		6

Soil Macroaggregation	La Danta	2		
	Llanos	5		
	Caqueta			1
	Loreto	1		
	Total	8		1

Table 3. Number of taxa (p<0.05) indicators of the 4 indicators of soil-based ecosystem services in the four study sites.

		Soil Biodiversity	Chemical Fertility	Physical Quality	Macroaggregation
La Danta (Western Nicaragua) Rousseau et al. 2013	High	Ant *Hypoponera sp.* 0.64	Diplopoda *Cylindroiulus sp.* 0.18		Ant *Atta sp.* 0.22
		Arachnida *Costabrimma sp.* 0.50			Arachnida *Pselaphocher sp.* 0.22
		Coleoptera *Anthicus sp.* 0.43 *Blapstinus sp.* 0.50 *Coproporus sp.* 0.50 Isoptera 0.82			
	Medium	Larva *Elateridae sp.* 0.54			
	Low		Coleoptera *Coproporus sp.* 0.18		

		Soil Biodiversity	Chemical Fertility	Physical Quality	Macroaggregation
ANTS Llanos orientales (Meta, Colombia) Sanabria et al. 2014	High	*Brachymyrmex sp2* 0.22 *Hypoponera sp.* 0.15 *Monomorium pharaonis* 0.14 *Pheidole inversa* 0.25 *Pseudomyrmex pallens sp.* 0.18	*Hypoponera creola* 0.22 *Hypoponera punctaticeps* 0.14		*Acromyrmex sp.* 0.23 *Nylanderia fulva* 0.28 *Pheidole inversa* 0.23 *Pseudomyrmex pallens* 0.18 *Solenopsis sp.* 0.12
	Low		Ectatomma ruidum 0.26	*Hypoponera creola* 0.28	

		Soil Biodiversity	Chemical Fertility	Physical Quality	Macroaggregation
TERMITES Deforested Amazonia Colombia (Caqueta) 8 different land cover	High	*Anoplotermes sp.* 0.34 *Cyrilliotermes angulariceps* 0.15 *Diversitermes diversimilis* 0.15 *Diversitermes tiapuan* 0.18 *Nasutitermes guyanae* 0.28 *Neocapritermes talpoides* 0.18		*Diversitermes tiapuan* 0.15 *Nasutitermes sp4* 0.28	
TERMITES Deforested Amazonia Colombia (Caqueta)	High		*Sylvestritermes holmmgreni* 0.13		
	Medium			*Aparatermes silvarii* 0.13	

		Soil Biodiversity	Chemical Fertility	Physical Quality	Macroaggregation
MACROFAUNA Large taxa and ant general. Deforested Amasonia Peru (Loreto) Vasquez et al. unp. data	High	*Anochelus* 0.13 *Centromyrmex sp.* 0.13 *Cremagaster sp.* 0.29 Chilopoda 0.54 Diplopoda 0.39 *Ectatomma sp.* 0.18 Gasteropoda 0.14 *Linopithema* 0.16 *Nylanderia sp* 0.21 Termites 0.54 *Trachymyrmex* 0.14 *Typhlomyrmex* 0.11	*Acromyrmex* 0.19 *Centromyrmex* 0.14 Coleoptera 0.36 Diplopoda 0.43 Isopoda x.xx *Pachycondyla* 0.23 Termites 0.54	*Pachycondyla* 0.24 Termites 0.37	*Nylanderia* 0.33 Termites 0.52
	Medium	Coleoptera 0.36	*Solenopsis* 0.35 *Typhlomyrmex* 0.13	Arachnida 0.40 Coleoptera 0.39 Diplopoda 0.51 Isopoda 0.40	*Acromyrmex* 0.16 *Camponotus* 0.23 *Pheidole* 0.33 *Pseudomyrmex* 0.18
	Low	*Solenopsis* 0.33	*Camponotus* 0.20	Blattaria 0.43 *Cardiocondyla* 0.39 *Hypoponera* 0.46 *Nylanderia* 0.16 *Solenopsis* 0.04	*Wasmannia* 0.17

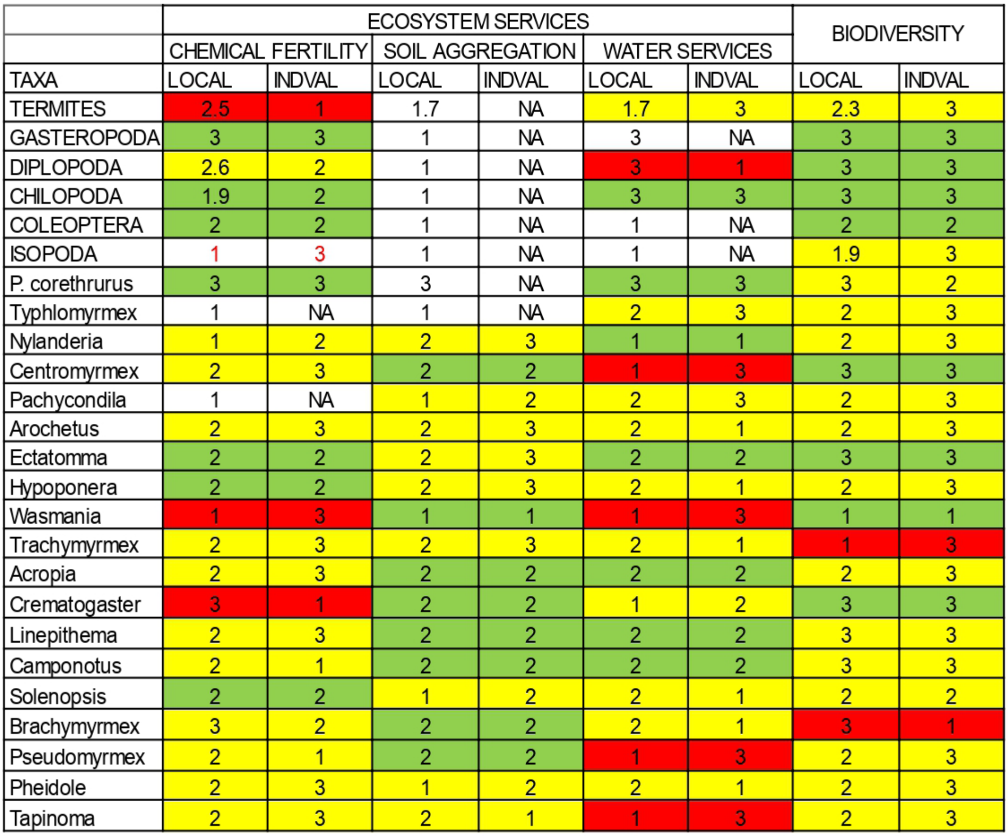
Table 4. Macroinvertebrate taxa as significant indicators of respectively low, medium, or high values of indicators of ecosystem services. Data indicate the indicator value of the taxa in a range of 0 to 1. All values significant at p<0.05.

Participatory method to assess ecosystem services

At the next stage of the study, we worked with farmers to establish a standardized system of ES assessment based on the observation of the presence/absence of particular groups of macroinvertebrates. We have therefore considered the 17 large groups used in the Macrofauna database and, locally, termites and ants, groups very rich in species, identified at the level of species or genus.

After a series of workshops intended to present the subject and to exchange basic knowledge, we went to the field to sample the fauna with the interested persons. After verifying that the macroinvertebrate group, genera or species found were known to these persons, we asked them what perception they had of their indicator value. Three options were proposed, for that, good, average or low value of two services, the physical functions and the chemical fertility as well as the biodiversity.

Table 5 shows the results of the first session, carried out at Yurimaguas in Peru, with a sample of 15 farmers.



	ECOSYSTEM SERVICES						BIODIVERSITY	
	CHEMICAL FERTILITY		SOIL AGGREGATION		WATER SERVICES			
TAXA	LOCAL	INDVAL	LOCAL	INDVAL	LOCAL	INDVAL	LOCAL	INDVAL
TERMITES	2.5	1	1.7	NA	1.7	3	2.3	3
GASTEROPODA	3	3	1	NA	3	NA	3	3
DIPLOPODA	2.6	2	1	NA	3	1	3	3
CHILOPODA	1.9	2	1	NA	3	3	3	3
COLEOPTERA	2	2	1	NA	1	NA	2	2
ISOPODA	1	3	1	NA	1	NA	1.9	3
P. corethrurus	3	3	3	NA	3	3	3	2
Typhlomyrmex	1	NA	1	NA	2	3	2	3
Nylanderia	1	2	2	3	1	1	2	3
Centromyrmex	2	3	2	2	1	3	3	3
Pachycondila	1	NA	1	2	2	3	2	3
Arochetus	2	3	2	3	2	1	2	3
Ectatomma	2	2	2	3	2	2	3	3
Hypoponera	2	2	2	3	2	1	2	3
Wasmania	1	3	1	1	1	3	1	1
Trachymyrmex	2	3	2	3	2	1	1	3
Acropia	2	3	2	2	2	2	2	3
Crematogaster	3	1	2	2	1	2	3	3
Linepithema	2	3	2	2	2	2	3	3
Camponotus	2	1	2	2	2	2	3	3
Solenopsis	2	2	1	2	2	1	2	2
Brachymyrmex	3	2	2	2	2	1	3	1
Pseudomyrmex	2	1	2	2	1	3	2	3
Pheidole	2	3	1	2	2	1	2	3
Tapinoma	2	3	2	1	1	3	2	3

Table 5. Comparison of farmers and scientists' evaluations of macroinvertebrates as indicators of specific range in soil-based ES indicators. Green boxes show total agreement, red boxes opposite perceptions and yellow, compatible evaluations. *P. corethrurus* is an invasive endogenic earthworm. *Typhlomyrmex* to *Tapinoma* are ant genera (unpublished data of Joel Vasquez, Ceila Lao, Patrick Lavelle and Julio Alegre).

Out of a total of 87 evaluations presented in this table, 30 were identical, 46 compatible and 11 opposed. This initial study shows a real potential for creating evaluation systems for ecosystem services based on field observation of presence of macroinvertebrate taxa. They could then be used in a standard methodology of evaluation for payment programs for ecosystem services. The use of this tool would be associated with a training program. Another interesting preliminary result is that, overall, the indicator value seems to be higher for macroinvertebrates identified in large groups (orders or families) than at genus or species level. This very important result will need to be confirmed in larger studies.

Conclusions

Soil macroinvertebrates are essential actors of soil functioning and their provision of Ecosystem Services. Located at a strategic place in the series of hierarchically organized elements of soil self-organizing systems, they are precise and reliable indicators of the condition of the soil and its functions. Much work remains to be done to analyze in more detail their response to the major physical, chemical, and biological parameters of soil condition. The existence of a standard methodology recognized by ISO and of a large associated database, with rapidly increasing numbers of data stored, allows rapid progress to be envisaged in the near future. It then remains necessary to strengthen studies that provide field actors, farmers, agronomists, and field technicians, with reliable tools linking their observation of macroinvertebrate communities present in the field to ES production levels that could be taken into consideration in policies of payment for ecosystem services. Such an approach would have two invaluable advantages. One is to provide the field actor with a tool that he can use based on his personal knowledge, which is not the case with the remote sensing methods or pedological analyzes currently used. The other advantage is of a more social nature. This approach emphasizes the capacity of observation and field experience of the farmer making him an active element of decision making and public policies. This approach breaks with the top-down transfer of knowledge and technology that have marked the farmer's relationship with the world of agricultural research and industry during the green revolution and considerably impoverished their initiative and self-esteem.

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Interdisciplnary perspectives from agronomical point of view

Chapter 8

Conserving and enhancing above-and belowground biodiversities and their relationship with other properties in tropical soils: the success of Brazilian agriculture

Fatima Maria de Souza Moreira, Marcos Gervasio Pereira, and Eduardo Fávero Caires

Current scenario and prospects for Brazilian agriculture

The territorial extension of Brazil is approximately 845 million hectares (Mha), and about 7.6% of this area is used for crop production (*Figure 1*). This cropland area represents only 3.42% of the global cropland area – estimated at 1.873 billion hectares. In addition, about 180 Mha in Brazil is used as pasture (21% of the total geographic area). Areas dedicated to the protection, preservation, and conservation of native vegetation in Brazil total about 560 Mha (66%). In relative terms, the percentage of total geographic area devoted to cropland is lower in Brazil than in most countries.

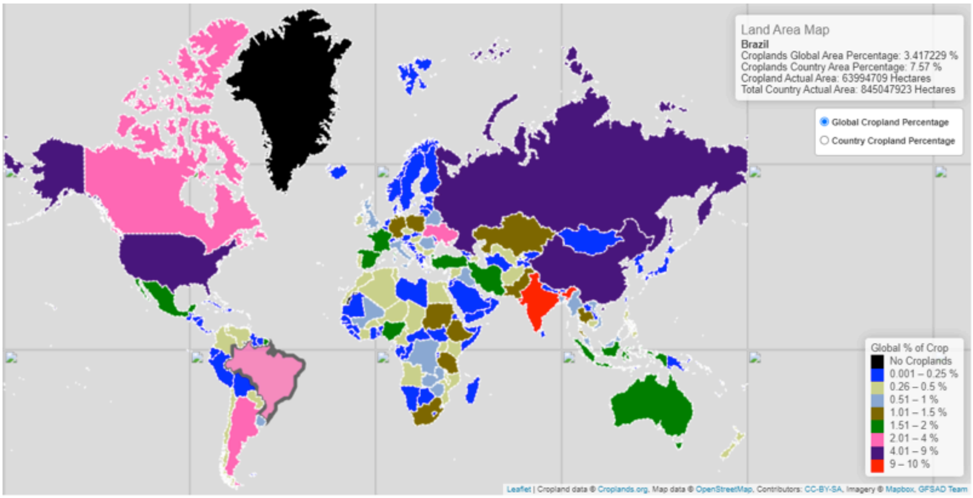


Figure 1. Image retrieved on 03 May 2021 from https://www.croplands.org/app/map/statsMap, maintained by the NASA EOSDIS Land Processes Distributed Active Archive Center (LP DAAC) at the USGS Earth Resources Observation and Science (EROS) Center, Sioux Falls, South Dakota, USA. 2018

The 7.6% occupation by cropland in Brazil contrasts with more than 80% in Moldova, San Marino, and Hungary; 70% to 80% in Denmark, Ukraine, Ireland, and Bangladesh; 60% to 70% of the Netherlands, the United Kingdom, Spain, Lithuania, Poland, the Gaza Strip, the Czech Republic, Italy, and India; and 18% of the United States of America (the USA) and

China. Globally, the largest net cropland areas are in India with 179.8 Mha (9.6% of global net cropland area), the USA with 167.8 Mha (8.9%), China with 165.2 Mha (8.8%), and Russia with 155.8 Mha (8.3%). Brazil ranks fifth, followed by Canada, Argentina, Indonesia, Australia, and Mexico. Brazil is recognized as an agricultural and environmental force since agricultural activities are conducted within an immense and diversified area of native vegetation.

The most prominent products of Brazilian agriculture are soybean (~136 million tons per year), maize (~109 million tons per year), sugarcane (~650 million tons per year), coffee (~2.8 million tons per year), orange (~17 million tons per year), cotton (~2.5 million tons per year), rice (~11 million tons per year), beans (~3,3 million tons per year), wheat (~6.5 million tons per year), cassava (~19 million tons per year), and cacao (0.25 million tons per year) (Conab, 2021). Grain production is gradually increasing in Brazil, along with the use of greater technology and modern cropping practices. Over the last 10 years (2010 to 2020), grain production in Brazil increased 70%, from 151 to 258 million tons (Figure 2). In this same period, cropland area increased only about 40%, from 47 to 66 Mha. The increase in grain production was mainly due to an increase in crop yields, showing the high sustainability of the growth of Brazilian agribusiness. Brazilian grain production is concentrated in the Midwest (46%) and South (32%) regions; the Southeast, Northeast, and North regions account for 10%, 8%, and 4% of grain production, respectively. Soybean and maize make up 85% of total grain production.

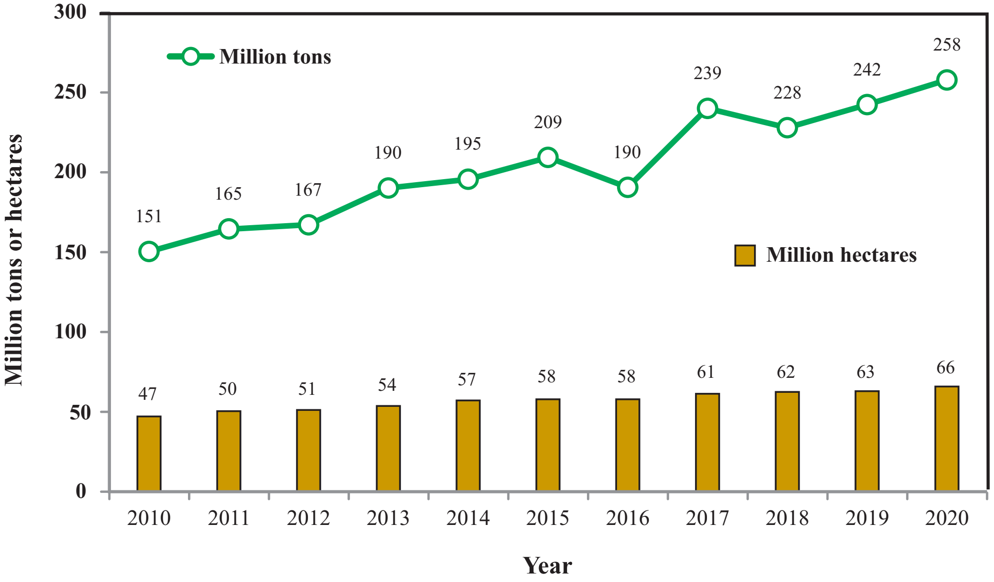


Figure 2. Expansion of grain production and cropland area in Brazil in the period from 2010 to 2020. Data obtained from Conab (2021).

Since Brazilian soils are highly weathered and have low nutrient reserves, increases in crop grain yield have been closely accompanied by increases in fertilizer consumption. Fertilizer deliveries to the agricultural market in Brazil increased from 24.5 million tons in 2010 to 35.5 million tons in 2018. Within this period, consumption of NPK fertilizers increased from 7.9 to 13 million tons (*Figure 3*). The average annual increase in the consumption of N, P, and K via fertilizers was 250, 100, and 285 thousand tons per year, respectively. Since soils in Brazil are naturally poor in P, they require relatively high P fertilization rates to ensure high grain yields. With soybean constituting about 45% of total grain production, slightly more K than N is used in fertilization. Soybean has both high efficiency in biological N_2 fixation and high K requirements, as large amounts of K are exported through harvests. Thus, N fertilization is often unnecessary in soybean, whereas the crop requires high input of K fertilizer.

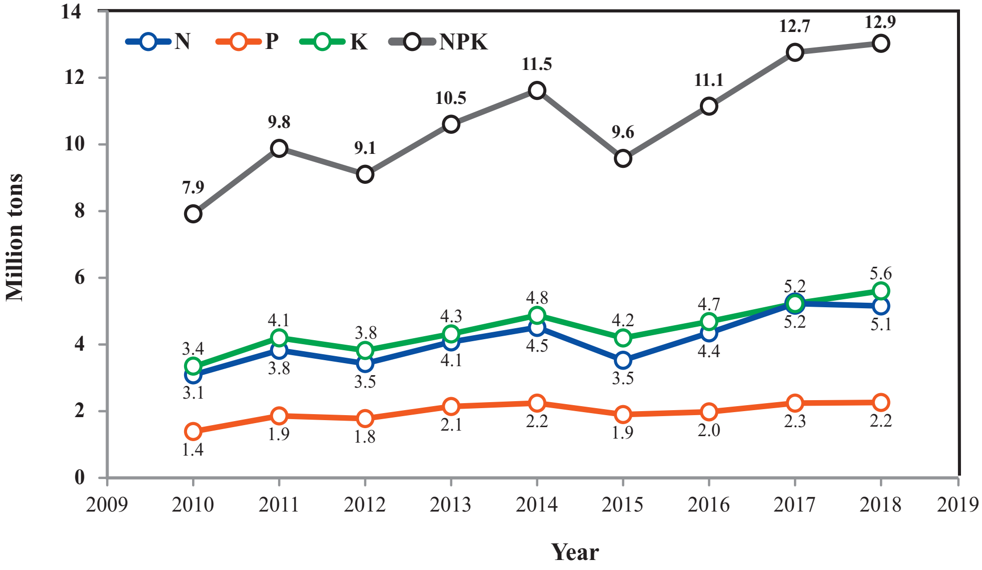


Figure 3. Evolution of NPK fertilizer consumption in Brazil in the period from 2010 to 2018. Data obtained from FAO (2021).

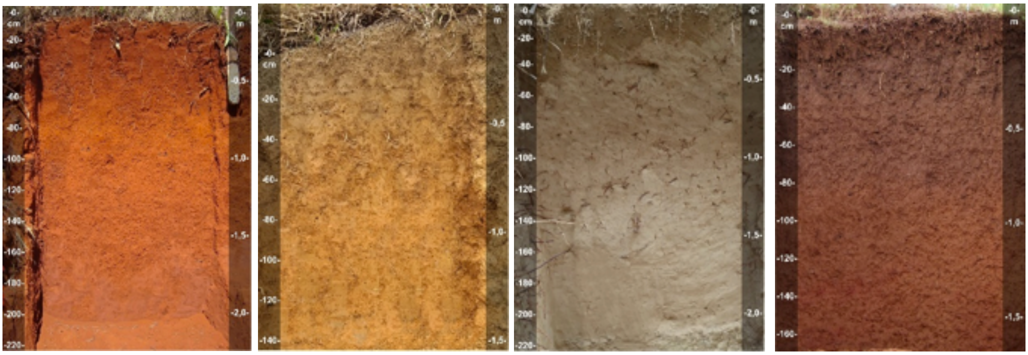
In this decade (2020 to 2030), Brazilian grain production is expected to increase to 318 million tons, while cropland area is expected to rise to 76 Mha. Internationally, expectations are that Brazil will have a nearly 52% share of world soybean exports and 23% share of maize and cotton exports. Such increases in grain production are expected to require even greater consumption of NPK fertilizers in Brazilian agriculture; therefore, fertilizer efficiency will be critical and fertilizer management practices should be improved.

A major challenge for expansion of Brazilian agriculture will be to achieve continual increases in crop yields with higher economic results while preserving soil natural resources and the environment. Greater investments are necessary in sustainable crop intensification processes and in recovery of degraded areas, with the aim of producing two or three crops per year in the same field through more complex cropping systems that have low greenhouse gas emissions and are adapted to climate change. Diversified production systems in no-till conservation agriculture and integrated crop-livestock and crop-livestock-forest systems, which are already in use in Brazil, should be improved and promoted. The use of cover crops and biofertilizers should be expanded, and other soil management practices related to improving the plant rooting profile should be adopted to conserve and enhance below- and aboveground biodiversity in such systems. These practices are expected to increase the carbon stock, nutrient cycling, water infiltration and storage, and soil productivity.

Properties of Brazilian soils

Brazil is a country of continental dimensions with great diversity of geological formations that function as parent materials, associated with wide climatic and topographic variability that contributes to generate soils with distinct morphological, physical, chemical, and mineralogical properties.

Latossolos (Ferralsols), *Argissolos* (Acrisols, Lixisols, and Alisols), and *Neossolos* (Fluvisols, Leptosols, Arenosols, and Regosols) constitute the main soil orders in Brazil (IUSS Working Group WRB, 2015). These three orders of soil together represent approximately 74% of the area in Brazil, and most of Brazilian cropland is composed of these soils. *Nitossolos* (Nitisols) are of limited occurrence compared to the previously mentioned soils, yet they have greater expression in the South region of Brazil, and due to their favorable chemical and/or physical characteristics, they are widely used for agricultural, silvicultural, and pasture activities (*Figure 4*).



(a) (b) (c) (d)

Figure 4. Main soil classes observed in the agricultural production areas of Brazil: (a) *Latossolo Vermelho* (Ferralsol); (b) *Argissolo Vermelho-Amarelo* (Acrisols, Lixisols, and Alisols); (c) *Neossolo Quartzarênico* (Arenosol); and (d) *Nitossolo Bruno* (Nitisol).

Each of these orders has specific characteristics that are the product of the action of pedogenetic processes, and due to these particular aspects, they display weaknesses and potentialities for agricultural use. The main characteristics and weaknesses of these orders are presented below.

Latossolos (Ferralsols) correspond to approximately 33% of Brazilian territory; they occur in all regions of Brazil, but have greater geographical expression in the North and Central-West regions. They are predominantly located in areas with flat to wavy topography and are deep and well-drained, which favors agricultural practices and minimizes erosion processes. Many *Latossolos* (Ferralsols) are acidic and have low nutrient reserves, which results in low natural fertility. Sometimes better natural fertility conditions are found when these soils are formed under drier climate conditions or produced from the weathering of basic rocks or sediments of a calcareous nature (Santos et al. 2018). They have kaolinitic and/or oxidic mineralogy with essentially variable or pH-dependent charges, which favors nutrient loss. Under higher acidity conditions, P fixation problems intensify. The reduced nutrient reserve of most *Latossolos* does not prevent them from being quite productive when well-managed, especially through use of techniques that maintain or increase the organic matter content, contributing to an increase in negative chargers in the soil.

Argissolos (Acrisols, Lixisols, and Alisols) are the order of second greatest occurrence in Brazil, corresponding to 28% of Brazilian territory, with greater expression in the North and Northeast regions. They have an accumulated clay horizon in the subsurface, which leads to slower flow of water inside the soil profile, favoring erosive processes, which can be intensified by the management practices used, the slope of the surface, and rainfall. *Argissolos* cover a huge range of soils, from those with high or low natural fertility to those with high saturation by aluminum. The cation exchange capacity of the clay fraction is variable, and soils with high and low activity clays can be observed. In addition, *Argissolos* are predominantly kaolinitic and have varying degrees of weathering. They have variable depth and can be seen in areas of topography ranging from flat to strongly wavy; their drainage can be good to imperfect.

Neossolos (Fluvisols, Leptosols, Arenosols, and Regosols) are mineral soils that have a lower degree of pedogenetic development, which may be due to topographic conditions that favor greater intensity of erosion processes, promoting constant rejuvenation of the soils; climatic conditions that disadvantage the weathering processes; the short time of formation of the soil; or the resistance of the parent material. These factors limit soil evolution. This order includes the following suborders: *Neossolos Litólicos* (Leptosols), *Neossolos Regolíticos* (Regosols), *Neossolos Quartzarênicos* (Arenosols), and *Neossolos Flúvicos* (Fluvisols).

Neossolos Litólicos (Leptosols) have varied natural fertility dependent on the nature of the parent material. The main limitation is the shallowness of the soil, which restricts root system development, water storage, and management practices. The location of these soils in the sloping reliefs also allows erosion processes to occur at greater intensity. *Neossolos Regolíticos* (Regosols), located in sloped areas, are deeper than *Neossolos Litólicos*, and generally have limitations similar to those observed for *Neossolos Litólicos* (Leptosols).

Neossolos Flúvicos (Fluvisols) are formed predominantly by fluvial sediments, with variable fertility and activity of the clay fraction. They have potential for agricultural use, but when located close to river areas, they may be subject to waterlogging in periods of greater rainfall, limiting agricultural mechanization and causing oxygen deficiency. They can also be used for environmental preservation when located in riparian forest areas.

Neossolos Quartzarênicos (Arenosols) are soils that have a sandy texture, with predominance of quartz. They are environmentally fragile because they have high susceptibility to erosion, low water storage capacity, and high losses by leaching, which can lead to aquifer contamination. In addition, the sandy texture favors rapid decomposition of organic matter. These soils have considerable geographical expression in the region called MATOPIBA, which encompasses part of the states of Maranhão, Tocantins, Piauí, and Bahia, a region that in recent years has witnessed great expansion of the agricultural sector (Donagemma et al. 2016).

Despite their limited extension compared to other soils (1.3%), *Nitossolos* (Nitisols) have wide agricultural use in the South region of Brazil, especially for grain cultivation. They are deep soils, with a clayey or very clayey texture and an accumulated clay horizon. Differentiation between their horizons is less pronounced compared to *Argissolos*, which helps impede erosion processes of great intensity. They may have good natural fertility, but are moderately acidic to acidic, with predominance of low or high activity clays of kaolinitic-oxidic composition.

Due to the intense weathering conditions to which most Brazilian soils were subjected in their formation, iron and aluminum oxides and kaolinite are the main constituents of the clay fraction (Kämpf et al. 2016). These minerals have low cation exchange capacity and can sometimes exhibit anion exchange capacity, a phenomenon that can be observed in iron oxides. Due to these characteristics, management systems that contribute to an increase in the organic matter content in soils are of fundamental importance.

Crop production systems

Before and after no-till conservation agriculture

In Brazil, conventional agriculture with intensive tillage through plowing and harrowing has led to large losses of soil by erosion, causing severe environmental degradation and loss of soil productive capacity. In the South of Brazil, with more slopy areas and higher rainfall intensity, intensive tillage led to an unsustainable condition of extensive soil erosion, and agriculture was at risk (*Figure 5*). This situation of widespread soil degradation persisted through the 1960s. Even though building terraces and contours helped check runoff and surface layer losses, it rarely curtailed erosion sufficiently to make this production system viable. In response, concerned farmers and researchers gradually began to look for other more sustainable production technologies.

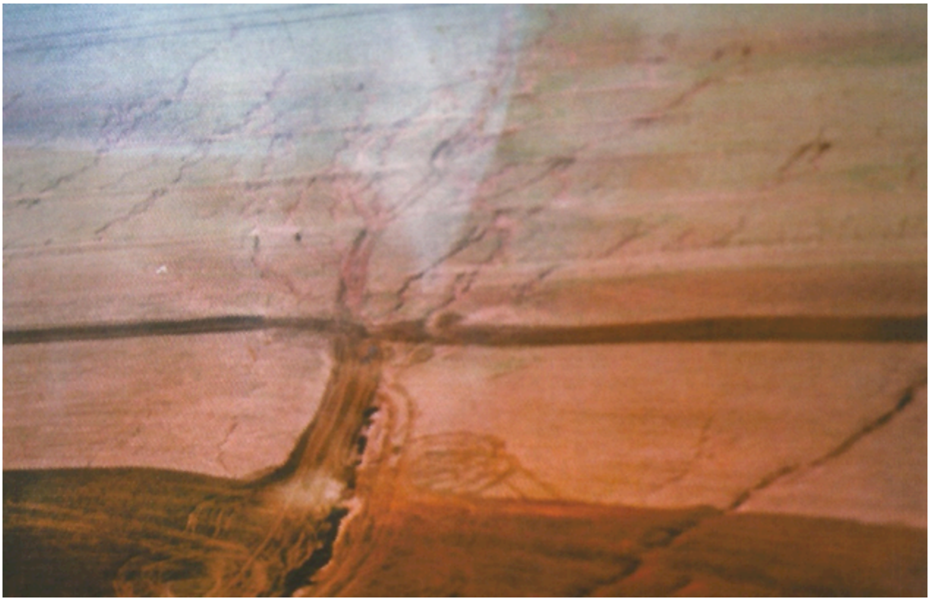


Figure 5. Water erosion processes resulting from intensive tillage in the Campos Gerais do Paraná, PR, region, southern Brazil (1970). Image obtained from Dijkstra (2020).

Conservation agriculture based on no-tillage systems (no-till) began to be practiced in southern Brazil in the early 1970s. No-till farming had three basic principles: (i) no soil disturbance, (ii) permanent soil cover by crop residues or cover crops, and (iii) crop rotation. In this cropping system, crops are seeded into untilled soil by opening only a narrow slot, trench, or band of sufficient width and depth to obtain proper seed coverage; no other soil tillage is done.

Residues from previous cash crops or green manure cover crops are to remain undisturbed on the soil surface after sowing. Crop rotation and growing cover crops are essential practices in this system.

The evolution of no-till farming in Brazil has been well documented (Bolliger et al. 2006, Calegari et al. 2020). No-till farming had limited expansion throughout the 1970s mainly due to the lack of suitable techniques to effectively control weeds, as well as the lack of planters able to work with large amounts of residues. During this period, planters had to be adapted and only 2,4-D and paraquat herbicides were available for weed management. The release of glyphosate in Brazil in the mid-1970s and the improvement of agricultural machinery boosted no-till farming in the 1980s. In addition, in the 1980s, no-till farming began to be used in the tropical, wet-dry savannah region (*Cerrado*) of central Brazil. Even so, the expansion of cropped area under no-till in Brazil was still slow until the early 1990s, when no-till farming reached about 1 million ha. It was from the 1990s on that cropped area under no-till greatly expanded, both in southern and central Brazil. The genetically modified (GM) crop technologies introduced in the 2000s further facilitated weed control using glyphosate. Currently, Brazil is one of the leading countries in the world in adoption of conservation agriculture based on no-till. The latest official information from the Brazilian agricultural census (IBGE, 2019) indicated that no-till farming is used on 33 Mha. Based on the rate of increase in cropped area under no-till in the last 10 years in Brazil, the cropland area under no-till farming is expected to expand to 40 Mha by 2030.

After the “Green Revolution” that brought a series of benefits to Brazilian agriculture, the change in cropping systems to conservation agriculture based on no-till was the biggest agricultural revolution in Brazil. No-till systems with rotation of diverse crops minimize soil and nutrient losses from erosion and increase organic carbon and nitrogen content through increased deposition of plant residues and decreased soil disturbance (*Figure 6*). The increase in soil organic matter increases cation exchange capacity (CEC), biological activity, and biodiversity.

Figure 6. Maize sowing and soybean emergence in no-till systems in the Campos Gerais do Paraná, PR, region. Images obtained from Dijkstra (2020).

One of the biggest challenges in no-till farming is to produce straw/plant residue in sufficient quantity and quality that remains on the soil surface to increase biodiversity and provide sustainability to this cropping system. Proper management of no-till requires the addition of at least 10 Mg ha^{-1} year^{-1} of dry phytomass, including legumes in crop rotation, which promote nitrogen input and organic matter accumulation (Bayer and Dieckow, 2020). High straw production is also important for more efficient weed control. This is a highly relevant aspect because there is currently considerable concern over increased use of glyphosate in GM glyphosate-resistant crops. Intensive glyphosate use has led to the selection of glyphosate-resistant weeds and microorganisms, changing microbiomes (van Bruggen et al. 2018). Increasing the production of straw that remains on the soil surface under no-till decreases weed infestation and helps reduce the use of herbicides, including glyphosate. A problem occurs in areas with low soil cover production, where farmers have increased the dosage and frequency of glyphosate for greater weed control. Improvements in this area and more research are necessary to allow farmers to verify that greater and more diversified production of cover crops not only decreases the use of herbicides and improves the ecosystem, but also results in greater economic return. No-till cropping systems require adequate strategies for supplying straw by integrating fast-growing cover crops and cash crops into the rotations.

In Brazil, several cover crop species have been used in no-till conservation agriculture. In subtropical southern Brazil, the most used cover crop species are black oat (*Avena strigosa* Shreb), radish (*Raphanus sativus*), hairy vetch (*Vicia villosa*), common vetch (*Vicia sativa*), field pea (*Pisum sativum subsp*. arvense), lupin (*Lupinus albus*), rye (*Secale cereale*), and rye grass (*Lollium multiflorum*). In the tropical Brazilian Cerrado region, the preferred cover crop species are millet (*Pennisetum americanum*), Crotalaria (*C. spectabilis, C. ochroleuca, C. juncea, C. breviflora*), buckwheat (*Fagopirum esculentum*), cowpea (*Vigna unguiculata*), pigeon pea (*Cajanus cajan*, normal, dwarf), *Mucuna aterrima* (grey, black, dwarf), lablab (*Dolichos lablab*), *Clitoria ternatea,* Sudan grass (*Sorghum* × *drummondii*), *Stylosanthes* sp., pinto peanut (*Arachis pintoi*), sunflower (*Helianthus annuus*), *Urochloa spp*. (syn. *Brachiaria*), *Paspalum* sp., and Guinea grass (*Panicum maximum*) (Calegari et al. 2020). Mixing cover crops has also been encouraged to increase the biodiversity of production systems. Mixed cover crops include two, three, four, or more species; for example, black oat + vetch, radish + black oat + vetch, buckwheat + radish + pearl millet, *Crotalaria* sp. + pearl millet or *Cajanus cajan* + pearl millet + *Crotalaria* sp., or pearl millet + *Crotalaria* sp. + buckwheat. As soil and climate conditions vary, each cropping system should have a mixture of cover crop species that are more suitable for fulfilling multiple functions in the soil. This is an important subject and of great interest in interdisciplinary research.

After about 50 years of no-till in Brazil, this cultivation system appears to have changed the original concept of agriculture entirely, bringing a great deal of learning and countless benefits. The success of no-till is based on accumulation of organic matter and an increase in below- and aboveground biodiversity. This requires diversification of cropping systems, high input of crop residues, and no impediments to plant root growth along the soil profile. These are aspects of concern in current no-till conservation agriculture in Brazil. Because of the usual surface applications of lime and fertilizers in no-till, the improvement in soil fertility in this cropping system has been largely restricted to the topsoil. For that reason, research on soil management practices that may favor the growth and deeper penetration of root systems in the soil profile (such as the use of lime, gypsum, polyhalite, cover crops, animal waste, and biofertilizers capable of promoting plant growth through hormone synthesis) should be encouraged. Some crops with specific deep rooting characteristics can improve subsoil structure. Taproot systems such as alfalfa (*Medicago sativa*) have specific pore geometries associated with structure related air-filled porosity and biopore formation. Uteau et al. (2013) found that alfalfa generated a greater connected air-filled porosity due to more intense shrinking/swelling processes. Although Brazil produces only 40 thousand hectares of alfalfa, mainly in the southern region, there is great potential for the expansion of its cultivation in Brazil. The expansion of the area cultivated with alfalfa could help in the recovery of the soil structure following intensive crop production. Increasing root growth in the soil profile increases the carbon stock in deeper layers and helps curb greenhouse gas emissions. Cooper et al. (2021) observed that the larger carbon content within no-tilled aggregates translated into a greater total carbon content throughout the soil profile (0–50 cm). Differences in carbon stocks at greater depths have been explained by better root growth conditions under no-till (Galdos et al. 2019). Sisti et al. (2004) showed that increased carbon accumulation in no-till soil below 30 cm depth could be explained by greater root density when compared with conventional tillage. In addition, world commodity markets have caused an overemphasis on soybean growing in Brazil, putting crop diversification at a certain risk. Since this reality will likely not change, at least in the short-term, greater efforts are necessary to increase diversification and the input of cover crops right after soybean harvest. This is a greater concern in the tropical Cerrado region, where sufficient moisture is usually available only in the spring-summer season, from October to March (*Figure 7*). Some techniques of overseeding forage species, such as *Urochloa* (syn. *Brachiaria*) *ruziziensis*, U. *brizantha*, and *Panicum maximum* cv. Mombaça through broadcasting by airplane at the end of the soybean crop cycle (development stage R5.5), have shown excellent results in integrated crop-livestock production systems. In addition, a second crop of maize intercropped with these forage species following soybean has provided several benefits and should be encouraged. The main objective is to keep the system biologically active by constantly adding organic residues and increasing biodiversity.

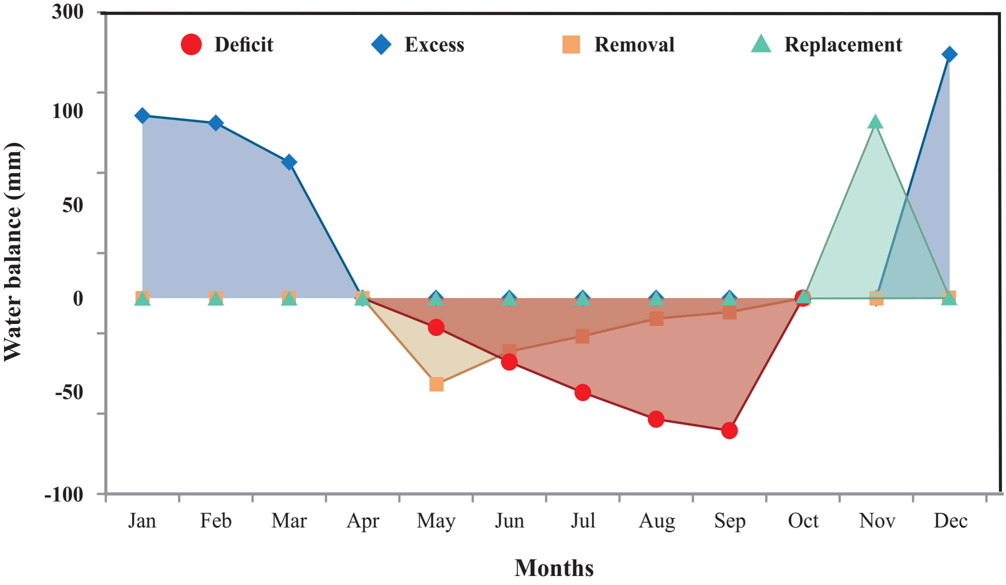


Figure 7. Extract of the normal climatological water balance for the period between 2004 and 2013, considering the soil water storage capacity of 100 mm at the Main Station of Embrapa Cerrados, Planaltina, DF, Brazil. Source: Silva et al. (2017).

Establishing integrated crop, pasture, forest, and livestock systems

Soils Brazilian institutions linked to the National Agricultural Research System (Embrapa) have long recommended systems based on no-till farming that integrate agriculture and livestock (Balbino et al. 2011). One of these systems is crop-livestock integration (CLI) and, more recently, the forest component was incorporated, creating the crop-livestock-forest integration system (CLFI).

Integrated agricultural production systems are sustainable strategies, designed to exploit synergies and intensify land productivity, combining annual agricultural, livestock, and/or forestry activities in different spatiotemporal arrangements. These systems contribute to changes in the physical, chemical, and biological properties of the soil (Vinhal-Freitas et al. 2010), promoting improvements in land quality (Lemaire et al. 2014). Therefore, they can help combat global food and energy insecurity and climate change in the coming decades (Moraes et al. 2014, Bieluczyk et al. 2020).

These systems began to be more widely used in the 1990s, and long-term experiments were developed with grain crops in association with pastures, especially in the South region of Brazil (Balbino et al. 2011) and then reproduced in other regions, with adaptation to environmental and cultural realities.

According to the FAO (2010), integrated production systems have the following benefits: i) improvement of production processes in the workforce, stability of economic factors, and risk reduction; ii) greater chances for producers to achieve their socio-cultural aspirations in an equitable way; and iii) greater food security to meet the needs of consumers regarding the quality of products and production processes.

According to Balbino et al. (2011), integrated production systems can be classified into four distinct modalities: (i) CLI or agropastoral, a production system that integrates agricultural and livestock components in rotation, consortium, or succession in the same area and in the same agricultural year or for multiple years; (ii) LFI or silvopastoral, a production system that integrates livestock (pasture and animal) and forestry components in consortium; (iii) CFI or silvoagricultural, a production system that integrates the agricultural and forestry components through the intercropping of tree species with agricultural crops (annual or perennial); and (iv) CLFI or agrosilvopastoral, a production system that integrates agricultural, livestock, and forestry components in rotation, consortium, or succession in the same area. The crop component can be restricted to the initial phase of implantation of the forest component or be part of the system for several years. Among these modalities, the one that has been most prominent in Brazil is the CLFI (Kichel et al. 2014). Some examples of integrated production systems are shown in *Figure 8*.


(a) (b)

Figure 8. Silvopastoral integration production system (a) and agrisilvicultural integration production system (b), Embrapa Pecuária Sudeste. Source: Wanderlei Bieluczyk.

Assessing the effect of the CLFI system in Cerrado areas, Zago et al. (2019) found that the system had a positive effect on microbial biomass and soil enzymes compared to pasture, reinforcing the importance of adopting more sustainable practices for soil health. In another study assessing the impacts of CLFI on soil microbiological indicators, Stieven et al. (2014) observed that, in general, the CLFI systems had positive impacts on soil microbiota.

Other authors, such as Assis et al. (2015), studied changes in the physical properties of the soil due to implementation of CLFI systems and found that such systems led to improvement in soil physical quality, identified by the improvement of the following attributes: hydraulic conductivity, microporosity, total porosity and bulk density, in relation to degraded pasture.

Studying the effect of nominal extensive grazing to CLI, and CLI to CLFI, system conversion on the quantity, quality, and origin of soil organic matter in southeastern Brazil in a six-year experiment, Bieluczyk et al. (2020) found that all the systems increased soil carbon and nitrogen stocks throughout time. In this study, CLI and CLFI systems enhanced 21% and 17% of soil carbon from 2010 to 2016. However, when comparing soil C accumulation tendencies in time (estimated by four points linear regression), converting extensive low-grazing intensity pasture to ICL was the most promising strategy, increasing soil C stocks at the rate of 0.28 Mg C ha^{-1} yr^{-1}. Further intensification, from CLI to CLFI reduced -0.22 Mg C ha^{-1} yr^{-1}, bringing no further benefits on soil organic matter accrual.

Agroforestry systems

Agroforestry systems (AFSs) are based on ecological succession and are analogous to natural ecosystems. Exotic or native trees are intercropped with agricultural crops, vines, forages, shrubs, etc. according to a pre-established spatial and temporal arrangement, with a high diversity of species and interactions among them. These systems are based on ecological, economic, and social interactions that exist in a production system, playing multiple roles such as promoting the recovery of degraded areas, ecosystem services and increased biodiversity, among others. In the Amazon, AFSs, called "terreiros", with a great diversity of native fruit and medicinal species and multiple uses, are commonly found close to rural indigenous communities, conserving biodiversity, including non-conventional plant species and increased soil organic matter (Noda et al. 2008).

AFSs have been considered more sustainable agricultural systems than traditional forms of agriculture and forestry. They can function as a viable option for the recovery of degraded areas, management of forest fragments, forest restoration in areas of permanent preservation and legal reserves and in the establishment of small plots with tree species for the purpose of producing wood for the producer's own consumption or as a source of food items. They provide various ecosystem services, such as carbon sequestration, biodiversity conservation, maintenance and/or improvement of soil fertility, prevention and reduction of erosion processes, improvement of water infiltration, wind break protection, pest control, and pollination enhancement. The great diversity of plant species creates favorable conditions for establishing ecological and environmental functions on the property, allowing for better stability of laborers in the field and greater income security for the producer, due to product diversification (Chang and Cheng, 2020). These systems have been identified as an alternative for more sustainable development of agriculture, especially for developing countries (Matthews et al. 2014, Nair et al. 2010).

In Brazil, regional diagnostics and research results have shown that AFSs are highly applicable in areas with agricultural and livestock activities. The main projects have been associated with cacao (*Theobroma cacao*), rubber (*Hevea brasiliensis*), oil palm (*Elaeis guineenses*), and coffee (*Coffea canephora* and *Coffea arabica*) crops (Alvim and Nair, 1986; Brienza Junior and GazelYared, 1991; (Souza et al. 2012, Gama-Rodrigues et al. 2010, Jagoret et al. 2014, Monroe et al. 2016, Ramos et al. 2018). In addition, other studies have shown that AFSs have good responses when used in areas where there are problems in different ecosystems resulting from deforestation and degradation.

This pattern was observed by Fávero et al. (2008), evaluating the recovery of degraded areas through an agroforestry system in the Vale do Rio Doce, Minas Gerais.

In general, AFSs can be classified into (i) agrisilvicultural systems that involve agricultural crops and trees, including shrubs and/or vines; (ii) silvopastoral systems that refer to the association of pastures and/or animals and trees; and (iii) agrosilvopastoral systems that combine agricultural crops, pastures and/or animals, and trees. In any of these systems, integration of the components can take place simultaneously or sequentially and in an infinity of possible combinations (https://www.embrapa.br/en/codigo-florestal/sistemas-agroflorestais-safs).

Through integration of trees in agricultural systems, production is diversified and sustained, promoting an increase in social, economic, and environmental benefits for land users at all levels. In particular, agroforestry systems are crucial for small farmers and other rural populations as they can promote an increase in the food supply, income, and health (http://www.fao.org/forestry/agroforestry/80338/en/). Some examples of AFSs in different regions of Brazil are shown in *Figure 9*.

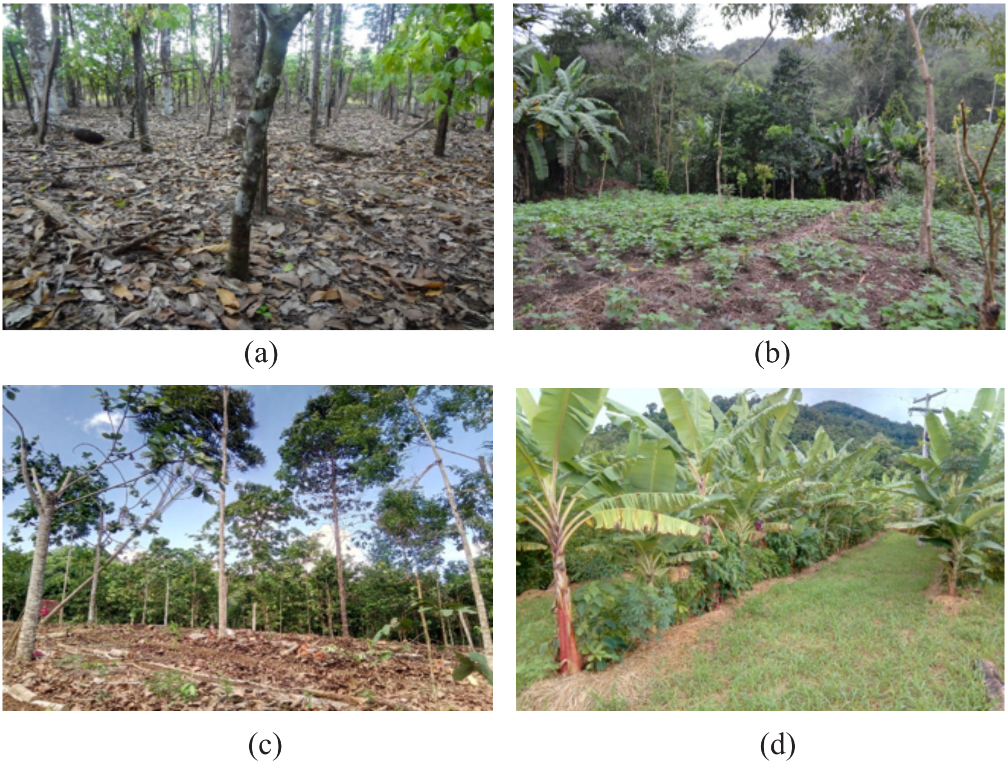


Figure 9. Agroforestry Systems composted of African mahogany and cacao, Tomé-Açu, PA, Brazil (a), source: Prof. Alberto Bentes Brasil Neto; AFS at the Universidade Federal de Santa Maria, Santa Maria, RS, Brazil (b), source: Prof. Ricardo Bergamo Schenato; Renovation of AFSs Jaguaquara, BA, Brazil (c), source: Prof. André Mundstock Xavier de Carvalho; and AFS including banana and cassava, Jaguaquara, BA, Brazil (d), photo: Prof. André Mundstock Xavier de Carvalho.

Assessing soil organic carbon storage in cacao (*Theobroma cacao* L.) AFSs in Bahia, Brazil, Gama-Rodrigues et al. (2010) highlighted the role of this system in mitigating the emission of greenhouse gases (GHG) through the accumulation and retention of large amounts of organic carbon in soils, demonstrating the benefit of the environmental services promoted by this system.

Monroe et al. (2016) quantified soil organic carbon storage and assessed its origin as an indicator of the relative contribution of trees (C3 plants) and grasses (C4 plants) four years after converting pasture to AFSs in southern Bahia. They observed that AFSs using cocoa and rubber were the most efficient systems in accumulating carbon in the first 20 cm of soil, and that AFS with cocoa was more efficient than with rubber in accumulating carbon derived from C3 plants. The C3-origin SOC decreased according to the following sequence: cacao row > natural forest > rubber row > pasture. In addition, the C4-origin SOC decreased according to the following sequence: pasture > rubber row > cacao row. The C3-origin C in the rubber row increased by 59.9 Mg ha^{-1}, and the C3-origin C in the cacao row increased by 81.7 Mg ha^{-1}at a depth of 0–20 cm after 4 years of establishment. The C4–C3 replacement rate in the rubber and cacao rows were 5.25 Mg ha^{-1}year^{-1} and 8.60 Mg ha^{-1}year^{-1}, respectively.

In eastern Amazonia Ramos et al. (2018), quantified C stocks in a palm oil (*Elaeis guianensis* Jacq.) AFS and a palm oil AFSs associated with cacao (*Theobroma cacao* L) in four distinct compartments: aboveground live biomass, plant litter, roots, and soil. The carbon stock until 30 cm depth was higher in the palm oil AFS with cacao (116.7 ± 1.5 Mg C ha^{-1}) compared to the AFS with palm oil only (99.1 ± 3.1 Mg C ha^{-1}). The same pattern was observed for the total plant litter stock, which was higher in the palm oil AFS with cocoa (3.27 ± 0.01 Mg C ha^{-1}) than in the palm oil AFS (2.26 ± 0.06 Mg C ha^{-1}). For the total stocks of root and soil carbon (0-30 cm), there were no significant differences between the AFSs. Carbon stocks varied between AFSs due to differences in above- and belowground stocks. In general, C stocks below ground varied spatially in response to management in the oil palm and non-palm areas. The results obtained have important implications for monitoring carbon dynamics at the ecosystem level and the refinement of soil management

Martinelli et al. (2019) studied aspects related to the mitigation of global warming and the provision of ecosystem services for small farmers in the Cerrado biome and found that AFSs have significant carbon sequestering capacity, represented by the negative values of global warming potential, ranging from (−263) to (−496) t CO_2 equivalent per hectare. In addition, it was noted that a large number of fruit trees allowed farmers to consume and sell a wide variety of products. Through adoption of the system, families also benefited from improvement of microclimate conditions and the aesthetic effects provided by the AFSs.

Plant biodiversity and its implications for soil quality

Brazil is well known for the megadiversity of its six biomes, consisting of vegetation with very heterogeneous characteristics (IBGE, 2019). This megadiversity is a consequence of the vast area of Brazil of mainly tropical climate, but also includes subtropical and temperate climates and a wide diversity of soils. For instance, at least 2.000 native Leguminosae species is estimated to occur in Brazil (Moreira, 2006). This figure represents 10% of all Leguminosae species throughout the world. Plant diversity also extends to edible plants, with some species typical of the cuisines practiced in each region of the country and generally grown by small farmers who maintain the diversity (many varieties) of this priceless germplasm. Some examples are *Euterpe* spp. (açaí), *Theobroma grandiflorum* (cupuaçu), and *Solanum sessiliflorum* (cubiu) in Northern Brazil; *Caryocar brasiliense* (pequi) in the Central-West; *Araucaria angustifolia* (araucaria) in southern Brazil; and even unconventional plants such as *Pereskia aculeata* (ora-pro-nobis), very popular in Minas Gerais in the Southeast of Brazil. *Vigna unguiculata* (cowpea) and *Phaseolus lunatus* (lima bean) are typically cultivated by small farmers, who preserve many subspecies and varieties in the North and Northeast of Brazil, where they are predominantly grown.

Considering epigeic and anecic organisms, such as some earthworm species, are known as components of soil biodiversity, it is logical to also consider plants as soil organisms, even more so because the root system of many species is much larger in volume and biomass than their shoots. Diversity in plant species that inhabit the soil provides for diversity of carbon substrates, due both to decomposition of deposited organic matter (plant residues) and the effect of plant roots (rhizosphere effect), which exude a great diversity and quantity of organic compounds that nourish soil organisms at different levels of the food chain under different vegetation covers (Berendsen et al. 2012, Kou et al. 2018). Thus, these carbon substrates allow the activity of all soil organisms to act as a driving force for biophysical (e.g. improvement of aggregation of soil particles by exopolysaccharides produced by fungi and bacteria as well as by fungal hyphae, improvement of water and air movements in tunnels, galleries and other structures made by soil fauna) and biochemical processes (e.g. nitrogen fixation, inorganic phosphate solubilization, organic matter decomposition and mineralization) that ensure the sustainability of ecosystems. As the main primary producers, plant species are the basis of food web and allow other groups of organisms (decomposers, litter and soil organic matter feeders, mutualists, pathogens and parasites, shredders, grazers and predators) to survive and provide ecosystems services. For instance, Braga et al. (2012) showed that land-use intensification (forest to pastures) in western Amazonia results in a reduction of the richness, abundance and biomass of dung beetles, and this in turn results in lower rates of dung removal as well as higher number of flies in the most simplified systems (pastures). Moreover, maintaining and enhancing biodiversity is of utmost importance to provide genetic resources for a great diversity of purposes, including agricultural management. Therefore, plant diversity is fundamental for soil quality.

The various production systems in constant expansion in Brazilian agriculture, such as no-till planting, crop-livestock integration, crop-livestock-forest integration, and agroforestry, consider the diversity of plant species and the conservation of plant residues on the ground (see 3.2 and 3.3). The use of trees in pasture also provides for animal welfare. The expansion of these types of management will certainly require species that contribute to food security, and their growth will be stimulated by biological processes.

Contribution of soil biodiversity to the sustainability of cropping systems in Brazilian soils and challenges in increasing utilization

Awareness of the importance of soils and their biodiversities have increased exponentially in recent years (FAO et al. 2020, Orgiazzi et al. 2016). Likewise, the development of reliable and accurate techniques for assessing and understanding biodiversity, especially microbial diversity, has increased exponentially. However, management of this biodiversity for sustainable agriculture and food security has not been developed in the same way. Development of molecular techniques has allowed evaluation of most microbial soil communities by culture-independent techniques, i.e., without cultivation and multiplication in synthetic media, and has revealed a huge diversity of both culturable and unculturable bacteria and fungi, as well as the effects of diverse soil management strategies on this biodiversity (Hug et al. 2016, Jesus et al. 2009, Carvalho et al. 2016, Lucheta et al. 2017, Damian et al. 2021) . However, only culture-dependent techniques can provide the genetic resources that will be selected for use in sustainable management in agriculture, environmental quality, and food safety/security, and for other purposes.

The 2030 Agenda for Sustainable Development was adopted by UN Member States in September 2015. Soil biodiversity has a strong relationship to most of the 17 goals included in the Agenda. The soil processes of nutrient cycling, organic matter decomposition, soil organic matter dynamics, soil structure maintenance, and biological control are carried out by a vast array of organisms and directly linked to the provision of food, fiber, and fuel, carbon sequestration, water purification, and reduction in soil contamination.

Sustainability is based on increasing the contribution of soil biological processes in agricultural and forest systems, reducing the use of fertilizers and agrochemicals and, hence, their effects on greenhouse gas emissions, and increasing food safety/security and soil quality. Brazil has good practices in this direction described below.

In 2020, the Brazilian Ministry of Agriculture (Ministério da Agricultura, Pecuária e Abastecimento - MAPA) launched the National ¨Bio-inputs¨ Program (Programa Nacional de Bioinsumos), which aims to meet growing demand from the productive sector and society that seek alternatives for bio-based inputs in agricultural systems and increasingly sustainable products. Therefore, the general objective of the Program is to expand and strengthen the use of bio-inputs to promote the sustainable development of Brazilian agriculture and improve the quality and reliability of bio-inputs, meeting national and international market requirements. Bio-inputs are produced by dozens of factories dedicated to products for biological control and inoculants to promote plant growth, a growing market both in Brazil and in other countries that are increasingly concerned with food safety and environmental quality.

Biological pesticides

According to the Brazilian Association of Biological Control Companies (Associação Brasileira das Empresas de Controle Biológico), biological pesticides are already used on more than 23 Mha in Brazil, with growth estimates of up to 20% per year. The most expressive example is in sugarcane (*Saccharum officinarum*), where 5.2 million of hectares are treated with natural enemies of pests. *Cotesia flavipes* and *Trichogramma galloi* are used in the cultivation of sugarcane to control the sugarcane borer (*Diatraea saccharalis*). To control leafhoppers, *Mahanarva posticata* and *M. fimbriolata*, products formulated with the fungus *Metarhizium anisopliae* are applied. To control sugarcane weevils, the fungi *Beauveria bassiana* or *Metarhizium anisopliae* can be used as microbial biopesticides. Several commercial products are also available and registered in MAPA for applying at about 20 millions of hectares in many crops with *Bacillus thuringiensis*, *Beauveria bassiana*, *Metarhizium anisopliae*, *Baculovirus anticarsia*, and *Trichoderma harzianum*, among others, for controlling pests, diseases, and nematodes in various crops (Bettiol, 2011; Parra, 2014; Bueno et al. 2020).

Legume inoculants in Brazil: advances and challenges

A good example of soil process that can be managed to improve plant growth is symbiotic nitrogen fixation in legume species. Leguminosae is a diverse family with about 20,000 species worldwide. Leguminosae species predominate in most Brazilian biomes. In addition to the economic importance of these species, most of them are able to establish symbiotic relationships with N_2-fixing bacteria, commonly named “rhizobia”. Estimations are of at least 2,000 species in Brazil, most of them able to form symbiotic relationships with rhizobia (Moreira, 2006).

The most significant example of successful application of a biological process in Brazil and other Latin America countries, specifically Argentina and Uruguay, as well as United States, is the inoculation of selected *Bradyrhizobium* bacterial strains in the exotic species soybean, as a result of efforts made by Brazilian scientists since the 1960s (Franco, 2009). Application of soybean inoculants is increasing (*Figure 10*), reaching a total of about 64 million doses in 2018/2019 applied on an area of about 36 Mha, yielding 120 million tons of grain throughout Brazil (Conab, 2021). Inoculation of soybeans with selected *Bradyrhizobium* strains (approved by MAPA) (*Table 1*) totally replaces mineral N fertilizers, leading to savings of about 5 billion dollars this year (https://globalfert.com.br/outlook-globalfert-2020/). The N_2 fixed by Brazilian soybean in 2018/19 saved about 19 million tons of urea, which would represent about 130% of all N fertilizers consumed in Brazil, 88% of which were imported.

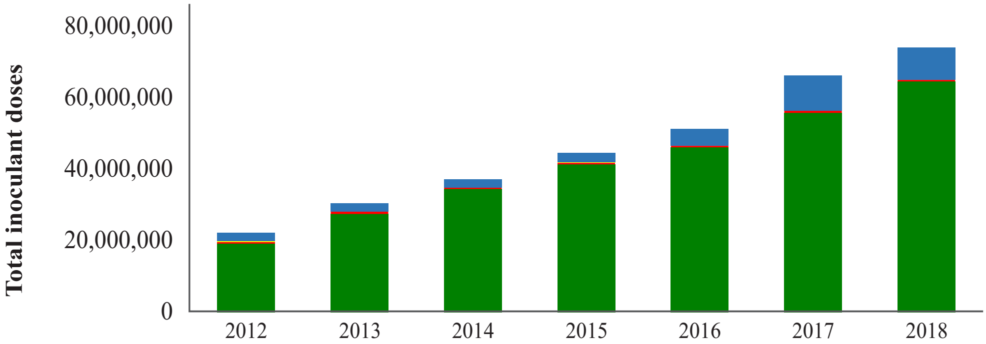
Table 1. Strains approved by the Minister of Agriculture, Supply and Livestock for some legume species

Legume species	Strains : SEMIA CODE (original code)	Identification
Glycine max (Soybean)	5019 (29 W),587, 5079 (CPAC15), 5080 (CPAC 7)	*Bradyrhizobium elkanii* (5079 & 587), *B. japonicum* and *B. diazoeficiens*
Phaseolus vulgaris (common-bean)	4077 (CIAT 899), 4088 (H 12), 4080 (PRF 81)	*Rhizobium tropici**, *R. Freirei**
Viga unguiculata (cowpea)	6461 (UFLA 03-84), 6463 (INPA 03-11B), 6462 (BR3267), 6464 (BR3262)	*Bradyrhizobium viridifuturi* symbiovar tropici*, *B.amazonense**,** *Bradyrhizobium yuanmingense,* *Bradyrhizobium pachyrhizi*
Acacia saligna *Chamaecrista ensiformis*	BR3804	*Mesorhizobium plurifarium**
Sesbania virgata	6402 (BR5404)	*Azorhizobium doebereinerae**
Stvlosanthes spp.	6154 (BR446)	*B. stylosanthis**
Centrosema spp.	690 (C100a)	*B. viridifuturi**
Neonatonia wightii (=Glycine wightii)	6149 (SMS 303)	*B. tropiciagri**
Desmodium ovalifolium (*=D. heterocarpon*)	6208 (CIAT 2372)	*B. embrapense*

*** New species, which description is based on isolates from Brazilian soils**
**** New species description submitted**

In addition to this enormous economic advantage, biological N_2 fixation from the air through *Bradyrhizobium* is a clean biotechnology, avoiding the lixiviation and volatilization of N compounds due to low N-fertilizer use-efficiency by plants. Unfortunately, soybean inoculation is currently the only success, because most inoculants produced by companies in Brazil are for soybean (*Figure 10*).

A decade ago, use of N_2-fixing bacterial strains in agriculture was almost exclusively restricted to soybean because, for a long time, it was believed that important crops such as cowpea (*Vigna unguiculata*) and common bean (*Phaseolus vulgaris*), mainly grown by small and medium farmers, would not respond to inoculation, due to their promiscuity with inefficient native rhizobia strains belonging to many species and genera. However, in recent years, use of inoculants containing efficient N_2-fixing bacterial strains has increased in these species. Although potential application is high, they have not benefitted as soybean has, even though Brazilian research institutions have strains selected for these crops and for about 100 other legume species (for forest, green manure, and forage species) (MAPA, 2011).



	2012	2013	2014	2015	2016	2017	2018
🟦 Others	2,575,227	2,148,788	2,254,986	2,874,619	4,597,753	9,710,256	9,169,169
🟨 Cowpea	180,914	239,260	46,524	96,259	67,624	94,730	66,288
🟥 Common-beans	270,448	477,848	339,340	403,820	377,563	658,603	279,948
🟩 Soybean	19,174,524	27,468,482	34,197,621	41,056,859	45,801780	55,215,175	63,945,667

Figure 10. Total inoculant doses sold according to crop from 2012 to 2018 (source: Associação Nacional de Produtores e Importadores de Inoculantes: http://www.anpii.org.br/)

Reasons for low adoption of N_2-fixing inoculants in other crop species compared to soybean are that soybean occupies large areas of Brazilian farmland and demands large amounts of inoculants, which is attractive to companies that produce them. These large farmers generally have consultants aware of the benefits of this biotechnology. Small and medium-sized farmers, who produce most of the food to supply the Brazilian population, do not know that this biotechnology exists. For the companies that produce these inoculants, it is more profitable to sell large amounts (wholesale) to large farmers than small amounts (retail) to small farmers. This is a typical example of the law of supply and demand, associated with lack of knowledge. Thus, effective communication channels must be established between scientists and social stakeholders directly linked with soil use and conservation. Biodiversity is of utmost importance to improve the contribution of symbiotic N_2 fixation in legume crops. Even soybean inoculation is being threatened by pressure from chemical fertilizer companies selling NPK formulas containing increased amounts of mineral N, which could lead to inhibition of biological N_2 fixation. Another threat is ¨on farm¨ production of inoculants produced without quality control, compromising a relatively well-established market of the certified inoculants. Nevertheless, the on-farm production of good quality inoculants and biopesticides may favor the expansion of the use of these bio-inputs to other species besides soybeans.

Multifunctional inoculants and co-inoculation of beneficial microbes

Researchers continue to select efficient N_2-fixing strains for important crops. Native soil communities occurring in Brazilian biomes such as the Amazon, Cerrado, and Atlantic Forest have been the sources of these genetic resources (Costa et al. 2020, Oliveira et al. 2020). In these studies, in recent years, researchers have considered rhizobia species as generally multifunctional.

In addition to biological N_2 fixation, they are able to perform diverse plant-growth-promoting processes, such as phosphate solubilization, phytohormone and siderophore production, and biological control of pests and diseases. Multifunctionality of plant-growth-promoting traits is also widespread in non-rhizobia plant-growth-promoting rhizobacteria (Oliveira-Longatti et al. 2014). Multifunctionality and co-inoculation of beneficial microbes represent potential windows of opportunity to explore for improving the contribution of biological processes in agroecosystems, as well as for increasing their use in other legume and non-legume species. Co-inoculation with *Azospirillum*, a non-symbiotic genus, is an increasing market for soybean, but also for inoculation in grasses such as maize, wheat, rice, and pasture species (*Figure 10*). The market for mycorrhizal fungi represents another window of opportunity, though not yet economically important and efficient, due to the widespread natural occurrence of these organisms and the lack of host specificity. However, stimulants of mycorrhization, such as formononetin, have proven to be a promising tool, especially in the event of low phosphorus inputs in soybean and maize (Ribeiro et al. 2016, Santos et al. 2020).

Concluding remarks

The immense biodiversity extant at the various types of Brazilian soils and climate conditions is a valuable source of genetic resources, with biotechnological potential that should also be considered in soil conservation programs. Brazilian diversity of many functional groups of soil organisms are still underestimated due to many reasons such as: the large size of the country territory with majority of areas with natural biomes with difficult access, low number of taxonomists, most surveys carried out in areas near research institutions. Despite of that, figures of native species already obtained are amazing: termites-300 (Constantino and Acioli, 2006), earthworms-253 (James and Brown, 2006) and dung beetles-726 (Vaz-de-Mello, 2021). Brazil also harbors 50% of all species of the world of the ubiquitous arbuscular mycorrizal fungi (Stürmer and Siqueira, 2006).

In Brazil, soil management systems under conservation agriculture have been evolving to increase plant diversity, and biotechnologies are already used on a large scale, especially for commodity crops such as soybean, sugarcane, and maize. However, the dissemination of biotechnologies, such as rhizobial inoculants, whose availability is still unknown to the vast majority of small and medium-sized farmers, who produce and conserve most edible species consumed by the Brazilian population, should be encouraged. Technologies that increase the growth and deeper penetration of plant roots in different crop systems should also be encouraged. An increase in the contribution of soil biological processes will be possible through more studies that reveal biodiversity and its functions and applications, as well as by increasing awareness of the role of biodiversity among the various social segments. Extension services and practices that popularize science should be increased to achieve this awareness. Knowledge of biodiversity is a strategic necessity from a political, economic, social, and environmental perspective. Effective communication between different sectors of society, government, and the scientific community must be established.

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Chapter 9

Soil management alters soil organic matter content affecting soil properties and agricultural sustainability in the Chilean Mediterranean environment

Edmundo Acevedo H. and Eduardo Martínez E.

Introduction

Agricultural intensification has retrograded the soil decreasing the natural sinks for CO_2, accelerating soil erosion, raising the greenhouse gas emission contributing to global warming, threatening biodiversity, and contaminating waters. The developments in agriculture require increased use of fuels, further contributing to the emission of greenhouse gases and to climate change. The agricultural model based on mechanical soil tillage, exposed soils and continued mono-cropping is accompanied by negative effects on agriculture's natural resource base to such an extent that future agricultural productive potential is jeopardized (Corsi et al. 2012, Corsi and Muminjanov, 2019). In agricultural systems, maintaining the carbon levels in the soil through an adequate agronomy is essential to their sustainability (Martínez et al. 2008a, 2008b). The carbon levels are affected by erosion, tillage and land use change, which finally affect crop yields and soil quality (Derpsch et al. 2000). Organic carbon affects soil physical properties such as aggregate formation, its structural stability, and chemical and biological properties of soils (Six et al. 2000). Drylands are particularly valuable for carbon storage due to their high degree of permanence, concerning the duration that carbon is stored in the soil, compared to humid areas (Laban et al. 2018). They make an important contribution to stocks of soil biodiversity and soil organic carbon, contributing to climate change mitigation.

In this chapter we discuss agricultural soil management in the Chilean Mediterranean environment and its interactions with soil properties. We rely in our experience in Central Chilean Mediterranean environment. The aim of this review is to highlight the relevance to maintain or increase the soil organic matter (SOC) as a sustainable agronomic practice, with emphasis on Mediterranean environments.

Mediterranean environments

The Mediterranean environment includes all the countries around the Mediterranean Sea, Northwest Mexico, California in the US, Central Chile, South-West Australia, and the Cabo region in South Africa (Boydak and Doğru, 1997; UNESCO-FAO, 1962; Di Castri, 1973). The Mediterranean climate is characterized by cool rainy winters and hot, dry summers with high solar radiation, high evaporation and a moderate influence of marine air throughout the year (Leisz, 1982; Seager et al. 2019) (*Figure 1*). It corresponds to Köppen´s (1923) olive climate, due to the cropping of olives in this environment. Most agricultural land in Chile is within this eco-region. Agriculturally, the Mediterranean environments are mostly affected by intensive tillage, overgrazing and the use of fire (Naveh and Dan, 1973, Boydak and Doğru, 1997, Derpsh et al. 2000).

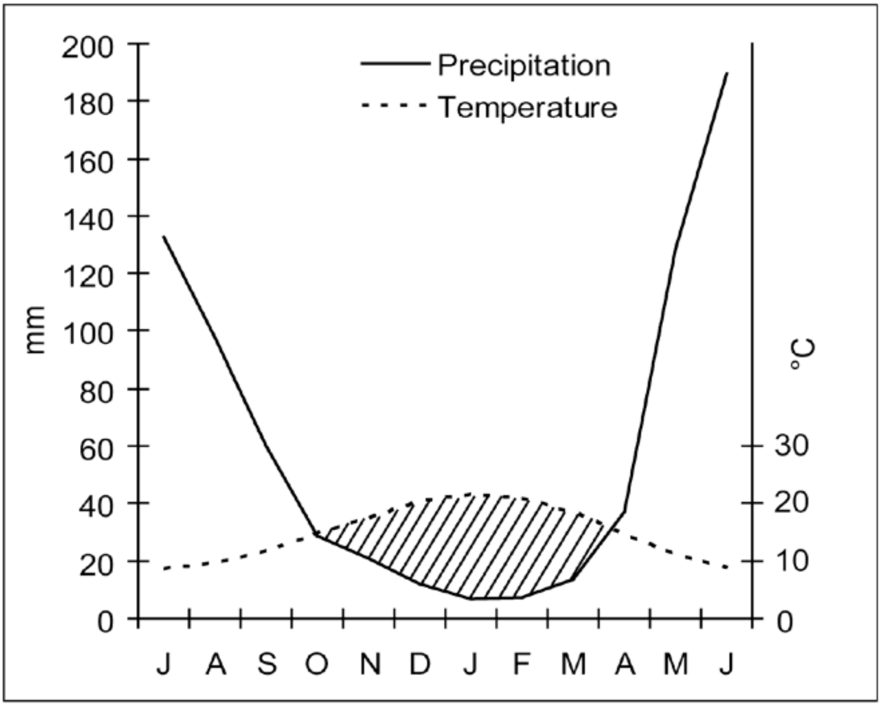

Figure 1. Climate Diagram for Talca, Chile (35°26’ S., 71°26’ W., 110 m.a.s.l).

Soil properties: organic carbon as a key element

Soil organic carbon (SOC) is the main element in the soil organic matter (SOM) and contains all the elements coming from the decomposition of organic residues, and metabolites from plants (rhizodeposition), animals and soil microbes (Laban et al. 2018). The soil organic carbon is related to the sustainability of the agricultural systems affecting the soil properties linked to crop yield (Bauer and Black, 1994; Carter, 2002, Wander et al. 2002, Acevedo and Martínez, 2003; Lal, 2010). However, establishing a clear direct relationship of dependence between SOC and soil productivity is complex (Govers et al. 2013) and commonly results from long term experiments.

Soil properties affecting productivity in terms of grain or biomass products are linked by SOC (Page et al. 2020). Soil organic matter affects the amount and availability of soil nutrients, contributing elements as N, usually deficient. In addition, SOM modifies soil acidity and the alkalinity towards neutrality, and increases the solubility of several nutrients, it provides colloids having high cation exchange capacity. Its effect on the soil physical properties is reflected in soil structure, pore size distribution, hydraulic, pneumatic and thermal conductivity and soil strength. SOC concentration depends on the local environmental conditions, but it is also affected strongly by the soil management practices.

Physical properties affected by Soil Organic Carbon

Soil aggregation.

Soil organic carbon has an important effect on the aggregation of soil particles (Blanco-Canqui and Ruis, 2018, Li et al. 2019), with a relationship between aggregate size and SOC content (Rothon, 2000, Martínez et al. 2008a) (*Figure 2*). As pointed earlier by Buyanovsky et al. (1994), the higher the labile SOC content, the larger the aggregate size (*Figure 2*). Nevertheless, the process of aggregate formation and aggregate strength is more complex, depending on several factors, including swelling and shrinkage processes, the biological activity and organic matter exudates playing a crucial role (Horn and Smucker, 2005). From the point of view of agricultural soil management practices, the conservation, and gains in SOC is crucial to maintain or enhance the stability of aggregates to avoid soil losses by erosion and surface seals.

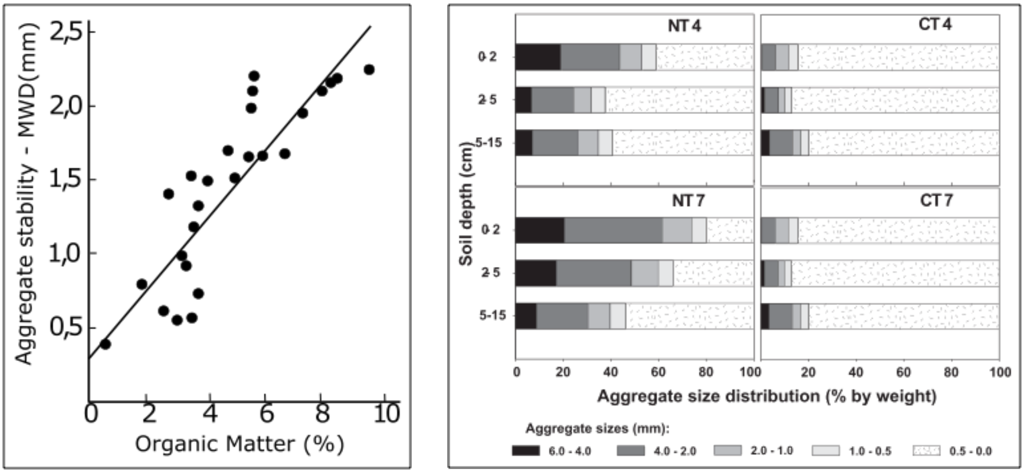

Figure 2. a) Relationship between aggregate stability (MWD) and soil organic matter (%SOM) for 26 British soils. (Adapted from Chaney and Swift, 1984) and b) aggregate size distribution after 4 and 7 years of No Till (NT) and continuous tillage (CT) (Martínez et al. 2008a).

Soil porosity and water retention.

The arrangement of aggregates in the soil matrix affects the porosity, water retention and the air capacity of soil (Acevedo and Martínez, 2003; Brunel et al. 2013, Obour et al. 2018, Neira et al. 2015, 2020). Soil porosity also affects the growth of plant roots. Porosity and water retention are two parameters closely linked to the number of pores, pore-size distribution, and the specific surface area of soils. SOM generally has a positive effect on the water holding capacity of the soil (Krull et al. 2004). Pikul and Allmaras (1986) studied the distribution of the pore space with different soil management and found that adding organic matter increased the macro pores, which retain water at lower energy.

The increase in SOC tends to decrease the soil bulk density (ρ_b) (Reddy, 1991; Krull et al. 2004), however, when the soil is managed without plowing as in No Tillage, even though there is an increase in SOC, there are other factors such as the weight of the planting machinery and the level of traffic affecting the final value of ρ_b which is dependent on the internal soil strength or the precompression stress (Dörner et al. 2013). Thus, soil ρ_b in No Tillage (NT) may increase (Apezteguía and Sereno, 2002; Lampurlanés and Cantero-Martínez, 2003), decrease (Dao, 1996) or have no change over time (Valle et al. 2004). Soil Organic Carbon decreases the specific gravity of solids (Huat et al. 2009) down to 1,4 g/cm^3 for peat.

Soil compaction and SOC.

Soil compaction consists in a reduction of the soil pore space caused by a force (weight) applied on top of the soil (Kulli, 2009). It affects the soil penetration resistance which is directly related to plant growth and development (Hazma and Anderson, 2005; Dexter et al. 2007), soil organic matter and soil cementing agents (Aggarwal et al. 2006). Soil strength, or mechanical resistance to failure represent the reaction of a soil to an applied force and has been widely used to estimate the soil compaction level. Compaction of the soil imply an increased soil strength and increased penetration resistance for the roots (Wästerlund, 2020). Soil strength is related to texture, structure, water content, land use, management history and the presence of soil organic matter (Payahuala and Dörner, 2009; Horn et al. 2019).

At all compaction levels, penetration resistance increases with decreasing soil water content (Kondo and Dias Junior, 1999; Lipiec et al. 2002, Aggarwal et al. 2006) and increases with increasing bulk density (Aggarwal et al. 2006). Some soil management practices such as subsoiling, bed cropping, and organic matter incorporation into the soil may decrease the soil penetration resistance significantly (Reddy, 1991; Aggarwal and Goswami, 2003).

The higher the organic carbon content at a given bulk density and comparable texture the higher is the mechanical strength defined as precompression stress (Horn 1981, Hartge and Horn, 2016).

Hazma and Anderson (2005) identified various effects of SOM on soil structure and soil compaction, including flocculation of mineral particles and changes in the mechanical resistance of the soil aggregates. The effects on soil compaction depend on the type of organic matter (Ekwue, 1990), the easily oxidizable organic matter having a greater effect on the mechanical behavior of the soil than total SOM (Ball et al. 2000). Soil compaction resistance is also affected by variations in the C/N ratio of SOM, type of soil and environmental conditions such as temperature and humidity (Hazma and Anderson, 2005).

As indicated, SOM increases aggregate size and stability and hence soil macroporosity, decreasing the soil penetration resistance. No till (NT) is an agronomic/agricultural practice that leaves organic residues on top of the soil; it uses high weight planting machinery; if this machine enters the field in a humid soil state, the soil macroporosity decreases (Kulli, 2009), increasing the soil penetration resistance although SOM has a buffering effect on the soil compacting forces.

Soil water infiltration.

SOM increases the water infiltration rate, but water infiltration depends on soil management and particularly on compacted soil layers that may be present in the soil profile. In a sandy loam soil, 10 Mg ha^{-1} of green manure increased the water infiltration rate (Reddy, 1991). Pikul and Allmaras (1986) in a 55-year experiment creating the conditions for higher SOM accumulation found an increased saturated soil water conductivity. Fuentes et al. (2004) found higher values of near saturated hydraulic conductivity in an Ultic Haploxeroll having natural pasture, 3.71% SOC in the 0-5 cm soil depth in No till vs. 1.82 % SOC in conventional tillage (using seed bed preparation). Aldea et al. (2008) found a decrease in water infiltration rate in an Entic Haploxeroll managed with no till for 5 years that could be reversed by chiseling, however, the soil decreased its water infiltration rate within the cropping season in which chiseling had been practiced. Blanco-Canqui et al. (2004) studied the effect of the soil tillage system, crop and traffic on the soil physical properties of a sandy loam Epiaqualf and did not find correlation between the SOM and the saturated hydraulic conductivity.

Soil chemical properties

SOM tends to move the soil pH towards neutrality. Organic residues are decomposed generating carboxylic and phenolic groups which behave as weak acids (Adeleke et al. 2017). If soil aeration decreases by compaction, CO_2 from the soil atmosphere increases and combines with water, generating carbonic acid which liberates H^+ acidifying the soil. If the soil is moderately acid, a decreasing soil pH may affect the availability of some nutrients and increase some toxic elements such as Al^{+3}. On the other hand, organic residues may increase soil pH liberating exchange bases from the residues (Pocknee and Sumner, 1997; Wong et al. 2000; Narambuye and Haynes, 2006). Nitrogen mineralization from plant residues increase the soil pH due to the formation of NH_4^+ which consumes protons.

The cation exchange capacity (CEC) of the soil increases with NT due to an increase in organic colloids. The CEC of the soil varies between 2 and 150 cmol (+) kg^{-1} soil according to the predominant clay type. The organic colloids depend on soil management and may have a CEC around 200 cmol (+) kg^{-1} soil or higher.

In a long-term irrigated experiment located in the arid Central Zone of Chile, Martínez et al. (2013) found that No Tillage (NT) promoted a more efficient use of the SOC generating soil C accumulation in time. Soil chemical fertility increased under NT, with higher levels of N, P, and K and a slight decrease of soil alkalinity, with major changes being observed in the top 5 cm of the soil. These chemical changes were attributed to the maintenance of crop residues on the soil surface, which contributed to a slow release of nutrients throughout mineralization and to the development of a better environment for microbial activity.

Organic carbon and soil biological properties

The SOC is essential for the soil biological activity. It provides energy resources to the mainly heterotrophic soil organisms (SO) in the form of labile C (carbohydrates, or low molecular weight organic compounds) (Borie et al. 1999). The SO decompose the soil organic residues and participate in the cycling of many elements used by plants. Furthermore, SO participate in the formation and stabilization of soil structure and porosity (Krull et al. 2004). SOM decomposition occurs in three phases: *1)* fragmentation and mixing with the mineral soil with the participation of the macro and megafauna (2-20 mm), *2)* rupture of big molecules by enzymes produced by fungi and bacteria and, *3)* assimilation and transformation of soluble products from phase 2 by soil microorganisms (microflora and micro fauna < 100µm) (Paul et al. 1999). The secondary products of the metabolism of SO and the rupture of the big molecules are accumulated in a complex colloidal substance (humus). The final products of decomposition of the organic residues and humus are energy, water and minerals, mainly CO_2 (Ontl and Schulte, 2012). If oxygen is not limiting, SOM decomposition is the product of respiration, if not it is the product of fermentation (Singer and Munns, 1996). Depending on soil redox the mineralized C forms CO_2 or CH_4.

Soil biota represents 1 to 3% of soil C and the microbial component varies from 100 to 1000 µg C g^{-1} soil (Paul et al. 1999). The soil biological activity acts in the solubilization and availability of the soil nutrients to the plants (Borie et al. 1999) and it is an indicator of early nutrient changes (Powlson et al. 1987). The soil biological activity can be determined by various methods depending on the size and functionality of the soil organism. The microbial component including nematodes, protozoa, fungi, yeasts, microalgae, and a diversity of bacteria (Paul et al. 1999) is usually studied using microbial biomass, enzymatic activity, and respiration (CO_2 production) as well as other methods (Alef and Nannipieri, 1995). The microbial biomass measured as biomass C can be used as a sensitive indicator of soil management (Zhang et al. 2015), soil toxicity due to heavy metals, and other contaminants. Enzymatic activity studies are based on the fact that bacteria and fungi produce extracellular enzymes which help to decompose SOM to assimilable forms.

The respiratory activity measured through CO_2 production is an indicator of the activity of aerobic organisms in the soil (Högberg et al. 2005). CO_2 production may change with the quality of organic material (Arrigo et al. 2002) and with seasonal variations defined by the climate. It is also sensitive to alterations produced by various tillage systems (Carter, 1991; Reicosky, 2002; Acevedo and Martínez, 2003) and crop rotations. Crop rotations and organic fertilizer practices have a positive impact on total SOC (Arshad et al. 2004) and on microbial biomass-C (C_{bio}). In Chile, in a Typic Haploxerand, after 8 years of field management with various rotations, the C_{bio} decreased with increasing soil use intensity i.e. lowest prairie presence in the rotation (Zagal and Cordova, 2005). In the same work, the microbial quotient ($Qmic = C_{CO_2}\ C_{total}^{-1}$) had the highest values for the more intensive treatments. A low $Qmic$ indicates that the microbial communities are more efficient in the use of their energy resources (Traoré et al. 2007). Another index of microbiological activity (Anderson and Domsch, 1993, Angelli et al. 2001) is the metabolic quotient ($qCO_2 = C_{CO_2}\ C_{bio}^{-1} h^{-1}$). A higher value of qCO_2 indicates that the microorganisms have lower efficiency in the use of energy resources, or that they are more efficient decomposing organic residues.

A particularly important group of soil microorganisms are the fungi that form mycorrhiza. They are present in all arable soils colonizing all crops and weeds (Hendrix et al. 1995; El Omari and El Ghachtouli, 2021). They may be ectomycorrhizal or arbuscular mycorrhizal fungi. The arbuscular mycorrhiza add SOC through the mass of their extracellular mycelia and through the production of a glycoprotein called glomalin (Zhu and Miller, 2003; Curaqueo et al. 2010, 2011) (*Figure 5*). Glomalin is relatively insoluble and highly resistant to decomposition, and it is directly related to soil aggregates (Wright and Upadhyaya, 1998).

Another group of organisms that benefit from organic residues in the soil are soil worms. They consume organic matter mixed with soil and obtain energy and nutrients generating galleries in the soil that favor soil macroporosity (Amador et al. 2003), they increase soil aggregation and water infiltration rate, and they also improve soil chemical conditions through their dejections or casts (Medina-Sauza et al. 2019). There is a high correlation between number of soil worms and quantity and quality of organic residues (Hong et al. 2011). In general cereal and grass crops such as wheat and prairies have more soil worms than crops leaving lower quantities of residues (Bot and Benites, 2005). Soil management along with SOC has an effect on the number and weight of soil worms (*Table 1*) (Acevedo and Martínez, 2003).

Management system	SOC§ (%)	Number (Earthworms ha^{-1})	Dry weight (kg ha^{-1})
Conventional Tillage	1.4	0	0
No Tillage, 3 years.	1.7	620,000	28
No Tillage, 6 years	1.7	2,760,000	104

§ Soil organic carbon in 0-5 cm depth.

Table 1. Number of earthworms in various tillage systems in a long-term experiment at the Antumapu Experiment Station of the University of Chile. Values collected after 5 years of a conventional tillage (CT) and no tillage (NT). (Adapted from Acevedo and Martínez, 2003).

Soil properties as affected by soil management

Conventional soil tillage, including soil bed preparation, increases decomposition and mineralization of crop residues (Koch and Stockfisch, 2006), increasing the quantity of nutrients present in the soil but decreasing SOC and increasing the CO_2 flux from the soil to the atmosphere (Reicosky et al. 1997, Martínez et al. 2008a) (*Figure 3*).

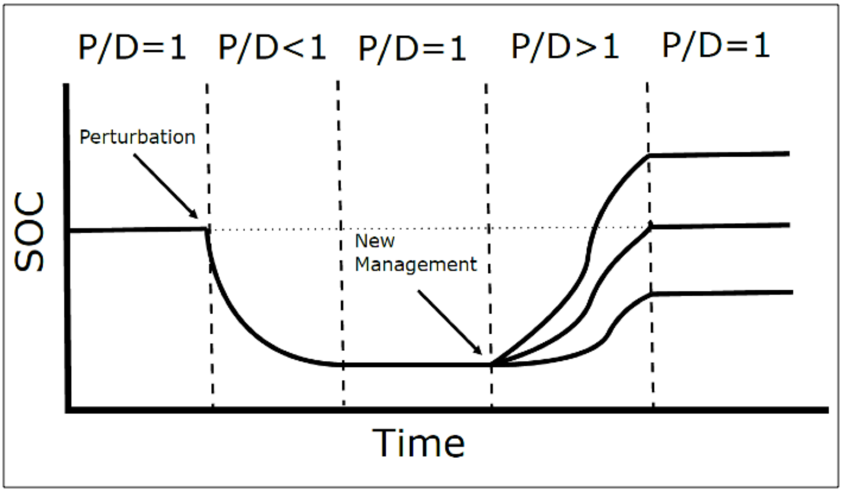

Figure 3. Soil organic carbon changes induced by soil perturbation followed by conservation management. Where P/D is the ratio of Production (P) to Decomposition (D) of organic residues. (Adapted from Johnson, 1995).

Soil Tillage temporarily increases total porosity which declines throughout the growing season. The pore continuity is worsened, and the bulk density increases throughout the season and may even results in smaller total pore volume and amount of coarse pores due to the running field management activities (Allmaras et al. 1967, Martínez et al. 2008a). No tillage can increase the water infiltration rate into the soil (Shukla et al. 2003, He et al. 2009, De Almeida et al. 2018) or reduce it (Valle et al. 2004), especially when crop residues are removed (Govaerts et al. 2009). Rhoton (2000), Lampurlanés and Cantero-Martínez (2003) and Martínez et al. (2008a) observed a greater resistance to penetration in NT with respect to continuous tillage (CT), a highly soil-specific phenomenon affected by soil management.

Other than nutrients, plants need adequate water, air, and an adequate temperature to reach their full productive potential, therefore, the stability of soil aggregates, which depends on the OM content of the soil (*Figure 2*) is important from the hydraulic and mechanical standpoint, as it favors the development of structural conditions that allow expression of the potential of crop yields. The aggregate stability gives information about the stresses that the soil is able to support without irreversible structural damage caused as a consequence of farming (Seguel et al. 2020). The penetration resistance of the soil indicates the strength of the plant roots needed for crop establishment and the Water Retention Curve (WRC) provides direct information about the volume of soil pores. From a plant standpoint the amount, continuity and interconnection of pores is important as it determines the availability of the water-air resources for plant roots (Neira et al. 2015).

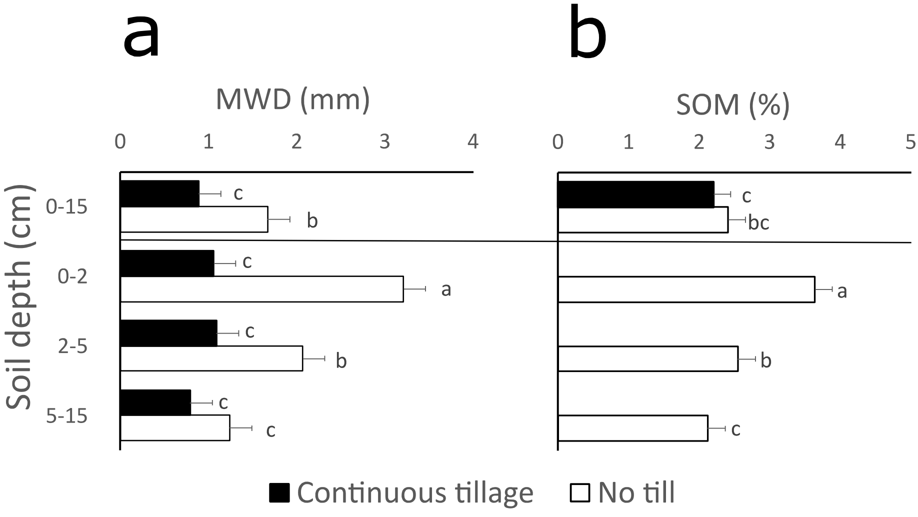

Figure 4. a) Mean diameter of soil aggregates and **b)** Soil organic matter in a Haploxeroll cropped under continuous tillage (CT) and no till (NT). Values obtained after four years of a wheat-maize rotation (Adapted form Reyes et al. 2002).

All mentioned soil properties are influenced by the tillage system and crop rotation, as they affect soil structure and its carbon content. *Figure 4a* shows values of mean diameter of soil aggregates for two soil management systems: CT and NT with straw mulching in a long-term experiment at Antumapu Experiment Station, University of Chile (Reyes et al. 2002). Clearly, the no till system favors a better soil structure due to the increased soil organic matter content

(*Figure 2 and Figure 4b*). The SOM concentrated mainly in the A horizon under NT (*Figure 4b*) along with N, P and K, the available soil water was significantly higher in the 0-15 cm soil depth, but the bulk density of the soil tended to be higher from 2 to 15 cm soil depth compared with conventional tillage.

Microbiological variables are considered suited indexes to evaluate the soil quality due to their sensitivity and ability to mirror changes of soil management. However, they frequently have high temporal and spatial variability (Curaqueo et al. 2010; Benintende et al. 2010) what becomes a concerned difficulty when selecting indexes of changes in soil quality in the different types of agro-ecosystems. They need to be calibrated and validated to know their variation with management. *Figure 5* shows arbuscular mycorrhizal parameters, glomalin fractions, water stable aggregates for the same experiment as in *Figure 4*.

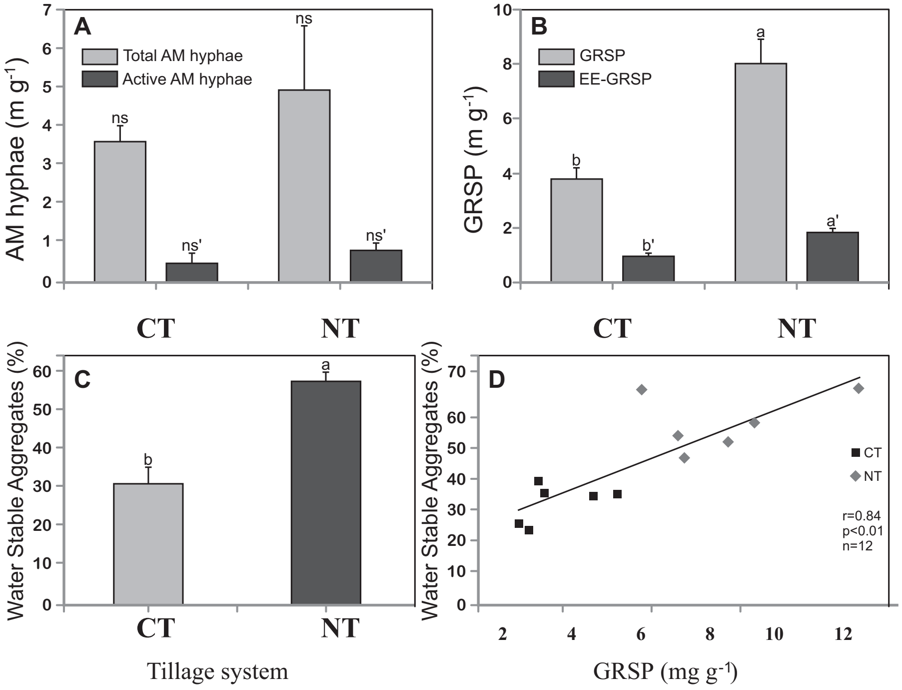

Figure 5. Arbuscular mycorrhizal parameters in a Mollisol of Central Chile managed six years under conventional tillage (CT) and no-tillage (NT) systems. **A)** Total and active AM hyphae. **B)** Glomalin fractions, GRSP: total glomalin; EE-GRSP: easily extractable glomalin. **C)** Water stable aggregates (WSA) in a Mollisol from Central Chile; and **D)** Correlation between GRSP contents and % of WSA in the agroecosystem studied. Different letters indicate significant differences among tillage systems according to the t-student test (P≤0.05). Errors bars indicate the standard error (SE) (Redrawn from Curaqueo et al. 2010).

Conservation agriculture

Conservation agriculture, a concept introduced by FAO (2009), was defined by Wall (2006) as "any management system that involves the characteristics of a clear reduction in soil movement with the final objective of its complete elimination, except for the small movement involved in sowing; the preservation of a permanent or semi-permanent organic soil cover; and the rotation of economically viable crops”. This type of agriculture is aimed at counteracting the negative effects of conventional agriculture on the renewable natural resources, (Acevedo and Silva, 2003; Wall, 2006; FAO, 2009; 2019). One of the central elements of conservation agriculture is the minimum and/or no-till. The soil is not inverted and the crop residues are left on top of the soil aiming at maintaining and/or increasing the soil organic carbon, essential element of soil sustainability (Rasmussen and Collins, 1991; Reicosky et al. 1995; Lal, 1997; Acevedo and Silva, 2003; Martínez et al. 2008a). No-till is a soil management practice that responds to an agronomy in which environmental sustainability is as central as crop yield. When the soil is not tilt and the crop residues are left on top of it, several agronomical consequences are generated. The crop residues are not incorporated into the soil and an accumulation and stratification of soil organic matter occurs at the soil surface (Rouanet et al. 1995, Martínez et al. 2013, Brunel-Saldias et al. 2018). The residues slowly decompose, adding organic matter to the soil which changes its physical, chemical, and biological properties.

The soil sustainability problem in the Chilean Mediterranean agriculture

The conservation agronomy/agriculture has rapidly extended around the world (Acevedo and Silva, 2003; FAO, 2019) but it is only partially used in the rainfed high yielding Chilean Mediterranean environments (Central Chile, having a high photo thermal quotient and rainfall above 800 mm/year). In these environments the summer rainfall is nil, therefore cereal residue decomposition (the main crop in the rotation) is limited by the lack of moisture (*Tables 2 and 3*) and the crop residues stay on top of the soil for a long period of time, from December-January through April-May in the southern hemisphere, almost through the planting of the next crop in the rotation, causing among others, physical problems to the planter if they are above 4 Mg ha^{-1} (García de Cortázar et al. 2003 ; Acevedo and Silva, 2003). At the break of autumn rainfall, the residues start to decompose at a time when temperatures are lower and limit the residue decomposition rate. The production of allelochemicals by the decomposing residues may limit crop germination and cause seedling mortality, particularly in the case of legumes, such as lupines following cereals in the crop rotation (An et al. 2002; Silva, 2007; Ordóñez et al. 2007; Silva and Acevedo, 2016; Silva et al. 2016). These problems do not occur in environments with summer rainfall (continental, tropical, subtropical) where temperature and moisture are adequate for residue decomposition and allelochemicals are washed out from the soil rhizosphere.

The conservation and efficient use of natural resources, especially nutrients, water and energy require maintenance or improvement of the biogeochemical cycles and appropriate technology for food production for the various ecosystems. It is necessary to develop sustainable agricultural systems allowing to maintain the biogeochemical cycles of C, N, P and S in a stationary state (ecosystem equilibrium) with high agricultural productivity and minimal environmental contamination. No-Till is central to this type of management. Crop yield in our irrigated long term NT experiment at the Antumapu Experiment Station of the University of Chile tended to decrease with increasing years of NT, with significant differences between CT and NT at the fourth and sixth year after NT implementation. The annual average wheat yield was smaller in NT as compared to CT (3,367 and 4,708 kg ha^{-1}, respectively). Plant emergence had a heterogeneous pattern under NT causing an irregular wheat cover. Furthermore, during the flowering stage, wheat of NT had fungi problems (*Fusarium* sp.). In general, fungi diseases intensify their damage when the soil has water infiltration problems (*Figure 6*) or when only Gramineae are considered in the rotation. In this trial, the wheat-maize rotation along with a decrease in soil water infiltration that occurred in the NT plots of this Mollisol (Martínez et al. 2008a) could have increased fungi damage. No significant differences in maize yield were found between NT and CT. Maize yield decreased during the first three years after NT implementation but this trend was reversed during the following years. As a result, the annual average of maize yield was similar in NT as compared to CT (16,543 and 16,241 kg ha^{-1}, respectively).

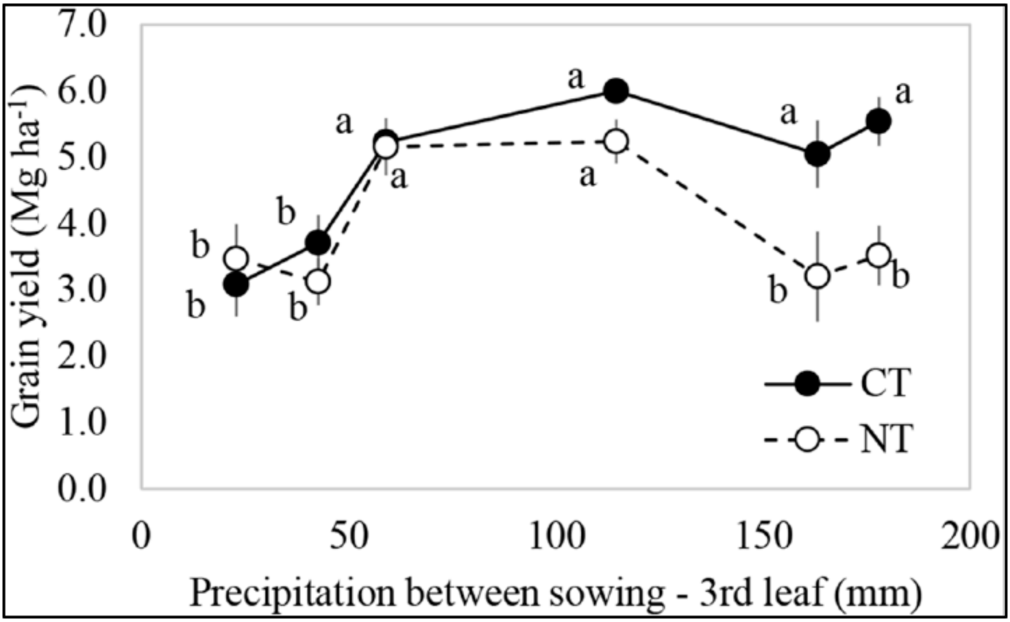

Figure 6. Effect of precipitation (rainfall + irrigation) between from sowing and 3rd leaf on grain yield of Conventional Tillage (CT) and No Till (NT). The lower yield after the 3rd leaf of wheat in NT treatment was attributed to hypoxia due to high rainfall in the period sowing-3rd leaf due to lower water infiltration rate of this treatment. Different letters indicate significant differences ($p \leq 0.05$) (After Silva et al. 2019).

In the high yielding Chilean Mediterranean environments winter crops, mainly winter cereals, peas, lupines, and canola are grown. Usually the cereal straw, having high C/N ratio, has low value and the farmers finally decide to burn it, defeating the purpose of organic carbon incorporation into the soil to provide sustainability to the agricultural system. Table 2 and 3 show the water effect on the rate of wheat straw decomposition at two residue levels having adequate temperature for decomposition (García de Cortázar et al. 2003, Silva et al. 2016). The results in *Table 3* indicate that it is not only the amount of water applied what affects straw decomposition but its frequency. Ten Mg ha^{-1} is a usual amount of winter cereal straw left on top of the soil after harvest (January/February) in the Chilean high rainfall Mediterranean (*Table 2*). Our experience shows that the amount of residual straw has to be decreased to around 4 Mg ha^{-1} to avoid problems to the planter in the next crop.

Decomposition time (days)	Quantity of residue (Mg ha^{-1})			
	Control	Sprinkled	Control	Sprinkled
0	5.000		10.000	
30	4.834 c	4.398 d	9.609 a	8.436 b
50	4.648 c	3.979 d	8.954 a	7.984 b
90	4.324 c	3.825 d	8.621 a	7.347 b

Different letters represent LSD $p \leq 0.05$ within each time of decomposition.

Table 2. Quantity of residue (kg ha^{-1}) left on top of the soil after 30, 50 and 90 days of decomposition, starting with 5 and 10 Mg ha^{-1} wheat straw residue under two situations: control (no watering) and low frequency sprinkler irrigation (After Silva et al. 2016).

Quantity of straw (Mg ha^{-1})	Water treatment (%)		Decomposed dry matter (Mg ha^{-1})	Weight loss of the straw (%)
3	+	20	0.37 a	15.0
	++	50	0.30 a	12.0
	+++	80	0.46 a	18.4
	++++	100	0.40 a	16.0
6	+	20	0.98 a	19.2
	++	50	0.90 a	17.6
	+++	80	1.04 a	20.5
	++++	100	1.06 a	20.9
9	+	20	1.06 a	14.0
	++	50	1.52 b	20.0
	+++	80	1.47 b	19.3
	++++	100	1.65 b	21.7

Different letters represent LSD $p \leq 0.05$ within each time of decomposition.

Table 3. Straw decomposition after three months, as a function of irrigation at the Antumapu Experiment Station of the University of Chile. The four watering levels, applied with equal frequency, are represented with symbols: lower (+) to higher (++++) and the average irrigation applied expressed as a percentage of maximum. (After García de Cortázar et al. 2003).

Crop and crop rotations

Worldwide food production needs to increase significantly mostly through sustainable yield increases to feed the world in 2050 (Cook et al. 2015). Increases in productivity achieved in the past are attributed in part to the significant use of fossil fuels. Energy use in agriculture is therefore also expected to rise, further contributing to greenhouse emissions. Productivity increases have often been accompanied by negative effects on agriculture's natural resource base, to such an extent that it affects its productive potential (*Figure 7*), this situation is further compounded by climate change impacts reducing the resilience of agro-ecosystems. Managing climate risks while improving resource-use efficiency and productivity of agro-ecosystems is therefore essential in order to reach food-security objectives. The integrated food–energy systems (IFES) can contribute to improved food security, energy access and adaptive capacity to climate change (Bogdanski et al. 2011).

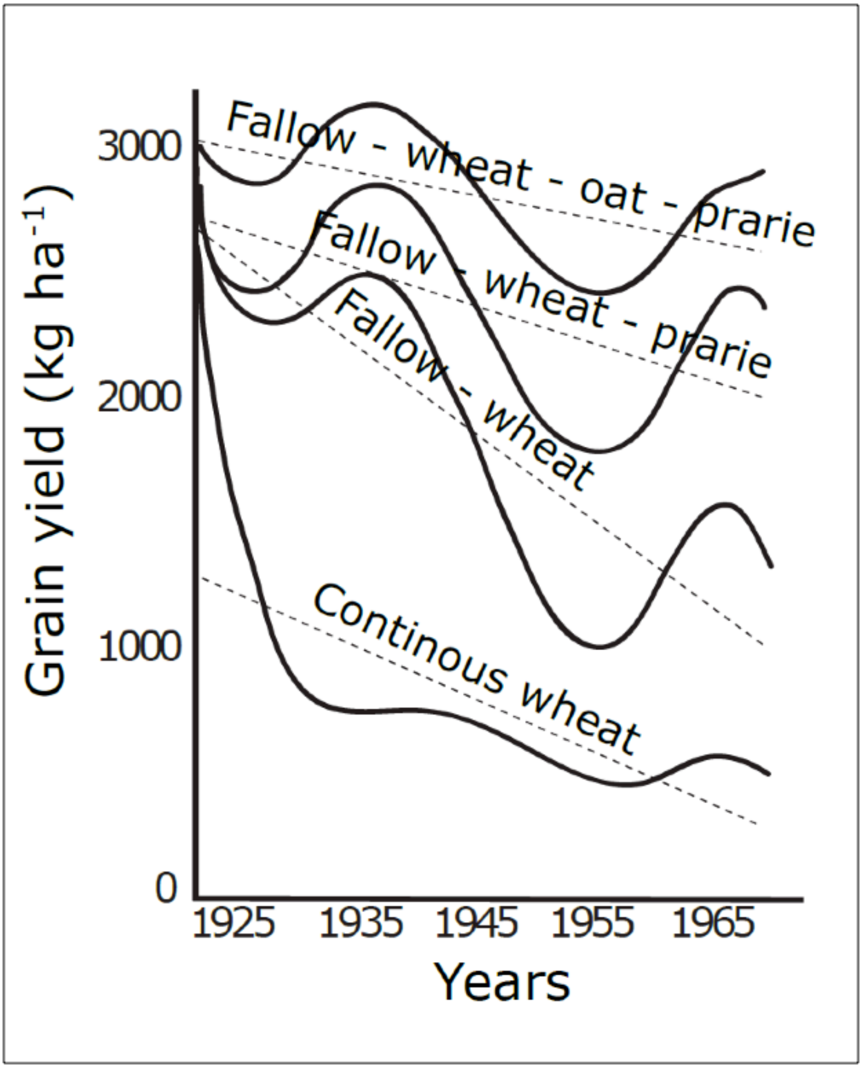

Figure 7. Changes in productivity of various crops and crop rotations. Analysis of a 40 years experiment (Vlek et al. 1981).

Modern agricultural yields of major food crops grown under relatively "good" conditions are considered to be the result of a mix of approximately 50% attributable to plant breeding and 50% attributable to agronomy (Fehr, 1984). These figures change when the crops are grown in stressful environments, the contribution of breeding decreasing as a result of the difficulty of breeding high yielding varieties for harsh environments, therefore the contribution of agronomy plays a major role, becoming crucial for sustainable agriculture and climate change, which for Mediterranean environments, shows an increase in temperature and a decrease in rainfall.

As indicated, the projected world food demand will require a sustainable intensification of field crops agriculture, fine tuning genotypes and agronomy for the various growing environments. No-till is central to agricultural sustainability, yet as mentioned, its adoption in Mediterranean environments is lagging behind, particularly in high yielding areas. The accumulated residues cause problems to the planters; allelochemicals limit germination and cause seedling mortality and sexual reproduction of pathogens occurs on the residue during the summer. When rainfall is high slugs thrive in the cool, humid environment provided by the straw (Acevedo, 2003).

Crop	**Problem** (Severity[1])	**Plausible Solution**
Wheat	Crop establishment after maize (*) Mechanical	- Reduce the quantity of straw (rowing, baling, animals, and biofuel).
	Nitrogen deficiency (**)	- Increase N fertilizer by 30%. - Use wheat genotypes with a higher N use efficiency
	Weeds (**)	- Rotation with oil crops and / or legume crops. - Development of wide spectrum herbicide tolerant wheat genotypes.
	Diseases (**)	- Use of tolerant varieties. - Avoid soil compaction (anoxia). - Use of fungicides.
Canola	Crop establishment (**) Mechanical	- Reduce the quantity of straw (rowing, baling animals, biofuel).
	Slugs (**)	- Rowing of the residue plus molluscicide.
	Weeds (*)	- Resistant varieties? - Rotations - Use of clearfield varieties.
Lupine	Crop establishment (***) Diseases	- Avoid soil compaction (anoxia). - Fungicide.
	Allelopathy	- Reduce the quantity of straw (rowing, baling animals. biofuel). - Change the sowing date from autumn to spring. - Use of varieties having lower sensitivity to straw allelochemicals.
	Slugs (**)	- Rowing of the residue plus molluscicide.
	Weeds (**)	- Use of more efficient soil-active herbicides in NT e.g., Linuron. - Development of varieties resistant to wide spectrum herbicides.
Maize	Nitrogen deficiency. (**)	- Increase N dose (30%). - Develop hybrids having a higher N use efficiency.

[1] *= low. **= moderate. ***= high

Table 4. Synthesis of the main problems faced by farmers introducing no tillage with crop residues on top of the soil in high yielding (above 4 Mg ha^{-1} of straw) Chilean Mediterranean environments and plausible solutions in rotations involving wheat, canola, lupine, and maize (Based on Acevedo et al. 2009).

Table 4, from Acevedo et al. (2009) propose agricultural practices and ideotypic traits of wheat for no-till that would help to overcome most of the production problems in these environments.

The proposed ideotypic traits are intended to overcome changes that occur in these environments when no-till practices are adopted, including soil mechanical impedance, anoxia, weed control, diseases, and allelopathy.

Crop rotations

Crop rotations are systems giving agricultural sustainability to crop production (Figure 7). In the Mediterranean Chile the crop rotations usually have only one harvest per year (Rouanet et al. 2005; Shertzer, 2013). The inclusion of diverse crops in the rotation is the most effective way of controlling pests and diseases, weeds and maintaining soil fertility. Contrasting crops are used in the rotation having differences in plant type (legume-grass), living cycle (perennials-annual), planting time (autumn-spring), agronomic requirements (low-high fertility), and others. The crop rotation affects the biodiversity of the system, 10-15 weed species are usually present in intensive rotations compared to 50 or more weed species in extensive rotations (Rouanet et al. 2005). It also affects the nutrients of the soil in quantity and absorption dynamics. Some crops in the rotations supply nutrients to the soil through symbiosis with bacteria and fungi. There is a close relation between the aerial biomass of legume crops and the quantity of fixed N, in the order of 21 kg of fixed N /Mg of dry matter of the legume crop (Mera and Rouanet, 2003) (*Figure 8*). Increases of 30-50% in the grain yield of a wheat crop after lentils, lupines, faba bean, or chickpea (Kirkegaard et al. 2008, Rouanet et al. 2005, Novoa et al. 1995, Espinoza et al. 2012) have been reported compared to the same crop without fertilizer. Species such as Lupinus albus in the rotation help to release fixed phosphorous of the soil to available forms to the plant through the exudation of citrates from their roots (Gardner et al. 1983) and enzymes such as acid phosphatase.

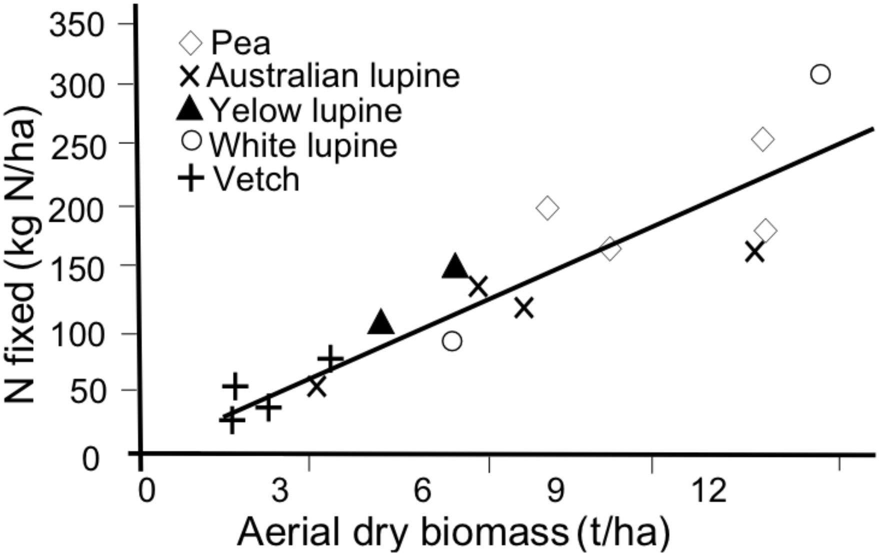

Figure 8. Aerial biomass of legume crops and fixed N by peas, Australian lupine, yellow lupine, white lupine, and vetch cropped at two locations in Central Chile (Adapted from Espinoza et al. 2012).

Crop productivity as an indication of soil sustainability

Crop productivity depends on the inputs of nutrients, water, pesticides, and others. Starting the second half of the past century there has been a continuous increase in crop production, mainly through improved crop varieties and agronomic intensification using improved agronomical practices. However, this was accompanied by a decrease in soil productivity, *Figure 7* (Vlek et al. 1981, Damrosch et al. 2000). Each increase in production required an even higher input vector. The productivity loss was found to be associated to organic carbon and nitrogen loss from the soil. Ryan et al. (2008), found that crop rotations produced benefits increasing the organic matter of the soil and soil fertility. In continuous crop situations a lower water infiltration rate, lower hydraulic conductivity and increased runoff have been observed, what could be reversed by a cereal-legume-pasture rotation (Ryan et al. 2008). Crop rotations also have a positive impact decreasing soil erosion as a result of a soil mulching of the residues. An example is the wheat-canola rotation that in Mediterranean environments quickly covers the soil in the period of high rainfall intensity (Karlen et al. 1994).

Crop rotations positively affect soil biodiversity as well as the plant biodiversity as a result of a better soil structure and soil organic matter content. In turn the increased biodiversity induces an increase of natural predators for crop pests and diseases, increases pollination of some crops and generates symbiosis (Karlen et al. 1994) Cereal based crop rotations including legume crops reduce nonrenewable energy demand, soil acidification and contamination as a result of lower N fertilizer use and lower use of agrochemicals such as pesticides and herbicides in general.

Rotation	1995	1997	1999	2001	2003	2005	Mean
Barley - wheat	5.54	3.36	0.72	5.21	3.33	4.35	3.75
Rye - wheat	4.30	0.97	0.18	4.55	3.46	3.35	2.80
Lupine - wheat	5.38	6.27	4.53	5.59	5.20	5.36	5.39
Continuous wheat	5.10	3.47	0.73	5.03	4.02	4.14	3.75
Triticale - wheat	4.53	4.11	0.50	4.52	3.89	4.40	3.66

Table 5. Wheat yield in two course rotations (Mg ha^{-1}) obtained in a 12 years experiment under Conventional tillage. (Silva et al. 2015; adapted from Mellado and Madariaga, 2003; Mellado, 2006).

Table 5 shows that while the same amount of NPK was annually applied to each rotation, the highest wheat yield was obtained when it was rotated with lupine, 44% higher than wheat in the continuous wheat rotation. This was explained by a sanitary effect of lupine controlling soil diseases and a higher amount of available P (54 mg P/kg soil vs. 14 mg kg^{-1} in the wheat monocrop) (Silva et al. 2015).

The quantity of straw has an effect on the next crop in the crop rotation. Wheat and oat were not significantly affected by the quantity of wheat straw in a study carried out in the foothills of the Andes in Central South Chile (Vidal and Troncoso, 2003), *Figure 9* shows, however, that canola and lupine decreased their grain yield significantly as the amount of wheat straw increased. In the case of lupine there was plant death caused by allelopathy and fungal attack (*Rhizoctonia* and *Fusarium*). In the case of canola most probably there were mechanical planting problems due to the small size of the canola seed.

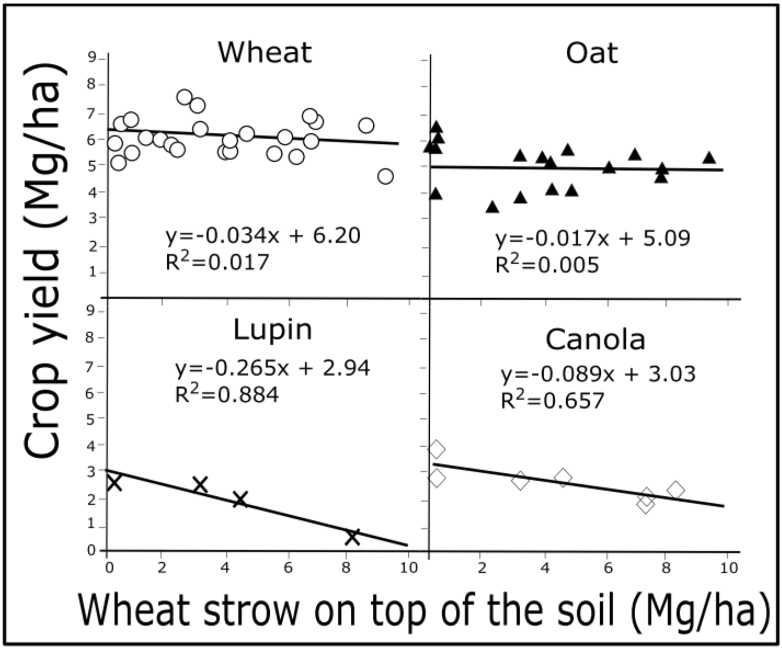

Figure 9. Wheat, oat, lupine, and canola yield related to the amount of wheat Straw present on top of the soil under no till management (Adapted from Vidal and Troncoso, 2003).

Conservation Agriculture emphasizes the importance of an intensive (in space) and diversified (in time) crop rotation in support of no till. Farmers must begin to regard crops as functional agronomic inputs, as a (partial) replacement for chemical fertilizers and herbicides. This means that farmers need to become familiar with a whole new system of production (see Acevedo and Silva, 2003; FAO, 2019).

Concluding remarks

Organic matter in the soil is most relevant to soil properties and agricultural sustainability, especially in Mediterranean environments like the Chilean Central Zone. Soil organic matter enhances chemical, physical and biological soil properties related to productivity.

Most agricultural ecosystems can be managed within sustainability boundaries and appropriate agronomy has been developed to this end. In our view the high yielding Mediterranean agricultural ecosystem is a special case which is described in this paper. Due to the characteristic dry summers, winter crops, particularly cereals are grown in rainfed agriculture in the wetter (southern) Central Chile. Cereal-legume and cereal-canola rotations dominate with one crop per year. The high C/N ratio of the cereal straw is a threat to its decomposition and allelochemical problems limiting conservation agriculture must be overcome. Some of the constraints to sustainable agriculture can be solved with agronomy but others need the development of specific varieties. Plausible variety traits and agronomic practices are presented in this review which may prove useful in developing increased agricultural sustainability in these environments.

Acknowledgments

The authors thank Dr. Paola Silva from the Soil-Plant-Water Relations Laboratory at the University of Chile for critically reading this manuscript.

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Chapter 10

Agricultural soil health and microbial biodiversity: organic amendments as a strategy for degraded soils

Carlos García and Teresa Hernandez

Introduction

The importance of soil as a natural resource, and the need for its conservation and protection is clearly established in the Resolution of the European Parliament on Soil Protection, of April 28, 2021. It focusses on agricultural soils, which are very complex because, in addition to the problems derived from erosion, soil degradation, or climate change, they are subjected to the actions derived from management, including fertilization, type of crop, etc. All this clearly has great impact on soil microbial biodiversity, with the consequent derived problems, since microorganisms conduct a large number of key processes in soil, among them, all those linked to soil organic matter (mineralization and stabilization). Some of the management and uses to which agricultural soils are subjected, lead to a decrease in soil organic matter, with the loss of organic matter being one of the problems that lead to their degradation, and lack of productivity and fertility, as well as to negative implications on soil biodiversity.

Therefore, in this Chapter, the authors focus on the need of using strategies to alleviate these organic matter losses. The addition to agricultural soils of quality organic matter (stable and sanitized exogenous organic matter, free of contaminants), capable of increasing the quality and health of the soils where they are applied can be a good strategy. Organic amendments will improve important soil properties and will positively interact with soil microorganisms, generating benefits on their microbial biodiversity, and increasing soil C content (C sequestration), one of the ecosystem services expected from our soils. The authors highlight the need of investigating what type of organic amendments are the most suitable to be used as a source of soil organic matter (perhaps a mixture of some of those found today: compost, biochar, manures, digestates, plant residues, ...), as well as increasing our knowledge about the interaction of this exogenous organic matter with the autochthonous one of the soils, and with the set of microbial populations existing in the receiving soils.

Soil and agriculture

Soil is a natural resource, essential for life on our planet; we have an obligation to protect and preserve it for present and future generations. Soil is a complex matrix, made up of mineral matter, organic matter, air, water and living organisms (worms, algae, bacteria, and fungi), combining to carry out physical, chemical and biological processes. It is, therefore, a living, dynamic entity, with continuous synthesis and degradation processes derived mainly from the activity of its microbial populations. Soil is one of the most species-rich habitats in terrestrial ecosystems and today we know that soil is not only the basis for agriculture, but also provides various ecosystem services that allow life on the Planet (FAO, 2015a and b), such as the provision of food, energy and raw materials, carbon sequestration, water purification and infiltration, nutrient regulation, pest control and recreation. Healthy soils provide these functions simultaneously, therefore, soil is crucial for fighting climate change, protecting human health, safeguarding biodiversity, and ensuring food security. Translated in political language, healthy soils are a key enabler to achieve the objectives of the European Green Deal such as climate neutrality, biodiversity restoration, zero pollution, land degradation neutrality, a sustainable food system and a resilient environment.

However, soil health is at risk in Europe and globally. For instance, 12.7% of European soil is affected by moderate to high erosion, causing an estimated loss of agricultural production in the EU of €1.25 billion per year. In Southern, Central and Eastern Europe 25% of soils show high or very high risk of desertification corresponding to about 411,000 km². Large parts of Southern Europe are likely to become desertified by 2050. In arid and semi-arid areas, in addition to the effects of erosion, compaction of soils, etc. (Sivakumar, 2007), climate change can aggravate soil health by damaging plant development due to reduced rainfall and increase in temperature, which reduces C inputs to the soil, potentially accelerating its degradation (Bisaro et al. 2014). The European Environment Agency recently concluded that the lack of a comprehensive and coherent policy framework which includes binding targets, incentives and measures to protect land and soil is a key gap that reduces the effectiveness of the existing policies and may limit Europe's ability to achieve future objectives.

The soil and its processes condition agriculture, affecting its productivity, functionality, and, therefore, its fertility. Agricultural land must be considered as the core of future sustainability, hence the interest in maintaining good health and quality. Europe is committed to this line, through programs such as "EJP SOIL" that has been launched in 2020, where the soil takes center stage for the agriculture of the future; or the new European pillars for "EUROPE HORIZON 2021-2027", where one of them is "Soil Health and Food", which clearly states that: "caring for the soil is caring for life". Due to various anthropic actions, sometimes derived from soil management not adapted to sustainability, as well as effects derived from climate change such as major droughts or excess greenhouse gases, the soil and the agriculture that develops on it may be affected negatively on its quality and health. Hence the interest in adapting agriculture to climate change, avoiding, through appropriate management and strategies, to negatively affect the soil and its degradation. In this context, the biodiversity of the soil, including here the microbial populations of the same, that are essential for the survival of the ecosystems, is threatened, and some of the factors indicated above, can undoubtedly have a negative influence on it. An important aspect in all of the above are the changes in land uses that are taking place, such as the intensification of agriculture and urbanization, overexploitation, pollution, etc., which can be causing damage to natural ecosystems. Once destroyed, their restoration is usually very expensive, and sometimes practically impossible.

Soil quality encompasses three basic components: biological, physical, and chemical characteristics. It is determined by concurrent functions such as sustaining biological productivity, maintaining air and water quality, and providing healthy conditions for plants, animals, and humans, while soil health generally refers to the biotic component of soil quality. Soil health has been defined as the capacity of a soil to function as a vital living ecosystem that sustains plants, animals, and humans (USDA-NRCS, 2019). The terms soil quality and soil health are not synonyms, but they are often used interchangeably in practice. Over the years, various indicators have been proposed to measure the health and quality of the soil (Bastida et al. 2008, Bünemann et al. 2018).

A healthy ecosystem is defined by the integration of nutrient cycles and energy flows, and by stability and elasticity in the face of alteration or stress. We know that global change (including the climate change), and soil management could affect to this healthy ecosystem. In general, the response of soils, as ecosystems, to disturbance has two components: resistance and resilience. Resistance is the inherent ability of the system to tolerate alteration and resilience is the buffering capacity and ability to regenerate. It is here where soil biodiversity plays an essential role since it conditions the recovery capacity of the edaphic system in the event of an alteration and the maintenance of its functional stability. Factors affecting soil resistance and resilience are soil type and vegetation, climate, land use, scale, and disturbance regimen (Griffiths and Philippot, 2013). We know that one of the greatest threats to many of the world's soils today is erosion. This erosion will have a more limited effect the higher the resilience of the soils.

There is a clear relationship between soil erosion and the loss of its organic matter and its biodiversity. The European Union, through the European Soil Data Center (ESDC), and the studies carried out by various researchers (Orgiazzi and Panagos, 2018), indicates that we should advance on issues such as: *i)* Quantifying the interaction between degradative processes in soils as erosion, and soil biodiversity; *ii)* Development and integration of a "Biodiversity Factor"; *iii)* Establish the ecological impacts on the soil living community, of degradative processes (erosion, loss of soil organic matter, etc.). A fundamental aspect in favour of establishing strategies for the conservation and protection of our soils, whether natural, agricultural or forest soils, is that it constitutes the largest reserve of terrestrial organic carbon, specifically, more than double the amount stored in vegetation. We know the importance of capturing CO_2 from vegetation, but even more important should be to increase the soil capacity as a C sink. Soil mitigates climate change by sequestering carbon and reducing greenhouse gas emissions (Guerra et al. 2021). World's soils should be part of any program dedicated to tackling climate change, thus ensuring future food. It is time to create a general awareness in favor of carbon in the soil, so that the soil is considered as a true living entity, a natural resource that must be protected, and not only as an inert support for plant growth. Soil is the basis for agriculture and the substrate for natural ecosystems across the planet.

Everything that has been exposed so far has been studied extensively for natural soils, including forest ones, but we need to improve our knowledge about what happens to the quality, health, functionality, and biodiversity of soils in agricultural systems as diverse as those that exist, with such varied approaches. A good part of our soils is dedicated to agriculture; we have to be able to be efficient in their productivity, but at the same time, sustainable in terms of the ecosystem services that they can generate. Agricultural soils are subject to a management that should be increasingly sustainable. Such management includes aspects such as the water used for irrigation (quantity and quality), the management of said soils, types of crops, and, above all, fertilization and any input they incorporate.

When it comes to processes that occur in soils, or their biodiversity, the complexity when these are agricultural soils increases considerably in relation to natural soils. To all this, we must add that climate change forces us in the case of agriculture and forest soils, not only to adapt them to said climate change, but also to try to collaborate to mitigate it with that agriculture that is proposed for the future. We consider that the challenge lies in proposing an "INTELLIGENT AGRICULTURE", capable of combining sustainable productivity with the possibility of avoiding negative effects on ecosystems.

Soil agricultural management and soil health

Soil productivity will be one of the main factors for feeding the planet. Changes that impact on said productivity (soil degradative processes, from erosion to contamination, compaction, excessive inputs, irrigation with low-quality and saline waters, etc.) will affect soil health and biodiversity. Faced with this, options for sustainable soil management in agriculture are necessary. There are four management practices that are considered critical to have a favourable impact on soil health, and therefore, on its biodiversity: *1)* Conservation practices such as cover crops. Cover crops could potentially be used across the tropics to increase soil health in these systems, and on occasion, could even increase the health of low-quality, degraded tropical systems, which could respond significantly to the improvement in soil biological functions and nutrient cycling resulting from cover crop use, in the subsequent crop. Such improvements in soil biological functions and soil biology would make the soils more resilient and improve their ability to adapt to a changing climate. Legumes as cover crops, or even a mixture of legumes and non-legumes, should be tested to know their usefulness as sustainable management, with little impact on the agricultural productivity of the soils (Delgado et al. 2021); *2)* We also know that an excess of tillage can destroy the structure of the soil, and help to mineralize organic matter, favouring the release of CO_2 into the atmosphere. It is necessary to increase our knowledge about tillage technologies so as not to harm soil health and to maximize the profitability of agricultural productivity; *3)* Crop rotation is another way of helping to maintain and improve soil health, and therefore, its biodiversity; in particular when plants such as alfalfa are included within the rotational system, thus improving soil health indicators such as carbon, nitrogen and aggregate stability; *4)* Lastly, important as a strategy for degraded agricultural soils and soils with low fertility, is the use of organic amendments a key factor within an adequate management to increase soil productivity and diversity. This type of strategy to combat soil degradation in agriculture, and favour C sequestration, thus mitigating climate change, will be discussed later in this Chapter.

There are many studies on this type of management, which are sometimes not easy to compare due to the large number of different organic amendments (different origin, different stability of their organic matter, different nutritional content, etc.) that are used. Many scientific works have been carried out on the effect of the management of agricultural soils on their health and biodiversity. Management techniques that focus on soil health are promising solutions to mitigate some environmental impacts and may increase economic returns. Widespread implementation of management practices that promote soil health, or “the continued capacity of soil to function as a vital living ecosystem that sustains plants, animals, and humans” (USDA-NRCS, 2019), is a hopeful counter to agroecosystem land degradation. As example, there is evidence of short- and long-term yield reductions with no-till in some cropping systems (Giller et al. 2015). A recent global meta-analysis compared yields under no-till versus conventional tillage using >5,000 paired yield observations across crop types (Pittelkow et al. 2015). In this study, an overall yield decline of -9.9% when no-till was implemented alone was observed. Yield declines were lessened when no-till was coupled with residue retention (-5.2%) or rotation (-6.2%), and negative yield impacts were further reduced when all three practices were concurrently applied (-2.5%) (Pittelkow et al. 2015).

Conventional management practices such as ploughing, cultivation patterns, as well as the use of pesticides and fertilizers, have had a negative influence on the quality of water and the atmosphere, since they have generated changes in the capacity of the soil to produce and consume gases like CO_2, nitrous oxide, and methane. We know that C and CO_2 are essential to manage essential processes in cells. What is more, with regard to CO_2 as a typical greenhouse gas, agricultural vegetation plays an important role in capturing CO_2 from the atmosphere within its photosynthesis process, including C in its own system and generating biomass. Part of said biomass can be incorporated into the soil, originating, on the one hand, the microbial activity that degrades said plant residues and emits CO_2 into the atmosphere (negative effect), but on the other hand, part of said carbon from plant remains will be incorporated into the soil in a fairly stable way (carbon sink, positive effect).

An important aspect for soil microorganisms is the use of conventional or conservation management in agriculture. The type of land management practices in agroecosystems impacts on the structure of microbial community and function through a variety of different mechanisms. Land-use changes also impact on soil microbial community structure through alterations in carbon availability and quality, pH and nutrient availability (Ogunsola et al. 2020). Since the ratio of fungal to bacterial population is commonly measured as indicator of microbial community structure, and the relative proportion of fungi is increased by no-till practices, crop rotations, and use of cover crops, biological mechanisms are regulating carbon and nitrogen exchanges between the land, water, and atmosphere. This reveals the importance of soil management and conservation approach in enhancing microbial activity for soil ecological intensification as well as buffering the soil to neutralize contaminants.

One aspect to highlight is that, as previously indicated, in agriculture, defective and sometimes irrational anthropic actions have promoted the loss of organic matter, reducing fertility, negatively altering different soil properties, and also increasing erosion and atmospheric CO_2. Consequently, inappropriate agricultural practices contribute to global warming of the planet. The high levels of gases associated with the greenhouse effect and the alterations in the hydrological cycles have produced changes in the global climate, as well as the reduction of ozone. To face this threat, it is required, on the one hand, to understand what is happening with a natural resource such as the soil, and how the agricultural management of said soil is affecting soil processes, and on the other hand, to propose alternatives for soil use that maintain both environmental quality and efficiency, that is, sustainable alternatives through viable and environmentally safe action strategies.

If we seek “sustainability” in agriculture, within a climate change scenario, we need a new vision and holistic approaches to ecosystem management. We have to face the growing demand for food, fibre, and environmental protection. The health and quality of the soil can be altered in the face of temperature variations, or constant drought processes. It is interesting to have knowledge to maintain a future agriculture adapted to these climatic changes. There, the soil has a lot to say, since, if it has good quality and health, it can mitigate the negative effects of the afore mentioned climate change. EJP SOIL proposes models to predict soil carbon sequestration, GHG emissions, soil degradation (soil compaction or soil functions), the provision of ecosystem services by agricultural soils, including agricultural production and climate change mitigation, under climate-smart sustainable soil management options. The outputs of the scenario modelling exercises could be compared when relevant with the quantitative targets proposed by the Mission Board on Healthy Soils and Food (e.g., current C losses in farmlands should be reversed to an increase by 0.1-0.4% per year). Everything stated in this section indicates that our obligation is to turn to agricultural systems that are sustainable. This will allow us to produce, keeping our natural resources in acceptable conditions (*Figure 1*).

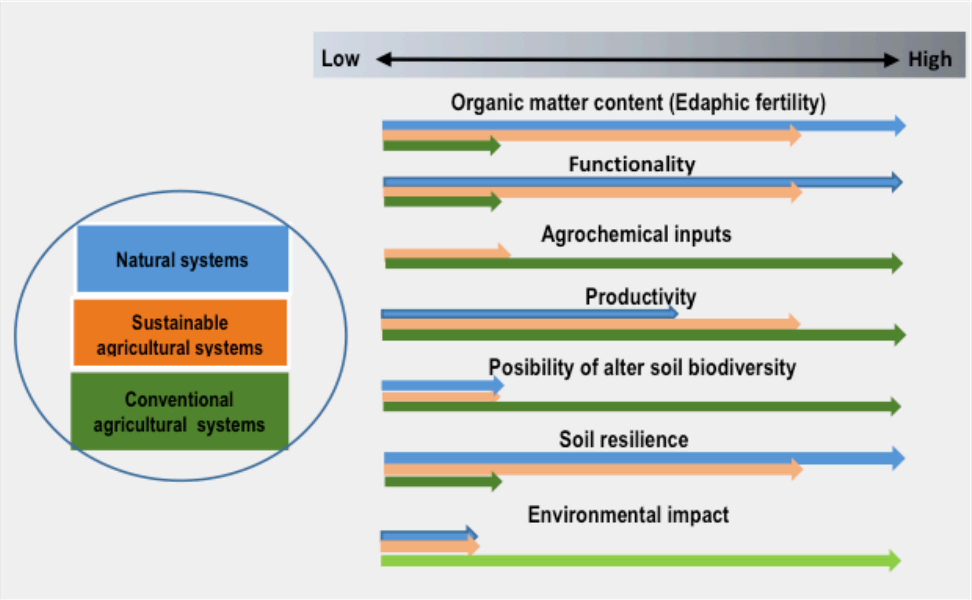

Figure 1. Some soil properties in different agricultural systems.

Microorganisms in agricultural soils

Microorganisms constitute the majority of the living part of the soil and are responsible for the dynamics and transformation of its organic matter. In this way, the microbial populations are immersed in an interaction framework that affects the development of plants (when carrying out processes of transformation of compounds to elements that can be assimilated by their roots) and the quality of the soil, ensuring the stability and productivity of both, agrosystems and natural ecosystems. The microorganisms of the rhizosphere contribute to plant growth by increasing the availability of limiting nutrients such as phosphorus and nitrogen. In turn, the composition and activity of the bacterial community is strongly influenced by the type of vegetation present in the soil. Some functions of microorganisms act on the availability of nutrients, on aspects of biological pest control, on the biodegradation of pesticides or other organic compounds, or even on the formation of soil aggregates.

The soil microbial community can be studied from different points of view. In the first place, to know how the microorganisms act and to do this we turn to the study of various indicators of microbial activity. Secondly, it is important to study its “size”, that is, microbial biomass. Third, it is important to know indicators related to the microbial structure and composition of the community. In this way, we will be able to know which microbial groups are more related to a certain process (i.e., restoration of a soil).

Microbial biodiversity in agricultural soils is a term that includes all components of biodiversity (genetics, species, and ecosystem levels) that are of importance for food and agriculture and that support the ecosystems where agriculture occurs (agricultural ecosystems). If we speak in general terms, biodiversity is the source of the plants and animals that form the basis of agriculture, and of the immense variety within each of the crops and livestock species. Countless other species contribute to the essential ecological functions on which agriculture depends, including soil services and water recycling. The great diversity of plants that have been selected and cultivated for millennia, and the animals that have been domesticated and raised, form the basis of the energy resources that farmers, ranchers, and other agronomists will be able to draw on now and in the future. FAO et al. (2020) defines soil biodiversity as *"the variety of life belowground, from genes and species to the communities they form, as well as the ecological complexes to which they contribute and to which they belong, from soil micro-habitats to landscapes"*.

Soil organisms provide essential services for the sustainable functioning of all ecosystems, and therefore are important resources for the sustainable management of agricultural ecosystems. Most of the biomass of our planet is found in the soil, where microorganisms use organic or mineral matter as a source of nutrients and energy. They play a fundamental role in soils, since are ultimately responsible for the state of organic matter and, in general, of the development and functionality of a terrestrial ecosystem.

Thus, the biological fertility of the soil is the result of many processes in which microorganisms intervene, such as the processes of initial soil formation from the mother rock, formation and maintenance of the soil structure, decomposition of organic matter, fixation and transformation of nitrogen, regulation of nutrient cycles and improvement of plant growth through the synthesis of growth factors and antagonisms with plant pathogenic microbes. Soil structure and its biota are interdependent; soil structure affects soil biota while it contributes to the formation and stability of soil aggregates. One of the most important functions of soil microorganisms is to break down the various kinds of organic matter of plant or animal origin. The ability of a soil to decompose agricultural plant remains is substantially controlled by the relative abundance, composition (e.g., fungi/bacteria ratio) and activity of soil microbiological communities, and by soil-plant microorganism interactions. Fungi decompose low-quality waste and recalcitrant organic compounds more efficiently than bacteria and, therefore, are associated with soils with lower fertility and drier conditions. On the other hand, bacteria thrive in environments where the entrances of more labile forms of C dominate. The microorganisms that inhabit soils, and that make up their microbial biodiversity, need carbon as an energy source (they oxidize it and return it to the atmosphere as CO_2), and they take it from plant remains and microbial material.

As it will be indicated later in this Chapter, within the various management that are being carried out on agricultural soils and that will affect the set of soil microbial populations, the incorporation of organic amendments to the soils to improve their quality and health, will undoubtedly affect soil microorganisms, as well as different physical properties (density, compaction, porosity), chemical (C and its fractions, nutrients), and biological (microbial respiration, enzymatic activities, etc.) (Coll, 2020). Within the set of soil microbial populations, those linked to the roots of the plants are of special interest. Exudates from living roots stimulate a quick response of soil microbes with acceleration of native soil organic C mineralization. Rhizosphere, a thin area of soil surrounding roots receiving C exudation from plants, represents a site of intense competition for available C and nutrients between surface-reactive particles and soil microorganisms. This competition can reduce the amount of available C to a critical level, becoming limiting for microbial growth and soil organic matter degradation. On the other hand, acceleration, or retardation of the degradation of soil organic C caused by root activity is termed rhizosphere priming effect (RPE) (Merino et al. 2015, Luo et al. 2014). Rhizosphere priming can also be affected by nutrient availability and substrate quality (Murphy et al. 2015).

These interactive effects may be of particular relevance in understanding microbial growth and nutrient supply in response to increased atmospheric CO_2 concentration and temperature. Thus, in the rhizosphere there are microorganisms capable of fixing N_2, as well as ammonifying and nitrifying bacteria, responsible for the conversion of organic N compounds to inorganic forms (NH_4^+ and NO_3^-), available to plants. Microorganisms can also dissolve insoluble minerals that control the availability of some elements such as phosphorus (both native and applied) through the release of some organic acids, or produce phosphatases that act on organic P. The availability of S, Fe and Mn is also affected by redox reactions, carried out by bacteria in the rhizosphere. Likewise, chelating agents can control the availability of micronutrients and participate in biocontrol mechanisms of plant pathogens. In addition, microorganisms improve soil structure by the formation and stabilization of soil aggregates through de excretion of compounds that bind clays and organic materials.

Within the whole set of microorganisms that inhabit the soil, we can include the concept of the MICROBIOME, which comprises the set of genes and microorganisms organized in a suitable way to fulfill functions of interest. Nowadays, and particularly in agriculture, there is a huge movement that is based on the knowledge of the soil microbiome, since from this microbiome actions and processes are suggested that may be of interest for agricultural productivity, soil health, and for functions such as solubilization of nutrients such as P, incidence on beneficial microorganisms normally linked to the rhizosphere, etc. It seems that the time has come to give interest to the management of soil organisms and direct their functions to achieve specific objectives. In the future, the management of soil microbial biodiversity may perhaps condition positive effects on soil health, fertility, and sustainability. We must not forget that we have to know: *1)* the best way to characterize and diagnose said microbial biodiversity through appropriate analytical techniques ranging from those that indicate microbial activity, enzymatic techniques, and others within what we know as “Omics”; *2)* what happens in the soil in relation to processes, functionality and effects: *3)* how to manage microorganisms. The challenge is that all of this is feasible in the near future. We should be able to “work à la carte”: depending on what is needed, build new “preparations” for the soils, and generate agriculture more in line with the knowledge we have. In any case, we continue to express the difficulties that exist with agricultural soils, where their management based on types of cultivation, management, fertilization, etc., clearly conditions their microbial biodiversity and their microbiome.

Threats to soil health and biodiversity

Climate change poses a major challenge for food production, and global warming and water scarcity have endangered agricultural systems, especially in semi-arid climates (Zalidis et al. 2002, Garcia et al. 2017). The sustainable management of the world’s agricultural soils and sustainable production are essential to reverse the trend of soil degradation and guarantee the current and future food security of the world (FAO et al. 2020). The degradation of the soil results in a loss of its fertility, in the decrease of its carbon concentration, biodiversity and water retention capacity, in the disorganization of the nutrient and gas cycles and in a reduction in the degradation of pollutants (Bai et al. 2008). Various threats such as pollution, erosion, soil compaction, inappropriate agricultural practices, influence of climate change, etc., affect chemical and physical soil properties; all of this can induce irreversible changes on soil biodiversity (Nawaz et al. 2013). Soil compaction produces a reduction in pore size that leads to lower oxygenation and higher water content, ponding, reduction of microorganism and worm activity, root development and productivity. The decrease in the content of organic matter in many soils is becoming an important degradation process, particularly in semi-arid Mediterranean European regions (Garcia et al. 2017). In addition, and as already mentioned above, various soil managements (for example, tillage), also contribute to a loss of organic matter from the soils. In areas with a semi-arid climate regime, the progressive degradation suffered by soils will primarily affect the vegetation they support (profitability and sustainability). This can lead to abandoning agricultural soils, which, in the existing climate conditions, will not be able to recover their fertility and productivity, and will be subjected to intense processes of water erosion. Therefore, there will be a loss of the most superficial layer of the soil, which is precisely where we find the most soil fertile part. This fertile part contains a large part of the soil's microbial biodiversity, so it can be concluded that, if agriculture and its vegetation disappear, a good part of the soils of semi-arid climates could then be deprived of their main barrier against degradation and erosion (its vegetation). This leads to a loss of organic matter and to a decrease in soil fertility, productivity, sustainability, and biodiversity. Common agricultural practices such as excessive use of agrochemicals, intensive crops, deep tillage, and over-irrigation have led to soil degradation, increased gas emissions, and contamination of water resources. The loss of soil organic matter is becoming the main cause of soil degradation, particularly in European semi-arid Mediterranean regions, where these negative effects are aggravated by inherent environmental factors, such as lithological substrate, relief, and semi-arid climate. Mediterranean soils rich in calcium carbonate and poor in organic matter are susceptible to erosion and significant C losses due to seasonal changes. Therefore, in order to implement sustainable agriculture in these soils, it is important to establish adequate management, which includes the contribution of external organic matter, ensuring the supply of nutrients and consolidating the continuity of the crops. According to Lal (2009), the soils of agroecosystems worldwide have decreased their organic carbon reserve by 25-75% depending on the climate, the type of soil and its historical management, and these losses may be of a magnitude of 10 to 50 t C / ha. This decrease in the C pool has led to a decrease in the productive capacity of these soils and in the efficiency in the use of the added inputs, as well as in their ability to sequester atmospheric CO2. All of the above (loss of organic material and organic C in soils subjected to degradative processes, including agriculture), already give a clear indication of the interest in increasing organic carbon content in agricultural soils, and to increase the fertility and productivity of the crop's soils. In this sense, the strategy of adding organic amendments that manages, on the one hand, to improve the edaphic fertility of the soils, and on the other hand, to sequester C in the soils, is an acceptable option (Coll, 2020). In addition, organic amendments provide both organic C and organic matter, and according to Laban et al. (2018), the biodiversity of the soil, its organic C, and its organic matter, are closely related. Most of the living organisms in the soil (bacteria, fungi, protozoa, insects, worms, etc.), are found in the first few centimetres of the soil, which is precisely where this organic part is, and they help to preserve the fertility of the soil.

As above indicated, a threat that has already been verified is climate change and its impact on agricultural soils. The microbial populations of agricultural soils, and their activity, are altered by climate change (rain regime, use of poor-quality water, increase in soil temperature, etc.), as well as by some of the inputs used or others managements carried out in agriculture that have sometimes been misunderstood. This type of highly varied anthropic actions has negatively conditioned soil biodiversity and the production of food generated by agriculture. In turn, agricultural land is under great pressure, due to the increase in the world population and the need to ensure its food.

For this reason, it is necessary to know how changes in land use together with climate change can affect the pool of microorganisms that live in it, since these will help to maintain agricultural productivity. It seems totally necessary to adapt our agriculture to present and future climate changes, so that the microbial biodiversity of the soils is not negatively influenced, and makes our agriculture lose efficiency. If we do not succeed, the life of the soils and the agriculture that they support will suffer, calling into question the much-desired sustainability.

Fertility and microbial biodiversity of agricultural soils with organic amendments incorporation

In theory, the addition of organic matter is carried out by the constant recycling of it, in the form of residues of living or dead plants and animals. However, in large part the soils of vulnerable zones, the C inputs are insufficient since the balance between the C inputs from the vegetation and the mineralization rate produced by the microbial activity of the soil is negative. In these areas, mineralization processes predominate over humification or C fixation, a common characteristic in these soils being the low content of organic matter and its exposure to degradative processes.

Degraded soils need strategies to reduce erosion and remedy the effects of degradation. Soils in semi-arid areas have very low microbial activity, low levels of microbial biomass and low content of organic matter (0.5-2%). If the characteristics of degraded soils in semi-arid areas are recovered and improved with the application of organic amendments, the cycle of nutrients (Clark et al. 2007, Hernandez et al. 2015) and their quality in general will be improved. As previously indicated, the degradative processes in soils affect natural soils, and in particular, agricultural ones. Soils lose organic matter, and this affects their fertility and functionality, as well as the biodiversity of said soil itself (remember that organic matter acts by providing substrates to microbial populations and it is an important part for the functioning of microbial populations that are found in soils).

To recover organic matter, one way that should be considered is to propose exogenous contributions that are capable of building new organic matter in the soils. However, this is not easy because the behaviour of exogenous organic matter and its ability to influence the generation of new stable organic matter in soils will depend fundamentally on the quality of said organic matter, and on the receiving soil, cultivation management and climate.

When it comes to adding “organics” to the soil, we must bear in mind if this is done in order to increase the nutritional contents of the soil fundamentally (organic fertilizers, potential substitutes for mineral fertilization, in the knowledge that the nutrients are provided gradually, depending on the possible mineralization of the organic matter), or are intended to improve soil health in general, increasing carbon and organic matter content, and achieving slow but consolidated improvements, with soil properties as varied as the physical, chemical, biological, biochemical and microbiological properties.

As exogenous organic materials capable of acting as good organic amendments for soils, organic waste such as manure, various agricultural waste, urban organic waste such as domestic waste, biosolids (although these have many problems of incorporation of pollutants, and have various limitations), organic by-products of different agroindustry types, organic waste conveniently treated by composting, for example (compost), and more recently, waste such as biochar (derived from pyrolysis processes of different organic waste), can be used as amendments to increase soil fertility, since while they provide nutrients to the crop, they improve the quality of the soil. These organic materials can partially or totally replace, depending on the residue and the requirements of the crop, the synthetic fertilizers used in the crop, while increasing the total organic matter in the soil (Hernandez et al. 2014, 2016, 2018).

However, here too, it must be based on a clear premise: the exogenous organic matter that is contributed to the soils must be of "QUALITY". This means that the sources of exogenous organic material that reach the soil must be hygienically healthy (without pathogenic microorganisms), without problematic content in heavy metals, salinity, or other emerging pollutants of anthropic origin, including microplastics), and with an acceptable stability of its organic matter, so that it can have a favourable effect on the physical, chemical and biological soil properties. The soil is a natural resource that must be conserved and protected, in particular its biodiversity. It cannot be a place where any organic matter is added due to the fact that it has carbon and nutrients. It must not contain toxic elements that could put the health and biodiversity of the soil, or any of the ecosystems that could be affected at risk. Sustainability criteria must be maintained, and for this, the quality of organic materials used as fertilizers or amendments is essential.

Building organic matter in the soil is a slow and gradual process. It may take up to a decade or more to increase soil organic matter to significant levels, however, its benefits can be seen after several additions of organic residues (Toor et al. 2015). An interesting initiative is the international initiative "4 per 1000" (to increase 0.4% or 4 per thousand each year the organic C in agricultural soils), launched by France on 1 December 2015 at the COP 21. It consists of federating all voluntary stakeholders of the public and private sectors (national governments, local and regional governments, companies, trade organizations, NGOs, research facilities, etc.) under the framework of the Lima-Paris Action Plan (LPAP). The aim of the initiative is to demonstrate that agriculture, and in particular agricultural soils can play a crucial role where food security and climate change are concerned.

On the other hand, the mineralization rate of organic matter varies greatly depending on the type of organic matter and environmental conditions. Adding new stable organic matter and maintaining native soil organic matter will undoubtedly decrease CO_2 levels in the atmosphere. In this sense, the Report on "Organic Matter and Biodiversity" of the European Strategy for Soil Protection, which is the preamble to a new directive, identifies the loss of organic matter as one of the main causes of soil degradation, and underlines that exogenous organic matter is currently an invaluable organic source to the soil, favouring the development of a stable plant cover (Garcia et al. 2017). The strategy of increasing the level of organic matter in the soils and getting part of said organic matter to collaborate in increasing the amount of sequestered C, will depend on the fact that in the processes that occur in the soil on said applied organic matter (mineralization and humification or stabilization), prioritizes said humification over mineralization. If the stability of the added organic matter is adequate and does not determine an excessive attack of the microorganisms on said organic matter, it will be possible to encourage the C sequestration in the soil, and the benefits that this ecosystem service entails (Hueso et al. 2012, Coll, 2020). On the other hand, the greater plant development caused by the addition of organic amendments will determine an increase in C inputs through plant remains and root exudates, contributing to the storage of C in the soil.

In this sense, we can indicate that the contribution of significant amounts of organic matter, through exogenous organic materials that makes up for their scarcity in these soils, whatever the type of organic material is, will affect the microbial populations of the soil, since with the organic material we are adding substrates to the autochthonous microorganisms of the receiving soil, as well as own populations contained in the organic material. We can point out that the addition to the soils of exogenous organic matter entails effects of biostimulation (diverse substrates for existing microbial populations), and of bioaugmentation, impacting with "direct" microorganisms on those existing in the soil. As we have already pointed out, the introduction of exogenous organic matter into soils will significantly improve their physical, chemical, and microbiological characteristics. Therefore, the use of organic amendments in the restoration of degraded and eroded soils can also counteract the effect of global warming by increasing stable and fixed carbon in the soil, avoiding its release as CO_2. Many of these effects will be driven by microorganisms, and they can be affected positively or negatively by the introduction of these amendments. The premise we should have with regard to the use of organics in agricultural soils is that they must not harm the microbial biodiversity of soils, and that biodiversity must be conserved, and improved if possible.

The stability and maturity of the organic amendment are two important factors to consider in order to obtain success in the recovery of degraded soils (soils with low organic matter content and/or high erosion level). Generally, for organic amendments the term "stability" related to the rate of degradation of their organic matter, while the term "maturity" refers to the decomposition of potentially phytotoxic organic compounds (Larney and Angers, 2012). The application of unstable or immature organic amendments may cause adverse effects on soil properties, such as bad odours, alteration of soil and water pH, immobilization of available N and addition of phytotoxins and animal pathogens in soil and water, excess of nutrients, heavy metals, etc.

Characteristics of some of the organic waste used as organic amendments for agricultural soils

The variety of organic materials, in many cases derived from residues, which are used as organic soil amendments, is high. We give below some indication about the properties of some of the most used.

Animal manure

Manure is the solid and liquid excrement of animals, mixed with the plant residues that have been used as a bed. It has long been the most widely used organic fertilizer of animal origin to replenish the natural fertility of soils. Its incorporation into the soil provides nutrients, increases moisture retention, and improves soil biological activity and therefore, soil fertility and productivity. Like all other organic fertilizers, manure does not have a fixed concentration of nutrients This will depend on the animal species, its age, its diet, and the plant residues that have been used as bed, among others. The concentration of undesirable elements such as salinity, excess of ammonia, as in chicken manure, heavy metals, or emerging contaminants derived from antibiotics in pig manure must also be taken into account, since may make them unsuitable for use.

Vegetable residues

Vegetable residues consist of the parts of the plants that remain after harvesting. It is an important source of organic matter and because they have a high concentration of carbon and are biologically active, although their C/N ratio is sometimes very high when their composition includes traces of lignin and cellulosic materials. Sometimes, even leaving pruning debris on the surface can increase soil aggregation and protect soil from the formation of crusts caused by the impact of rain and frosts that break up soil aggregates into fine particles. Although they normally present a low N or P content, they are good carbon sources for soils, encouraging the fixation of C in the soil.

Sewage sludges (Biosolids)

Among the possible sources of exogenous organic matter that could be part of organic amendments that can be applied to agricultural soils, are the sludges generated in urban wastewater treatment plants. They are produced in a large quantity and are a pasty material (normally their moisture content is about 70-80%, Murcia, 2013). Sewage sludges are commonly used as organic amendments in soil restoration as they contain macro and micronutrients, which are a good source of nutrients for plants, and organic constituents that provide beneficial conditioning properties for the soil. On the other hand, their incorporation into the soil allows the recycling of nutrients and eliminates or reduces the need to use commercial fertilizers in crops. The countries are legislating on the possible applications of sludges from urban wastewater treatment, with regulations that fundamentally include the treatments to which they must be subjected prior to their use in agriculture (aerobic or anaerobic treatment, thermal drying, even pyrolysis, etc...), to prevent its application to the soil from causing any harm to its health and biodiversity.

This type of regulation is essential, since the sludges generated in treatment plants can cause problems that prevent them from being added to agricultural soils as organic amendments. On the one hand, the low stability of its organic matter can pose a problem of competence for nutrients between plants and soil microbial populations, which could access to nutrients that will not be then, available to the plants (Murcia, 2013). Another problem is due to some pathogenic microorganisms such as salmonella, *Escherichia coli* or *Clostridium prefingens*, which can cause health problems. Also, their high concentration sometimes of heavy metals, which could pass at some point to the plant and the trophic chain. In this sense, Murcia (2013) points out that in recent years, the sludges generated in urban wastewater treatment plants has greatly decreased their content of heavy metals (Cd, Cu, Zn, Cr, Pb, Ni, Hg). In addition, sewage sludges can also contain harmful compounds such as detergents, heavy metals, salts, and pesticides due to effluents from municipal and industrial facilities, organic toxins (emerging pollutants sometimes derived from drug residues) and hormone disruptors, whose presence in the treated sludges will depend on its origin (urban wastewater or industrial water) (Garcia et al. 2017). For sludges generated in urban wastewater treatment plants, they are considering setting limits on the concentrations of some organic compounds such as LAS (alkylbenzene sulphonates), DEHP (phthalates), NPE (noniphenols), AOX (organic halogenated), PAH (polycyclic aromatic hydrocarbons) and dioxins; but this has not yet been established, and some countries have taken the initiative in this regard (Murcia, 2013). All of the above indicates that although the sludges from urban wastewater treatment have a good content of organic carbon, as well as N, P and micronutrients, they have various problems that must be considered to classify them as good organic amendments. for soils.

Green manure

Green manures correspond to the plant biomass produced by the sowing of some species of grass, legume or cruciferous or other plant species, which allows organic matter to be incorporated into the soil. Green manures allow the incorporation of organic matter and other nutrients used by plant material. The grass species favour the formation of soil structure, while the legumes incorporate nitrogen due to the effect of symbiotic fixation, which allows enriching the plant tissues with N, and by direct decomposition of the nitrogen-rich nodules. It is an economic organic matter.

Biochars

They come from the pyrolysis carried out on organic residues of different origin. They have a high stable C content, which will undoubtedly benefit soil fertility, and the sequestration of C in soils. The characteristics and action of biochar will depend, not only on the residue from which the biochar comes from, but also on the pyrolysis technique itself, and specifically on the pyrolysis time and temperature. Bolan et al. (2021), in a study on the capacity of using biochar to store C in the soil observed that depending on the pyrolysis temperature (from 500-600 degrees, up to >1000 degrees), biochar with different quality of C and resistance to being mineralized was obtained. In agriculture, the potential of biochar is given by the increase in the availability of nutrients in the soil, motivated by an improvement in the soil cation exchange capacity (Liang et al. 2006), soil structure and porosity, which favors the microbial habitat and, therefore, increases the biological activity. Biochar also increases the water available to plants (Diatta et al. 2020), and environmental benefits are obtained such as reducing nutrient doses or minimizing their loss through leaching. In general, Spanish agricultural soils, with very low organic carbon content, are good candidates for the extensive application of biochar. Soil current and potential fertility can be improved, as well as its capacity as a carbon sink in the context of the fight against climate change.

Composts

Within current agriculture, and possibly future agriculture, looking towards sustainability, is the use of compost as an organic amendment. Through the composting process, it is possible to transform a fresh organic waste into one (compost) that is biologically more stable, and also sanitized, free of pathogenic microorganisms. Composting is an inexpensive, well-known and well-studied process. It is a biooxidative and aerobic process, where the microorganisms existing in the organic mass to be composted, are those that carry out the process: An adequate aeration of the material is needed, as well as a humidity adapted to the microbial action. The organic material to be composted goes through a mesophilic phase (<42º C), followed by a thermophilic phase (about 60-65ºC), thus achieving the incorporation of different microbial populations adapted to those temperatures, as well as the sanitization of the material (pathogenic microorganisms, as well as weed seeds are destroyed). At the end, a maturation stage at room temperature consolidates a very good stabilization of the organic matter. In particular, composting is very suitable for the stabilization of the organic fraction derived from domestic waste. It is also usable with manure, sludge, or other organic wastes, to which may be advisable to add a structuring agent before composting to facilitate aeration. The product of the composting process is the "compost", similar in appearance to humus and with little resemblance to the original material. It is a product stabilized by biological processes, easy to handle, store and odourless.

Digested from organic waste-methanisation processes

Anaerobic digestion consists of a process where it is possible to degrade the organic matter anaerobically, obtaining on the one hand biogas rich in methane that can be used as a source of thermal or electrical energy and, on the other, the residue of the digestion (digested) with the appearance of semi-liquid sludge. They are organic wastes from several origins (agricultural wastes, organic manures, urban organic wastes such as domestic wastes and sewage sludges) after biomethanisation processes. This type of material is becoming more and more common, which, depending on the biomethanisation system (mesophylic or thermophilic), and of the initial organic material, will have more or less nutrients and C content, and hygienisation level (absence of pathogens and phytotoxic compounds). The direct application of the digested to the soil from an agronomic point of view is currently the most used application in Europe for the management of the digested. This is a well-considered application when treating a digested with properties that make it a good candidate as an organic amendment and with potential value as a fertilizer due to its NPK content. The benefits that the digested provides to the soil and crops have been demonstrated, although its direct application must always be integrated into a fertilization program and follow a code of good agricultural practices (Arthurson et al. 2011).

Effects on soil derived from the use of organic amendments

Improving productivity of degraded soils is key for achieving sustainable food production. As already indicated, the use of organic amendments in agriculture constitutes a common and useful strategy to improve the fertility, health, and edaphic productivity of soils, in a rational and environmentally appropriate way, and benefit its biodiversity (Urra et al. 2019). However, it must be taken into account that the application of organic amendments to agricultural soils can also bring some risks, because there are amendments that may contain contaminants such as heavy metals, pathogenic microorganisms, or even emerging pollutants derived from pharmaceutical residues, persistent pesticides, or microplastics (Pampuro et al. 2017, Asgharipour et al. 2016). Undoubtedly, since the use of organic amendments in agricultural soils is a good strategy to improve productivity and soil health, there are many studies aimed at mitigating the possible risks associated with the use of organic amendments in agriculture (Urra et al. 2019).

Therefore, it should be noted that when we talk about the addition of organic amendments to agricultural soils as a strategy to maintain and improve their fertility, we always refer to the use of quality organic amendments, with stable organic matter and free from pathogens and contaminants.

From a general perspective, many of the organic amendments used in agriculture today (different origin, different level of stabilization) are composed of a chemical part (from carbon and its structure to its content in nutrients and chemical elements), and of a "living and organic" part formed by the set of microbial population inherent to the amendment itself. Both parties will interact with the recipient soil, depending on both the characteristics of the amendments, and the doses tested, as well as the type of crop, also including seasonal variations. All this, together with the complexity of the "soil" matrix, highlight the difficulty of generalizing about this type of strategy or management. Despite this, there are evidence in favour of incorporating organic amendments into agricultural land, which indicate the benefit of this strategy, and which are now exposed.

The addition of organic amendments to agricultural soils has been shown to improve the physical, chemical, and biological fertility of the soil and to reduce soil pathogens (Hernandez et al. 2014, Chocano et al. 2016) (*Figure 2*). They improve the physical fertility of the soil, mainly by improving the stability of the aggregates, porosity, and the soil water holding capacity (Mabuhay et al. 2006, Hernandez et al. 2015, 2017).

Abiven et al. (2009) indicated that the effect of the easily biodegradable amendments on soil aggregates is intense but transitory, while amendments more resistant to degradation, such as ligno-cellulosic ones, have a less intense effect, but more durable.

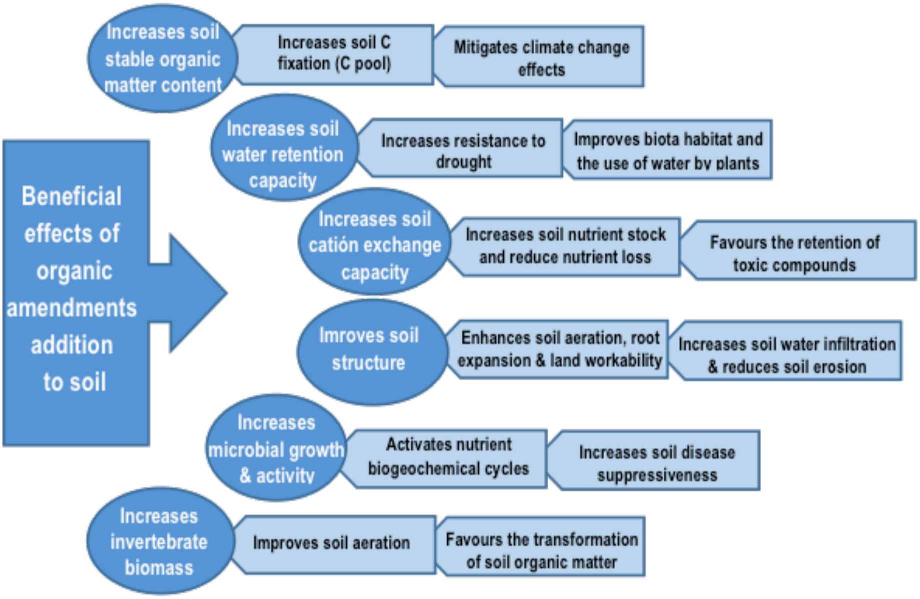

Figure 2. Addition of organic amendments to soils.

CUnagwu (2019) investigated the effects of four organic amendments on the physical, chemical, and biological soil quality indicators (SQIs) of a degraded sandy loam soil. He indicated that organic amendments increased Olsen P and available K compared to control soil (unamended), as well as microbial biomass C and total organic C.

Sulok et al. (2021) in an assay of pepper culture in soils with organic amendments (biochar and compost), observed a positive effect on physical (bulk density and porosity) and chemical (total organic C and available P) soil parameters, and even, on parameters such as soil respiration and abundance of microorganisms (bacteria, fungi and actinomycetes). They also noted that crop yields were achieved with organic amendments, similar to those obtained with mineral NPK. This suggests that the application of organic materials, compared with the use of synthetic fertilizers, have an immediate positive effect on soil fertility and microbiota, while the increase of crop productivity is of longer-term nature.

Readyhough et al. (2021), in a study on tomato cultivation using different manure amendments concluded that organic amendments, apart from improving some soil physical properties such as porosity and water holding capacity affected the microbial community of the rhizosphere (bacteria and fungi), particularly fungi. This fact confirms that organic amendments affect plant yields, due both to their involvement of the microbial community, and to the positive effect on the physical properties of the soil.

In addition, other studies (Gomez et al. 2006) have shown that the addition to agricultural soils of manure-based organic amendments, cause an increase in the bacteria and fungi abundance and activity. This is particularly due to the increased activity of bacterial populations, with fungal populations less responsible for this effect. The carbonaceous substrates incorporated with the amendments, and other nutrients, allow to increase the microbial biomass, and the breathing of the microbial populations. The addition of organic amendments not only influences an increase in microbial biomass (organic material is added), but the amendment causes changes in the structure of the soil microbial community, also increasing diversity (Jangid et al. 2008). But not only do organic amendments have a direct impact on the microbial populations in the soils where they are applied, but the enzymes, especially hydrolytic enzymes involved in the C, P and N cycles of that soil, also play a key role in the decomposition of soil organic matter and nutrient control, and therefore affect the quality and health of soils, and their functionality (Garcia et al. 2017). This is because most enzymes are extracellular and are produced by micro-organisms.

There are experiments where they have combined managements, specifically the incorporation of organic amendments, along with tillage. Treonis et al. (2010), in an experiment on crop rotation, where organic amendments, buried (tillage), and not buried (no-tillage), were introduced into the topsoil layer, found a positive effect on the organic matter of the receiving soil, abundance of protozoa, soil microbial respiration and PLFAs of bacteria and fungi.

Calbrix et al. (2007), in a comparative study of the effect on the soil microbial community, of various organic amendments (sewage sludges, manure, and compost) and mineral fertilization, on a mustard-beet crop, indicated that organic amendments impact on the soil's potential metabolic activity, on its genetic structure, and on the amount of soil microorganisms. The study was conducted at three different seasons, and they concluded that organic amendments affect microbial populations in a general way, although seasonality is even more important than the different organic amendment used.

All this reflects the difficulty of generalizing on the effects of organic amendments in soils, especially those where populations of soil microorganisms are involved, since aspects such as seasonal variability itself plays a decisive role.

Scotti et al. (2015) carried out a review of the effects of organic amendments on soils where intensive farming was applied in crops under plastic. The interest of this work is due to the fact that this type of intensive agriculture induces a loss of organic matter by mineralization, due to soil conditions under plastic, which leads to a decrease in soil fertility. Therefore, the introduction in the system of organic amendments is considered of interest, to recover the lost organic matter. The authors see positive the use of organic amendments in this type of agriculture, to improve soil fertility and soil health, and showed that this effect will depend to a large extent on the organic matter used as an amendment. They also point out that more research is needed to identify the best organic amendments, both single and mixed, in order to maximise their benefit on agricultural soils, and their future sustainability.

Different studies have also shown that the application of organic amendments is an effective strategy to recover soil organic C stock, helping to mitigate climate change and avoiding land degradation and C losses by erosion and other degradative processes. Degraded soils present a low microbial activity that will negatively affect their quality, fertility and natural productivity, since the biogeochemical cycles of the important elements in the soil develop in degraded systems with great difficulty, as manifested by the low values of enzymatic activities detected in them. The incorporation of organic amendments into soils will produce a reactivation of its microbiological and biochemical properties, stimulating microbial proliferation and metabolic activity, as a consequence of the contributions of new labile sources of carbon that will serve as a substrate for the soil biota (Hernandez et al. 2015). This increase, in turn, results in an increase in the enzymatic activity of certain enzymes, in particular hydrolases, involved in biogeochemical cycles of interest in the soil, as well as in some metabolites in the soil.

This fact is particularly important with enzymes such as phosphatases, which are favoured with the entry of organic amendments to the soil, and which allow the hydrolysis of organic phosphorus to inorganic, which can be assimilated by plants. Compost of organic waste stimulates microbial activity in soils, increasing the mineralization of organic matter, thus supporting the growth and activation of microorganisms (Scotti et al. 2016, Hernandez et al. 2018).

Controversy exists as to whether or not the microbial community present in the exogenous organic matter incorporated into the soil will alter the native microbial community of said soil. According to Saison et al. (2006), the intrinsic microbial activity of the amendment joins the activity of the original soil population with a minimal impact on the native microbial community. On the contrary, for Ondoño et al. (2014) the incorporation of exogenous organic matter to the soil matrix can modify its microbial community by incorporating a considerable number of microorganisms linked to it (fungi and bacteria), which can result in a competition or antagonism with those already existing on the soil.

As described above, organic amendments are widely used as an adequate strategy for the restoration of soil organic matter levels and the improvement of soil quality and fertility in arid and semiarid ecosystems. However, the effect of soil restoration using organic amendments on the structure, composition, and diversity of soil microbial communities is barely known. Certain studies have found no significant changes in the structure of the bacterial communities of soils subjected to organic amendment (Crecchio et al. 2009). Conversely, other studies did find a change in the community structure after amendment. For instance, Cytryn et al. (2011) found that members of the *Bacteroidetes* proliferated in compost-amended soil. Precisely, *Bacteroidetes* are usually categorized ecologically as copiotrophs and are more abundant in soils with high carbon availability (high C mineralization), indicating that this phylum plays an important role in decomposing organic materials in soil (Fierer et al. 2007). Similarly, Bastida et al. (2013) observed that the abundance of Bacteroidetes was also correlated to the level of organic carbon after long-term restoration (25 years) in Southern Spain. These authors concluded that vegetation development after soil restoration in the long-term (25 years) induced changes in the phylogenetic structure of the bacterial and fungal communities but without a significant influence on microbial diversity. In South-West Sydney (Australia), Ge et al. (2010) observed that the effect of the rate of organic wastes on the soil bacterial community varied with the type of waste.

Long term field study is recommended to further evaluate the effects of these organic amendments on the overall soil health, including soil microbial populations. In this sense, in a long-term experiment, Zanen et al. (2008), carried out a field experiment with several organic amendments with a completed rotation of seven years. Yields suggest significant differences due to different organic amendments after seven years. The study shows that organic amendments used within the legal framework of organic farming may impact soil fertility and biodiversity indicators (soil microorganisms) within seven years.

Bonilla et al. (2012) carried out a study on avocado crop and they indicate that organic amendments affect soil properties in one or more of the characteristics that were analysed. In addition, molecular analysis of soil bacterial communities by DGGE allowed the detection of the influence of all the analysed amendments on bacterial community composition. This effect was stronger in the superficial layer of the avocado soil. Rahman et al. (2017) indicated that poor soil structure is a serious problem of soils which makes it difficult for agricultural workability and crop growth. Straw amendment significantly enhanced soil respiration, microbial biomass carbon (MBC), and phospholipid fatty acid (PLFA), and hence improved aggregation ($P < 0.05$). But biochar from this straw had no impact on soil microbial activity as well as on soil aggregation in comparison to the control ($P > 0.05$) due to its recalcitrant nature. This study showed that straw amendment technique has the potential to be an option to improve soil structure by promoting fungal abundance.

Gravuer et al. (2019) drive a study from Global Change Biodiversity; a global meta-analysis of 92 studies in which organic amendments were added to arid, and semiarid soils. They found that organic amendments, on average, provide some environmental benefits (decreased runoff, increased soil carbon, soil water holding capacity, aboveground net primary productivity, and plant tissue nitrogen), as well as some environmental harms (increased concentrations of soil lead, nitrate, and phosphorus runoff, increased soil CO_2 emissions). This study suggests that trade-offs among outcomes are inevitable; however, applying low-N amendments was consistent with both maximizing benefits and minimizing harms.

Luo et al. (2018) after a meta-analysis of 106 peer-reviewed papers, concluded that “organic amendments can improve microbe-mediated soil ecosystem functioning, long-term soil fertility and crop productivity, relative to mineral fertilization, on a global scale”.

The use of organic amendments such as composts can also be an efficient tool in the control of different plant diseases caused by soil-borne pathogens. This suppressive effect of organic amendments is attributable to the increase in total microbial biomass and activity caused by the amendment and has commonly been related to a general suppression mechanism, in which all the microorganisms contribute to create adverse habitat condition for the pathogens (Bonilla et al. 2012). However, there is also evidence of a specific suppression mechanism in which a particular group of microorganisms are involved, affecting pathogen growth by competition for resources, direct attack (microparasitism) and/or antibiotic production.

Conclusions

Future agriculture must apply sustainable management criteria that can benefit soil health and biodiversity. Its contribution in the increase of stable C in soil, is an attraction for such “smart” agriculture to adapt to climate change, and also contribute to mitigate its negative effects. The variety of microorganisms that live in the soil, and in particular the bacteria and fungi that make up its microbial biodiversity, are responsible for many of the processes that accredit the life and sustainable functioning of the soils, increasing their fertility and productivity. Managing the microbiological biodiversity of the soil, in favour of agricultural productivity in the future, is a challenge, but there are problems that must be solved, such as methodological ones, integration and harmonization of databases, etc. Our view is that, if we want to have sustainable agricultural systems, particularly in large areas where climate change is causing serious soil degradation problems, and in particular, high decreases in the soil organic matter content, and therefore, loss of productivity, fertility and biodiversity, the option of getting agroecosystems, where organic "quality" amendments have a leading role, can be a key strategy to increase organic carbon, improve soil health, and positively influence microbial biodiversity. To improve our knowledge on the different organic amendments that we can use in sustainable and future agriculture, anticipating their effects on the receiving soils, and in particular on their microbial biodiversity and their health, is essential to ensure that the strategy of adding exogenous organic matter to those soils is truly useful in increasing their ecosystem services, both to improve their health and biodiversity, and to improve their role in C sequestration, thus mitigating the effects of climate change. Governments, agricultural producers, researchers, and other stakeholders should channel their collective resources towards strengthening investment and legislation in sustainable soil management practices.

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Chapter 11

Food & nutrition security and soil security: the soil biodiversity connection

Ayoade Olayiwola Ogunkunle and Victor Okechukwu Chude

Introduction

Agriculture faces a three-fold challenge today: The production of safe and nutritious food will need to increase to meet the growing demand and ensure food security for all. The sector has to generate jobs and incomes and contribute to poverty eradication and rural economic growth. Furthermore, it has a major role to play in ensuring the sustainability of natural resources and in combating climate change.

As the world's human population continues to rise and is expected to reach 8.6 billion by 2030, 9.8 billion in 2050 and 11.2 billion in 2100 (UN, 2017), it is paramount to respect and protect natural resources, including soil, water, air, minerals, and biodiversity that support life on earth, including humanity. Currently, it has been estimated that inadequate management of resources are putting unprecedented pressure on global systems, and it is estimated that one to six billion hectares (up to 30%) of land has been degraded globally (Gibbs and Salmon, 2015). Land degradation negatively affects 3.2 billion people, threatens sustained human well-being and is a major contributor to climate change and biodiversity loss (IPBES, 2018). Global initiatives to meet these challenges include the UN 2030 Agenda for Sustainable Development and the UN Convention on Biodiversity's Strategic Plan for 2020 (Bach et al. 2020). Generally, these agendas address areas to improve human life and environmental sustainability, they rely on the participation of all countries and stakeholders, and will require innovative, timely, and interdisciplinary approaches.

Seven existential challenges facing human survival were identified by Bouma and McBratney (2013), they emphasized the need to understand these challenges interdependently, rather than as separate processes. Of the identified challenges, Food Security is unarguably the most widely explored, followed by climate change, water security, energy sustainability, biodiversity protection and ecosystem service delivery. Soil Security is a lesser-known challenge, not widely discussed beyond the soil science discipline; yet without soil we would not have food, water, ecosystems, or biodiversity (Holt et al. 2016; Wall and Nielsen, 2012).

Soil is home to more than 25% of the earth's total biodiversity and supports life on land and water, nutrient cycling and retention, food production, pollution remediation, and climate regulation. Accumulating evidence demonstrates that multiple sustainability goals can be simultaneously addressed when soil biota are put at the center of land management assessments because the activity and interactions of soil organisms are intimately tied to multiple processes that ecosystems and society rely on. With soil biodiversity at the center of multiple globally relevant sustainability programs, we will be able to more efficiently and holistically achieve the Sustainable Development Goals and Biodiversity Targets.

Soils are central to supporting natural systems and human well-being (Soliveres et al. 2016). This is well captured in Figure 1, yet to date soil biodiversity —the diversity of life in soil which drives ecosystems, sustains life aboveground, and maintains healthy landscapes— has remained largely overlooked in global agendas. For example, the term 'soil biodiversity' does not appear in any UN documentation while forests, wetlands, rivers, and dry lands have received specific attention to their benefit. Soil-dwelling organisms, including bacteria, fungi, nematodes, earthworms, moles, and even plant roots, contribute the majority of living biomass on Earth (Bar-On et al. 2018) and represent more than 25% of all described species (Decaëns et al. 2006, Bardgett and Van der Putten, 2014) not to mention the genetic diversity represented within these species. The activity and complex interactions of soil organisms provide the backbone for many ecosystem functions, including nutrient cycling, pathogen control, water infiltration, foundations to food webs, and supporting agroecosystems. Soil is one of the components of land, an important part of the ecosystem and all the elements of biodiversity occur within or on the top of the soil. In fact, soil is formed from the interaction between climate and living organisms (biota or biodiversity including man) on the parent rock as conditioned by the landscape over a period of time. Thus, biodiversity is involved in soil formation and continues to sustain the soil in its functions for food production such as water and solute flow, filtering and buffering, nutrient cycling, and structural support (Seybold et al. 1998). Here lies the importance of soil biodiversity to humanity. It is crucial for Soil Security and Food and Nutrition Security.

Figure 1. Role of Soils (security) in Water and Food Security (Modified from Mohtar, 2015)..

This chapter lays emphasis on the unique importance of soil biodiversity on ecosystem functions and its impact on food and nutrition security by bringing together relevant information on its contributions to sustainable development through soil security. We focus on the need to see soil biodiversity as the nexus between Soil Security (Sustainable Soil Management) and Food and Nutrition Security (The outcome of Soil Security) without which neither of them can be achieved and hence Sustainable Development may be a mirage.

Food and nutrition security

In order to provide enough food for the growing world population, global food production will need to rise by about 60% by 2050. Food security is a flexible concept as reflected by the many attempts to define it in research and policy usage. The concept of food security originated some 50 years ago, at a time of global food crises in the early 1970s. Even two decades ago, there were about 200 definitions for food security in reviewed articles (Maxwell and Smith, 1992), showing the contextual dependent features of the definition. The current widely accepted definition of food security came from the Food and Agriculture Organization (FAO) annual report on food security: “The State of Food Insecurity in the World 2001”: Food security is a situation that exists when all people, at all times, have physical, social and economic access to sufficient, safe and nutritious food that meets their dietary needs and food preferences for an active and healthy life (FAO, 2002). The last revision to this definition happened at the 2009 World Summit on Food Security which added a fourth dimension – stability – as the short-term time indicator of the ability of food systems to withstand shocks, whether natural or man-made (FAO, 2009).

Dimensions of Food and Nutrition Security

Food Security is becoming more challenging over time due to changing supply and demand, pushed by a growing population, changing climate and environmental conditions (Hwalla et al. 2016). For a nation, community, or individual to possess security in food and nutrition, they must have access to suffcient safe and nutritious food at all times to facilitate an active and healthy life (FAO, 2009). Originally known as ‘Food Security’, the term has been expanded to include nutrition, because both quality and quantity are important (Hwalla et al. 2016).

Food and Nutrition Security possesses dimensions: *availability, access, utilization,* and *stability*. These categories have clear definitions, but they are hard to assess, so development of more consistent assessment is needed to make it easier to compare and identify change in security status and facilitate more rapid identification of problems as they arise.

(i) ***Availability:*** This is the physical existence of suffcient quantity of food of appropriate quality (Hwalla et al. 2016). This dimension of Food Security is directly related to agricultural production, management and availability of land to support crop production.

(ii) ***Access to food:*** This means adequate income or resources to acquire safe and nutritious food (Leroy et al. 2015). The food can reach the consumer (transportation infrastructure) and the latter has enough money for purchase. To such physical and economic accessibility is added socio-cultural access to ensure that the food is culturally acceptable and that social protection nets exist to help the less fortunate. Access may either be through markets (indirect) or by home production (direct) and is controlled by food affordability and availability, respectively (FAO, 2019).

(iii) ***Utilization:*** This is defined by how humans use food, including how the body uses food and absorbs nutrients, food choices, preparation and hygiene (Gross et al. 2000). The individual must be able to eat adequate amounts both in quantity and quality in order to live a healthy and full life to realize his or her potential. Food and water must be safe and clean, and thus adequate water and sanitation are also involved at this level. A person must also be physically healthy to be able to digest and utilize the food consumed. Both undernutrition and obesity are gaining in importance, stemming from the inability to receive the right amount of food and nutrition (Fanzo, 2014).

(iv) ***Stability:*** Stability refers to the security of availability, access and utilization, and their ability to be maintained throughout changes and conflict. It is the ability of the nation/community/household/person to withstand shocks to the food chain system whether caused by natural disasters (climatic, earthquakes) or those that are man-made (wars, economic crises). To safeguard against environmental, social, and political change, global policies are needed. The UN Sustainable Development Goals (SDGs) brings Food Security to the forefront, provides guidelines and standards to encourage and raise awareness of global challenges (UN General Assembly, 2015).

Soil security

Over the last 30 years the world's crop area has expanded by some 5 million hectares annually, with Latin America alone accounting for 35% of this increase. The pool of unused, suitable cropland is unevenly distributed, with greatest current potential for arable land expansion in South America and, to a lesser extent, southern Africa (Fischer et al. 2002). While FAO projections (FAO, 2002) indicate that most gains in production (around 90%) will be achieved by raising productivity through increasing yields and cropping intensity, it is still expected that the area of arable land will be increased by around 70 million hectares globally. An increase of around 120 million hectares in developing countries (responsible for an expected 21% of growth in crop production in these areas between 2005 and 2050) will be partially offset by a decline in arable land of some 50 million hectares in developed countries, resulting in a lower per capita decline of arable land in developing countries, than would be expected when looking at the population growth alone (Bruinsma, 2009).

The concept of Soil Security is relatively new and was first communicated by Koch et al. (2012). Soil security refers to the maintenance and improvement of the world's soil resources so that they can continue to provide food, fiber and fresh water, make major contributions to energy and climate sustainability, and help maintain biodiversity and the overall protection of ecosystem goods and services (Koch et al. 2012). It is the ability of soil to sustain functions to provide planetary services and human wellbeing. Soil security is a new concept that has arisen during a time of emerging international response to the increasingly urgent problems that face the global soil stock. Soil Security is a lesser-known challenge, not widely discussed beyond the soil science discipline; yet without soil we would not have food, water, ecosystems, or biodiversity (Holt et al. 2016; Wall and Nielsen, 2012). It is more holistic than land suitability or land quality (Blum and Eswaran, 2004; Klingebiel and Montgomery, 1961) with capability/suitability classes defined on a number of land characteristics/qualities. The land space in the world is finite (fixed and cannot be expanded), so also is the amount of land available in every nation. In addition, the rapid and continuous expansion in population resulting in urban development, industrialization and other non-agricultural uses which are competing with agriculture for the dwindling available land. This underscores the need to secure the soils of the land allocated to agriculture in terms of quality and sustainable management.

Dimensions of soil security

History stands as a warning to our modern societies. Whole civilizations have fallen and collapsed when their stock of fertile soils washed or blew away. Many clarion calls to preserve our soil stocks have been made; (Jacks and Whyte, 1939; Hyams, 1952; Kellogg, 1956; Hillel, 1992; Montgomery, 2007), they must not be ignored. Soil is multifunctional, the functions include nutrient cycling, habitat and biodiversity, carbon sequestration, bioremediation, filtration and buffering, physical stability and support and hydrological cycle. Hence, assessment is needed to understand the ability of the soil to perform these functions. Dimensions of Soil Security were conceptualized to provide the means for assessment. The practical measure of soil security is expressed by Food Security, particularly in terms of two of the dimensions: Availability and Stability.

Increased and intensified agricultural production has pushed the soil to its limits in many regions around the world, resulting in degradation and ultimate loss of agricultural land. Soil degradation may result from loss of soil structure, chemical imbalances (e.g., salinity), nutrient loss, or loss of soil altogether by erosion (Lal, 2001). As soil is a finite resource, many food production regions are at risk of losing this, due to mismanagement resulting from lack of knowledge, or lack of care. Building knowledge and care is essential for securing soil, but it cannot be achieved without understanding the inherent link and interactions between Soil and Food Security.

These dimensions pertain to the physical aspects of the soil, e.g., Condition and Capability, as well as socioeconomic factors influencing those physical aspects, e.g., Connectivity, Capital, and Codification (McBratney et al. 2014). The dimensions were chosen to be measurable so that they may provide the means for assessment, comparison, and a reliable and accurate estimation of Soil Security. Formal Soil Security assessment is gaining traction. Studies have been undertaken in Tasmania as part of land suitability assessment (Kidd et al. 2015), followed by spatial exploration of Soil Security (Kidd et al. 2018). Soil security assessment has also been applied in Papua New Guinea for cocoa production (Singh et al. 2019), and in China for agriculture (Yang et al. 2018). Furthermore, the study by Bennett et al. (2019) has set out provisions for the implementation of an Australian Soil Security Framework, enabling other countries to follow suit and bring Soil Security policy to the forefront.

- ***Capability:*** This refers to the physical potential of the soil, similar to the suitability class in land evaluation. The assessment of soil capability aims to identify the optimal biophysical state of the soil (characteristics) for a given use, or what the reference state of the soil is (McBratney et al. 2014, Kidd et al. 2018). For optimal production we need to match the land use with its capability, asking "what can the soil do?" Capability is evaluated by observing soil properties, such as texture, structure, consistence, CEC, hydraulic conductivity, bulk density etc. and comparison of these with another soil of the same type, but one that is in a more natural state (McBratney et al. 2017). Capability is the potential of the soil for a given purpose.

- ***Condition:*** We can think of condition as the 'health' of the soil for crop production– its current state rather than its reference state. The condition of the soil is influenced by land management practices and land use; if the activities do not match the capability of the soil, degradation will occur, and the condition will decline. The soil health indicators used to evaluate soil condition include nutrient content, pH, biodiversity and organic carbon content i.e. properties that are prone to change due to use and/ management (Field and Sanderson, 2017). Soil is resilient and it is possible for the condition to improve, but only to a certain extent. Care must be taken to prevent human activities and land management pushing the condition of the soil beyond tipping points of recovery. This can occur due to the pre-compression stress resulting in change from elastic to plastic soil deformation leading to irreversible changes in soil functions (Horn and Blum, 2020). In order to avoid this, sustainable land/soil management practices such as was suggested by Ogunkunle and Chude (2018) must be adopted.

- ***Capital:*** Land is the real wealth of a nation as it is basic to human existence. Placing value on the soil improves the likelihood of it being included in decision/policy making (Robinson et al. 2009). Soil capital may refer to financial, manufactured, human, social or natural forms (Kidd et al. 2018, McBratney et al. 2014). Soil natural capital is the stock of natural assets which provide resources or ecosystem services, and so to achieve sustainable development, natural capital may be valued through the ecosystem services the soil provides (Dominati et al. 2010). Ecosystem services and natural capital are often undervalued, decreasing the consideration of the resources in decision making (Costanza et al. 1997), thus degrading the condition of the soil.

- ***Connectivity:*** This dimension refers to how humans relate to soil and extends beyond monetary value. Obviously, people who appreciate the value (usefulness) will treat it well. The relationship can be grouped into those who *know* and understand soil, those who *know of* soil, and those who are *aware of* the existence of soil. The move into urban areas and mega cities is increasing the gap between humans and soil (FAO and ITPS, 2015). Despite its importance, connectivity is one of the lesser explored Soil Security dimensions and further exploration is needed in defining connectivity (McBratney et al. 2014), evaluating it, and understanding what makes people care.

- ***Codification:*** To ensure continued provision of goods and services while maintaining the condition of the soil, there is a need for education, policy, and regulation (McBratney et al. 2014). Stronger global and national policy will bring soil to the forefront, allowing greater weighting in decision making processes and stronger guidance on soil management. Policy integrating Soil Security is gaining traction, and a key aspect to successful policy development is through cooperation between soil scientists and policymakers (Hill, 2017). Proper codification, including clear and candid communication with stakeholders, is essential to link science with society, a link that needs improvement.

Soil biodiversity

Unsustainable farming practices are associated with a decline in species diversity, driven by conversion of natural habitats for food production as well as by pollution. The impact on biodiversity is amplified along food supply chains through energy use, transport and waste (Dudley and Alexander, 2017). A major contributor to biodiversity loss from the food and agriculture sector is intensification and the increased application of agrochemicals, including pesticides and fertilizer. Ecosystem functions and services frequently decline in parallel with biodiversity loss, including decline in water supply, water quality, clean air, and climate regulation. The global loss of ecosystem services due to land use change has been estimated to cost US$ 4.3-20.2 trillion per year (Costanza et al. 2014).

One factor that cuts across Sustainable Soil Management, Food and Nutrition Security and Soil Security but has not been given proper consideration until recently is Soil Biodiversity. Most of the biodiversity of agricultural systems resides in the soil. Food web interactions among the soil biota (including plant roots) have large effects on the quality of crops (affecting human and animal nutrition or other utility), the incidence of soil-borne plant and animal pests and diseases (affecting production levels), and the beneficial organisms that, e.g., cycle nutrients or are predators of the pest species. This is because the availability of water, nutrients and certain microorganisms at the root surface is mediated by such interactions. Farmers manage, consciously or inadvertently, soil biodiversity in the face of unpredictable disturbances and stress agents.

Despite the clear importance of soil biodiversity in the provision of essential ecosystem services (provision of food, fiber and fuel, filtering of water, source of pharmaceuticals, carbon and nutrient cycling, soil formation, GHG mitigation, pest and disease control, decontamination and remediation), its proper use and management is not up to scale. It is only just over a decade ago that initiatives and research networks were established to contribute to the knowhow, conservation, use, and sustainable management of soil biodiversity. These include the establishment of the International Initiative for the conservation and Sustainable Use of Soil Biodiversity in 2002, the establishment of the Global Soil Biodiversity Initiative in 2011, the Global Soil Partnership in 2012, and the publication of the Global Soil Biodiversity Atlas by the European Commission in 2016 (Orgiazzi et al. 2016).

There is increasing attention on the importance of biodiversity for food security and nutrition, especially above-ground biodiversity such as plants and animals. However, less attention is being paid to the biodiversity beneath our feet, soil biodiversity. Yet, the rich diversity of soil organisms drives many processes that produce food or purify soil and water. In 2002, the Conference of the Parties (COP) to the Convention on Biological Diversity (CBD) decided at its 6th meeting to establish an International Initiative for the Conservation and Sustainable Use of Soil Biodiversity and since then, the Food and Agriculture Organization of the United

Nations (FAO) has been facilitating this initiative. In 2012, FAO members established the Global Soil Partnership to promote sustainable soil management and increase attention to this hidden resource. The Status of the World's Soil Resources (FAO and ITPS, 2015) concluded that the loss of soil biodiversity is considered one of the main global threats to soils in many regions of the world. Soil biodiversity and the ecosystem services it provides will be critical to the success of the recently declared UN Decade on Ecosystem Restoration (2021-2030) and the upcoming Post-2020 Global Biodiversity Framework. Soil biodiversity could constitute, if an enabling environment is built, a real nature-based solution to most of the challenges humanity is facing today, from the field to the global scale. Therefore, efforts to conserve and protect biodiversity should include the invisible array of microorganisms that make up more than 25% of the total biodiversity of our planet.

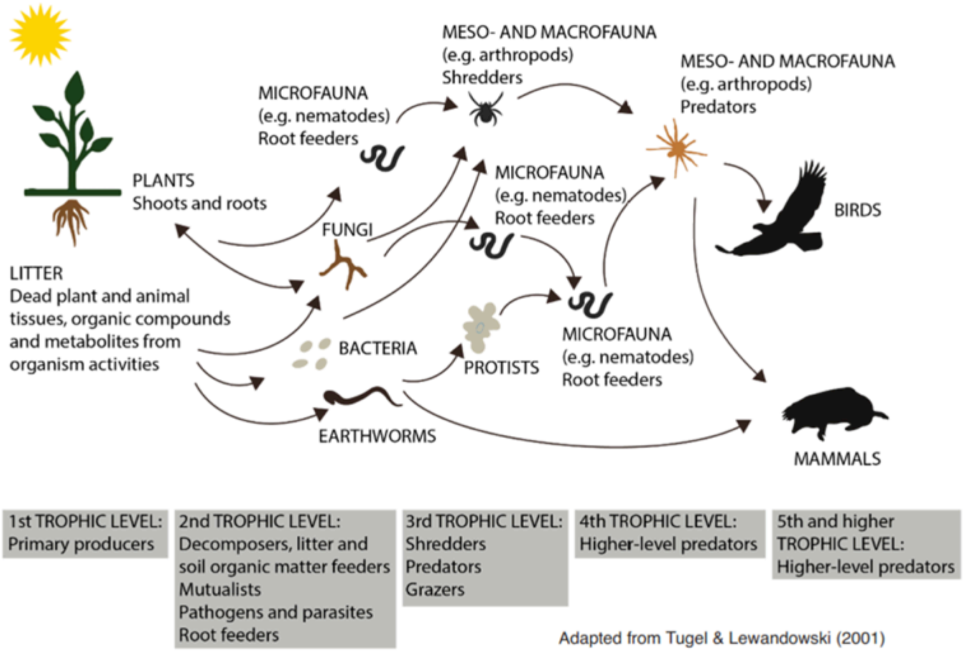

Figure 2. Food webs formed by soil biota complex for sustaining ecosystem functions. (Source: Larbodière, et al. 2020; adapted from Tugel and Lewandoswki, 2001).

Most of the biodiversity of agricultural systems resides in soil. Soils are one of the main global reservoirs of biodiversity, more than 40% of living organisms in terrestrial ecosystems are associated during their life cycle directly with soils (Decaëns et al. 2006). Soil biota interacts and forms complex food webs that support a range of ecological processes as shown in *Figure 2*. These processes are essential to maintain a functional soil ecosystem and are therefore essential for soil health (Security). Food web interactions among the soil biota (including plant roots) have large effects on the quality of crops (affecting human and animal nutrition or other utility), the incidence of soil-borne plant and animal pests and diseases (affecting production levels), and the beneficial organisms that are responsible for e.g., nutrient cycling or predators of the pest species. This is because the availability of water, nutrients and certain microorganisms at the root surface is mediated by such interactions. Four challenges are driving research on soil biodiversity and agricultural sustainability.

(i) **Understanding the "importance" of soil biodiversity for the resistance and resilience against stress and disturbance:** The relationships between soil biodiversity and ecosystem functioning are not straightforward (Brussaard et al. 1998). Hence, the insurance hypothesis (Yachi and Loreau, 1999) which suggests that ''high'' biodiversity confers an insurance against ecosystem malfunctioning under stress or disturbance, needs urgent and serious attention.

(ii) **Understanding the "importance" of soil biodiversity for the sustainable use of soil resources:** Soil biodiversity includes resistance and resilience, and this is related to efficient use of natural resources, such as water and nutrients. This holds promise for relieving pressure from agriculture on natural areas in agricultural landscapes and beyond, and even for providing habitats for species with conservation value from ''natural'' areas. Soil micro-, meso- and macrofauna play a key role in the physical breakdown of plant residues, allowing the soil microorganisms to liberate the nutrients and energy bound up in the plant material. The role of soil organisms in agriculture has many beneficial effects beyond plant nutrition. For example, soil microbiota such as arbuscular mycorrhizal fungi and nitrogen fixing bacteria can minimise cost and dependence on synthetic nitrogen fertilizers in agriculture, and enhance soil fertility and environmental sustainability, including reducing greenhouse gas emissions from the energy-intensive manufacture of nitrogen fertilizer.

(iii) **Managing soil biodiversity:** Whereas aboveground biodiversity is widely managed by choosing livestock and livestock breeds, crops, their varieties, rotations and sequences as well as the botanical composition of field margins and non-productive elements in agricultural landscapes, in most cases soil biodiversity can only be managed indirectly and the options for such management are less evident. The rhizosphere is the narrow region of soil directly influenced by root secretions/exudates and associated microorganisms' interactions. Leguminous plants receive nitrogen, a limiting nutrient in many soils in the form of ammonia, thanks to rhizobium, an N fixing bacteria that form nodules in the plants' roots. In return, Rhizobium receives nutrients and habitat from the roots. Mycorrhizae (symbiont fungi in roots) play a key role in providing ecosystem services such as soil fertility, soil formation and maintenance, nutrient cycling and improvement of plant root exploration of the soil.

(iv) **Valuing soil biodiversity:** The intrinsic value of soil biodiversity is less obvious than for above-ground biodiversity. For instance, it is much easier to convince land users and/or policy makers on the necessity to maintain above-ground biodiversity to reduce soil loss/erosion than for the same people to appreciate soil degradation (physical, chemical and biological) within the soil. It is even much more difficult to appreciate the value of soil microorganisms unless by experimental evidence e.g., significant effect on crop yield. Therefore, making a case for increasing and/or maintaining soil biodiversity will need to be substantiated with data (e.g., in terms of crop yield or economic gain/loss) unlike for the above-ground biodiversity which is more obvious. We suggest that identifying the value of soil biodiversity in terms of economic benefits should be an area of research aimed at sustaining soil biodiversity. This can be part of a wider strategy of conserving and using agro-biodiversity for sustainable soil management/agriculture (Perrings et al. 2006).

Soil biodiversity and agricultural sustainability

Sustainable food production is essential to the future of humanity and nature on earth. Soil and the biodiversity within it is the foundation of sustainable agriculture. Sustainable agricultural actions focused on soil biology are needed across cultures and farming contexts. There is an abundance of research from industrial row-cropping systems, including robust evidence that tillage negatively impacts soil organisms with larger body size including earthworms, collembola, and mites (Cavigelli et al. 2012). No-till and reduced-till systems generally support a greater proportion of fungi and bacteria compared with conventional systems (Cavigelli et al. 2012). Tsiafouli et al. (2015) found that management with annual tillage reduced soil biodiversity and shifted communities in favor of small-bodied organisms compared with grass/legume rotations and grasslands managed without any tillage. Smaller, less taxonomically diverse soil communities can lead to lower plant productivity, less N turnover, and greater P leaching (Wagg, 2014), which could lead to reduced crop yields and increased loss of nutrients from agroecosystems. In addition to tillage, presence of cover crops and retention of litter are important factors for soil communities. Cover crops, which are grown in row-crop fields outside of the primary crop growing season prevent extended periods of time when fields have exposed soil. Furthermore, the greater soil biodiversity and the functional redundancy it provides may increase the capacity of soil to continue functions like nutrient turn-over and plant productivity under global change scenarios.

The relationship between soil biodiversity and sustainability is important in explaining the contribution of soil biodiversity to the efficiency of resource use. It is clear that soil biodiversity will only be meaningful, when integrated with above-ground biodiversity to sustain ecosystem functioning. The knowledge gained will be useful only, if inspired by, and combined with farmers' knowledge, perceived problems and opportunities for application.

Soil is very complex, and man remains ignorant of the identities and activities of many of the creatures living in it. Nonetheless, it is clear that important features of soils derive from the organisms that inhabit it. Global change, in its broadest sense, is considered the main concern for biodiversity conservation, ecosystem function, and service provision (Foley et al. 2005). A substantial proportion of Earth's terrestrial surface area has been converted for human use (Millenium Ecosystem Assessment, 2005), and about 30% of soils worldwide are considered degraded with many areas highly impacted by pollution. Over 40% of Earth's land surface area is classified as arid to semiarid ecosystems, and this percentage is increasing owing to desertification (Reynolds, 2007).

Furthermore, anthropogenic activities have resulted in substantial changes in nitrogen (N) and phosphorus (P) fluxes with inputs to the biosphere increasing astronomically altering global nutrient cycles and causing imbalances among C, N, and P (Sardans and Penuelas, 2012). Associated with these observed global changes, there has been a loss of species in many terrestrial ecosystems and the introduction of novel species (Bardgett and Wardle, 2010). Such gains and losses of species can be considered a consequence but also a driver of global change with potentially significant impacts on ecosystem functioning (Wardle, et al. 2011). Belowground communities are no less affected by environmental changes than aboveground communities, and significant responses to changes in land use, climate, nutrient deposition, invasive species, and other global changes are already evident with potential implications for ecosystem functioning in both natural and managed systems (Wall et al. 2012). These changes to soils are raising global attention toward efforts to sustain soils and their living capital for the future.

It is the living communities—soil biodiversity—within soil that drive the processes central to plant growth, directly impacting human health and well-being through crop and livestock forage production (Bender and van der Heijden, 2015). Illustrations of the value of soil biota for supporting global food production are abundant and diverse. For example, soil fauna and microorganisms drive terrestrial nutrient cycling by decomposing dead plant and animal material and converting it to forms readily used by living organisms (Falkowski, 2008).

The value of soil biota is apparent in China where traditional rice-fish farming leverages rice paddies as fish habitat, where fish feed on microorganisms decomposing dead rice leaves. Fish feeding behavior loosens soil allowing more oxygen to infiltrate, further stimulating microbial decomposition of both plant material and fish excrement, which in turn liberates nutrients that are used by growing rice plants (Liu et al. 2013). Similarly, in industrial row-crop systems, rotating legumes with grain crops leverages nitrogen (N)-fixing bacteria associated with the legumes to build soil N which grain crops like maize subsequently use.

The ways in which soil biodiversity interfaces with multiple ecosystem functions makes it a natural focus for advancing a holistic global sustainability agenda. Soil biodiversity is at the heart of natural solutions for climate, biodiversity, and humanity, including protecting natural areas, restoring degraded ecosystems, employing sustainable agricultural practices, and adapting urban areas for nature and people. As we work toward a sustainable future, we cannot afford to overlook the critical and diverse asset, right beneath our feet.

The nexus: food & nutrition security and soil security

Without securing the soil, nutritious food will be much harder to produce in the foreseeable future. How we access food influences the relationship individuals and communities have with the soil. To visually demonstrate this, it is possible to map the relationship between Food and Nutrition security and Soil Security, in line with a subset of the UN Sustainable Development Goals. The realization of Sustainable Development Goals (UN General Assembly, 2015) has been a major step in placing the importance of soil care on the international agenda. Soil directly relates to seven of the goals, with the remainder indirectly related through the contribution of soil to ecosystem services (Keesstra et al. 2016). Some others are goals which are related to both soil and food, and thus highlight areas in which interactions between Soil Security and Food and Nutrition Security lie. Considering the natural factors such as climatic and environmental systems, and anthropogenic factors such as socioeconomic and political systems which affect the interaction between Food and Nutrition Security and Soil Security, the perceived strength of these interactions are identified through the dimensions (*Table 1*). In this Table, if interactions were strong, they were denoted with an asterisk. Under each of the Food and Nutrition Security dimensions the interaction with each of the Soil Security dimensions are systematically examined.

Soil Security Dimension	Food and Nutrition Dimension			
	Availability	Access	Utilisation	Stability
Capability	**	*	**-	*
Condition	***	*	**	**
Capital	* -	-	* -	**
Connectivity	**	* -	**	**
Codification	***	*	-	***

Table 1. Interactions between Dimensions of Soil Security and Food and Nutrition Security. Asterisks (*) identify connections and hyphens (–) identify disconnections (Modified from Pozza and Field, 2020).

Food and nutrition Security is food sufficiency or sustainable food production. Sustainable food production must be based on sustainable soil management which depends on the state of the soil. The four dimensions of food security: *1)* Availability - National. *2)* Accessibility - Household. *3)* Utilization - Individual. *4)* Stability - may be considered as a time dimension that affects all the levels and must be intact for optimum food and nutrition security. More recent developments emphasize the importance of sustainability, which may be considered as the long-term time (fifth) dimension to food and nutrition security. Food and nutrition security is best considered as a causal, linked pathway from production to consumption, through distribution to processing, recognized in a number of domains, rather than as four “pillars/dimensions”. Food and nutrition security/insecurity are dynamic, reciprocal and time dependent and the resultant status depends on the interaction between the stresses of food insecurity and the coping strategies to deal with them. Universal indicators for measuring food security are challenging. Different indicators may be applied to different levels of food security. Measuring food security at the household level involves five categories of indicators-dietary diversity and food frequency, spending on food, consumption behaviors, experiential indicators, and self-assessment measurements.

The biodiversity connection between food & nutrition security and soil security

From the foregoing discussion, it is very clear that Food and Nutrition Security is intrinsically linked to the soil. Trying to accommodate growing food demand through unsustainable intensification and management practices is degrading the soil and the world is running out of arable land. To avoid further degradation, land/soil needs to be used based upon what it can do. We need to care for the soil and take measures to maintain its capability and condition or work to restore what has been lost through regenerative agriculture. The linkage between Soil Security and Food and Nutrition security is supported through the explicit development of local, national, and international policy frameworks which explicitly recognise soil and its contribution and therefore the need for it to be secured.

With focus on 2030, Morgan et al. (2017) listed a set of goals where policy would contribute to securing soil, and in-turn have links to Food and Nutrition Security. The ultimate idea of these goals was for Soil Security to be integrated into policy through the SDGs and legally recognised by 50% of national governments, while providing universal indicators to assess soil sustainability, and enabling nation states that export food to take account of Soil Security and its agricultural production.

Due to the interconnectedness of soil and food, approaches to reducing global food demand can help improve both Food and Nutrition Security, as well as Soil Security. Likewise, increased implementation of Soil Security assessment, or assessment of its dimensions, will assist with the development of more consistent methods of assessing Food and Nutrition Security. Food and nutrition security depends on soil security. The six major factors that influence soil security have been identified as: water, climatic factors (rainfall and temperature), man, energy, and biodiversity (*Figure 3*). Of these six factors, biodiversity stands out. It is both within the soil and above it and it is also partly a product of the soil and also supplies part of the energy required in the soil through the activities of micro and macro-organisms.

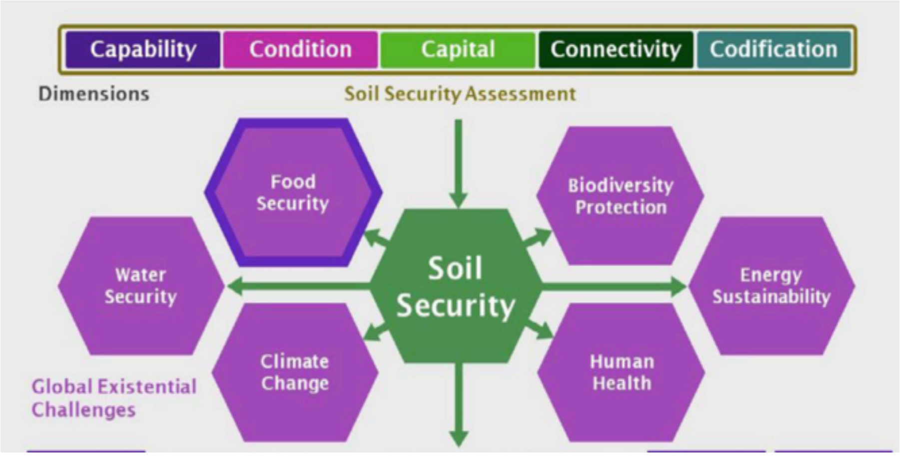

Figure 3. Major Factors influencing Soil Security (Modified from Pozza and Field, 2020).
Note: Food and Nutrition Security is both a Factor and a Product of Soil Security.

It is obvious that soil biodiversity is inseparable from the soil and any attempt to do that leads to soil insecurity and this automatically results in food and nutrition insecurity: this is the unique connection. Natural factors such as climatic and environmental systems, and anthropogenic factors such as socioeconomic and political systems affect the interaction between Food Security and Soil Security; the perceived strength of these interactions are as shown in *Table 1*.

Soil organic C factor in the biodiversity connection

Soil should be considered an ecological system and a finite resource. Conservation of soil biodiversity and/with soil organic carbon (SOC) through sustainable farming practices is essential to improve agricultural land health and productivity. Improved soil biodiversity is closely correlated with an increase in carbon stored in soil, for it sequesters organic carbon and is a natural carbon sink. It is, therefore, required for reducing GHG emissions. The global carbon (C) cycle includes a significant C stock in terrestrial soils, including agricultural soils. Global land and soil degradation is strongly linked to the depletion of this stock.

Furthermore, it has been established that if organic carbon in all the world's agricultural soils were to increase by 0.4 per cent annually the global production of three of the most important food crops — maize, wheat, and rice — could increase by up to 23.4 per cent, 22.9 per cent and 41.9 per cent respectively between 2020 and 2050 (Gazdar, 2020). Additionally, hydrological cycles also benefit from increased soil carbon. The capacity of soils to store water could increase by up to 37 billion cubic meters. The increase in soil moisture from 4‰ (parts per thousand) has the potential to reduce reliance on irrigation, with estimated global savings of $44 billion per year (Gazdar, 2020).

A relative change in SOC concentration may be assumed to correlate with changes in soil biodiversity and the generation of supporting ecosystem services (Brady et al. 2015). The 0.4 per cent comes from the initiative launched by the French government in 2015 called 'Four per thousand' (4‰) to improve agricultural ecosystems for increased food security and combat climate change (Larbodière et al. 2020). This is achieved by increasing the soil organic carbon SOC by 0.4 per cent in the top 30-40 cm of soil (where most arable and pastoral plants grow) annually through economically viable and environmentally sound agronomic practices. This is illustrated in Figure 4. The benefits of 0.4 per cent SOC could result in increased resilience for vulnerable farming communities, and reduced exposure to projected risks associated with climate change such as floods, droughts, and storms. This is due to nothing else but enhanced soil biodiversity.

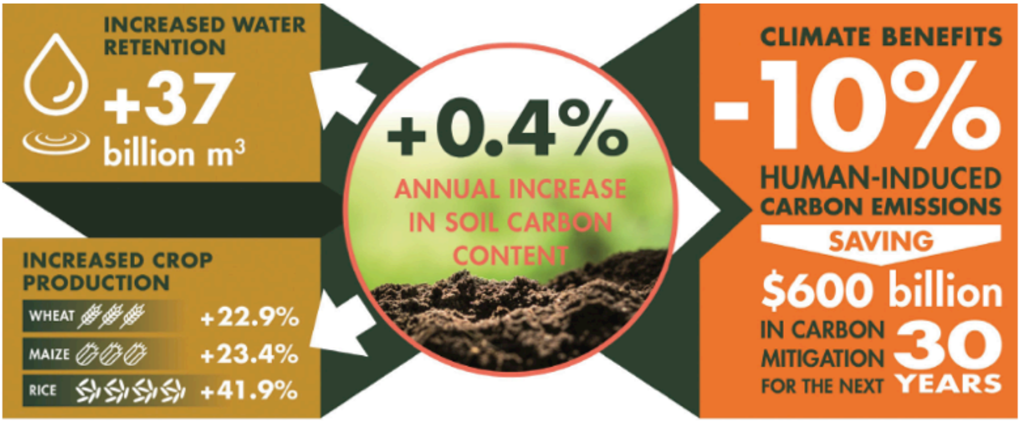

Figure 4. The pathway towards the 4 per 1000 benefits from increased organic carbon in soils (Source: Larbodière, et al. 2020). *Potential benefits from increasing organic carbon in soil.*

Conclusions

Soil biodiversity influences all the soil functions that secure the soil for Food and Nutrition Security. This is clearly revealed in the interactions between the dimensions of Soil Security and those of Food and Nutrition Security. The physical (crop yield) and economic (financial) benefits and the climate change mitigation effect of SOC confirm the need for further research on the value of soil biodiversity in the CONNECTION function between Soil Security and Food and Nutrition Security.

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Chapter 12

Can soil improving cropping systems reduce the loss of soil biodiversity within agricultural soils?

Felicity Crotty, S. Emilia Hannula, Moritz Hallama and Ellen Kandeler

Summary

Soil biodiversity, including abundance and function of species living in the soils are important aspects of soil health, and acknowledges that soil is a living ecosystem. The decline of soil biodiversity can lead to a simplification of the soil food web and the inter-relationships as a result of poor soil management. The impact of agricultural management has been shown to reduce diversity, with damaging consequences for nutrient cycling, soil structure and potentially crop yields. However, research is often siloed and the effect of soil biodiversity on crop yields is overlooked by soil ecologists focusing on other ecosystem services; and the role of soil biodiversity on maintaining soil quality and improving crop yields is missed by agronomy researchers. Introducing specific soil improving cropping systems (SICS) have previously been considered as a method to combat soil threats (such as soil erosion or compaction) but have been overlooked as a method of reducing soil biodiversity losses within agriculture. As soil biodiversity loss is interlinked with these other soil threats, SICS will also potentially reduce the impact of them. Here, we review the range of SICS that can be utilised to reduce the threat of soil biodiversity loss, evaluating the effect of SICS on biodiversity across a wide range of organismal groups and consider the impact this will have on the sustainability of agricultural management. There are examples of measures and practices that combine high crop yields with the promotion of soil biodiversity. Selecting specific cropping systems designed to maintain or increase soil biodiversity (e.g., cover crops), promoting the stabilisation of the soil environment (minimum tillage), reducing chemical amendments (targeted spraying and biocontrol), or increasing biological amendments (adding organic matter) are all potential SICS that can be utilised.

Introduction

Soil quality and the protection of its functions are not specifically mentioned as a sustainable development goal (SDG) by the United Nations, but soil-related ecosystem services are implicitly linked to several SDGs, especially *No poverty* (SDG 1), *Zero hunger* (SDG 2), *Good health and well-being* (SDG 3), *Clean water and sanitation* (SDG 6), *Climate action* (SDG 13), and *Life on land* (SDG 15) (Bach et al. 2020). The European Commission defined the soil as having seven basic functions: *1)* Biomass production (including agriculture and forestry); *2)* Storing, filtering, and transforming nutrients, substances and water; *3)* Biodiversity pool (habitats, species and genes); *4)* Physical and cultural environment for humans and human activities; *5)* Source of raw material; *6)* Carbon pool; *7)* Archive of geological and archaeological heritage; (European Commission, 2006). Agricultural production is only one of these seven soil functions, the biodiversity as a pool is one of the others; it is up to agricultural management and the use of soil improving cropping systems (SICS) to prevent the loss of one function due to another. Research has shown that agricultural intensification across Europe has reduced soil biodiversity (Tsiafouli et al. 2015), however within extensive agricultural systems soil biodiversity can reach similar levels as permanent grasslands (van Eekeren et al. 2008).

Intensive agricultural systems have also been shown to impact the ecosystem services delivered by soil biodiversity, for example De Vries et al. (2013) showed that carbon and nitrogen cycling were directly linked to the soil food web across land use systems. It is commonly acknowledged that conserving soil biodiversity is key to improving and sustaining soil quality (e.g. Firbank et al. 2008, Handa et al. 2014, Crotty et al. 2015). Soil quality refers to "the continued capacity of soil to function as a vital living system, within ecosystems and land-use boundaries, to sustain biological productivity, maintain the quality of air and water environments, and promote plant, animal, and human health" (Doran and Zeiss, 2000). In current agriculture, usually yield maximization is prioritised over other ecosystem services provided by the soil such as nutrient cycling and carbon storage (Bender et al. 2016), key functions largely performed by soil organisms. Bünemann et al. (2018) review on soil quality showed how research to date has prioritised soil physical and chemical features as measures of soil quality, rather than biological analyses. This is likely due to the difficulties in studying these organisms, such as the difficult taxonomy combined with a paucity of specialists (Porco et al. 2014), opacity of the environment (Crotty et al. 2012), being mostly invisible or hidden within the soil matrix (Geisen et al. 2019) and general lack of funding available (Kim and Byrne, 2006).

The response of diversity, abundance, and function of soil organisms to soil management constitutes an important aspect of soil quality (Mbuthia et al. 2015) and life within the soil (i.e., soil biodiversity) in essence represents the soil's health (Paoletti, 1999). Soils are among the most abundant and diverse habitats on earth (Bender et al. 2016). This diversity in species and function is explained extensively in both the European Atlas of Soil Biodiversity (Jeffery et al. 2010) and the Global Atlas of Soil Biodiversity (Orgiazzi et al. 2016); as well as the recent report on the state of soil biodiversity (FAO et al. 2020). There is a greater biomass of soil fauna residing below-ground than normal stocking densities of livestock grazing above-ground (Crotty, 2021).

Together these organisms participate in functions important to the maintenance of soil quality. Throughout this review, soil biodiversity is discussed in terms of abundance as well as diversity; this is because biomass (abundance) is an indicator for biodiversity of the whole soil ecosystem. Both aspects – abundance and diversity are necessary for soil health and a fully functioning soil food web to perform ecosystem functions.

Review aims and hypothesis

This review will focus on soil improving cropping systems that reduce soil biodiversity loss, with a focus on temperate Europe. Here, SICS are defined as any agricultural management strategy that modifies the soil environment to improve the "health" or quality of the soil. A healthy soil is a sustainable soil that continues to produce profitable crop yields within agriculture, whilst also reducing biodiversity loss. If ecosystems are severely modified through intensive agriculture, then in theory SICS implementation is going to have a greater relative effect on reducing soil biodiversity loss than in more extensive farming systems, especially when leading to a more diverse landscape. However, even in a more extensive system, we hypothesise that SICS can still help to reduce soil biodiversity loss (*Figure 1*).

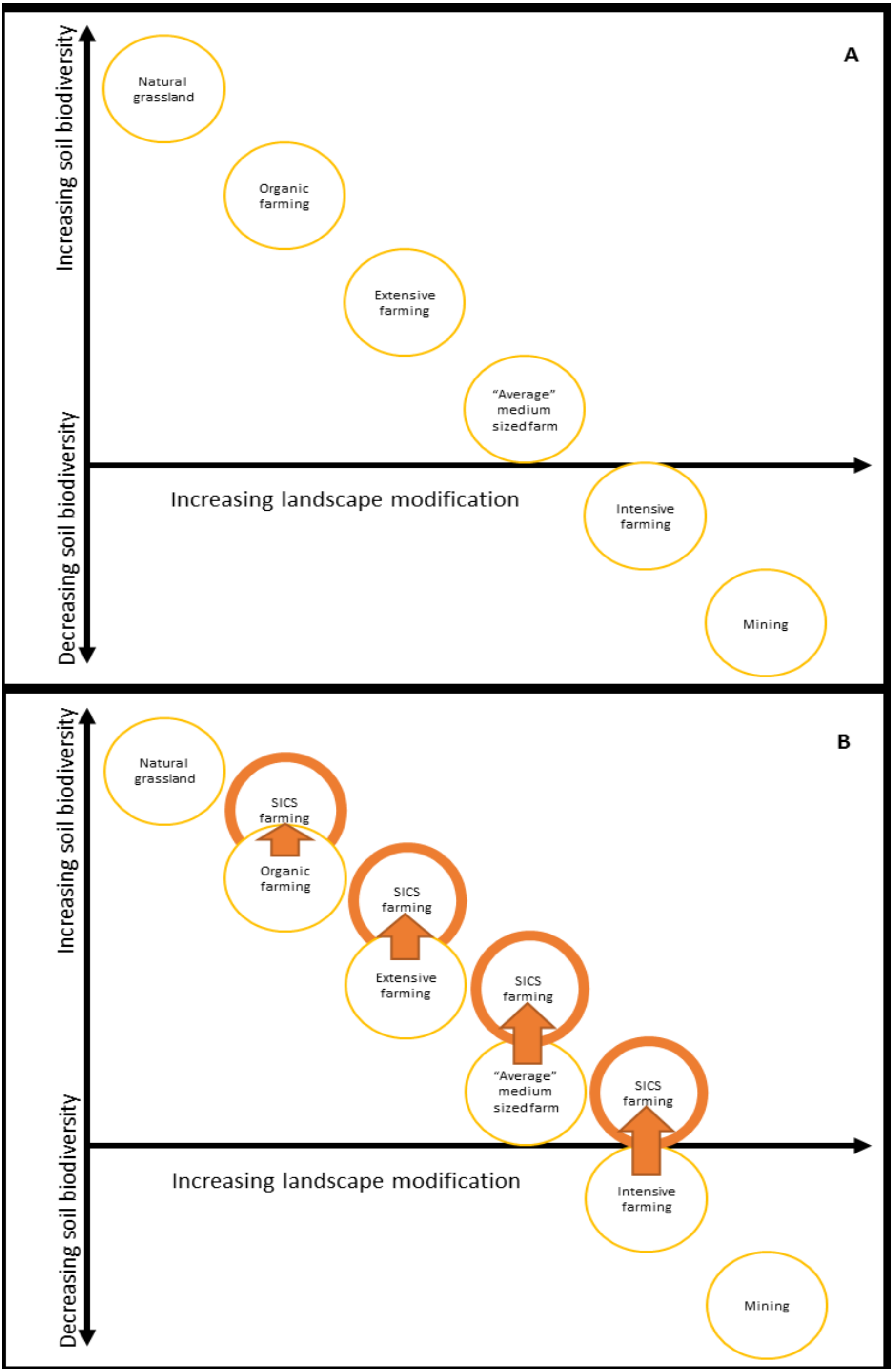

Figure 1. Visualisation of the effect of land management on soil biodiversity (**A**) and the effect soil improving cropping systems would have if they were implemented (**B**).

Key types	Cropping systems		
Crop	- *Cover crops* - Cash crops - Catch crops - Mixed cropping - Strip cropping - Companion cropping - *Intercropping* - Monoculture - Double cropping	- Crop rotation - Deep rooting crops - Vegetable crops - *Brassicacea* inclusion - Nitrogen fixing plants - Landraces (natives) - Permanent cropping (fruit) - Biodiverse mixes - Wild flower mixes	- Pollinator mixes - *Headland alternatives* - *Grass or grass/clover leys* - Fallow - Set aside - Buffer strip - *Agroforestry* - *Energy crops/forests*
Physical soil environment changes and water	- *Non-inversion tillage* - *Reduced tillage* - *Minimum tillage* - *Spike aeration*	- Soil conservation - Terracing - Soil stripping - Subsoiling	- Contouring - *Drainage management* - Irrigation management - Drip irrigation
Chemicals	- *Plant protection products* - Pesticide use - Soil amendments	- *BCSR amendment* - *Liming* - *Gypsum*	- Biofumigation - Solarisation - *Synthetic fertiliser*
Biological amendments	- *Organic fertiliser* - *Manure / slurry addition* - *Biodigestate* - *Mulching / living mulch* - Composting	- Residues - Compost tea - *Organic farming* - *Biochar* - Woodchip	- Mycorrhizal amendments - Bioaugmentation - Probiotics - Paludiculture - *Bio control*

Table 1. Overview of potential soil improving cropping systems. Cropping systems in italics are discussed more fully within this review.

There are many different types of SICS (*Table 1*) and these can be broadly grouped into five main categories – crop choice, physical/environmental change within the soil, chemicals, biological amendments and technology, but how they fit together within agriculture needs further study (*Figure 2*). Each SICS will affect biodiversity differently and different components of biodiversity will respond to different SICS – the aim of this review is to develop our understanding of the impact of these SICS on preserving soil biodiversity and reducing its loss. Most mechanisms that increase biodiversity with SICS are based on either change in (1) the distribution, quantity and quality of organic matter (OM) or (2) in improvement of habitat conditions for soil organisms (soil structure, aeration, and/or increased water holding capacity). Consequently, the focus of our review is to give an overview of the effect of different agricultural practices on functional groups of soil biota and soil biodiversity in general to depict the already available possibilities to manage agroecosystems in a more sustainable way by maximising biological functions providing ecosystem services.

Measuring biodiversity

Measuring biodiversity is incredibly challenging (Tibbett, 2015; Geisen et al. 2019), even more so in agricultural context (Lemanceau et al. 2015). Whilst blindly enhancing soil biodiversity infers random inclusion of many species (Bender et al. 2016), maintaining or increasing everything may lead to the inclusion of a greater diversity of undesirable organisms, e.g., pests (Simon et al. 2010) and weeds (Sanyal and Shrestha, 2008). However, a biodiverse agroecosystem might also decrease the opportunity for single, harmful, organisms to dominate (Wall et al. 2012). For more information on soil borne pathogens see Box 1.

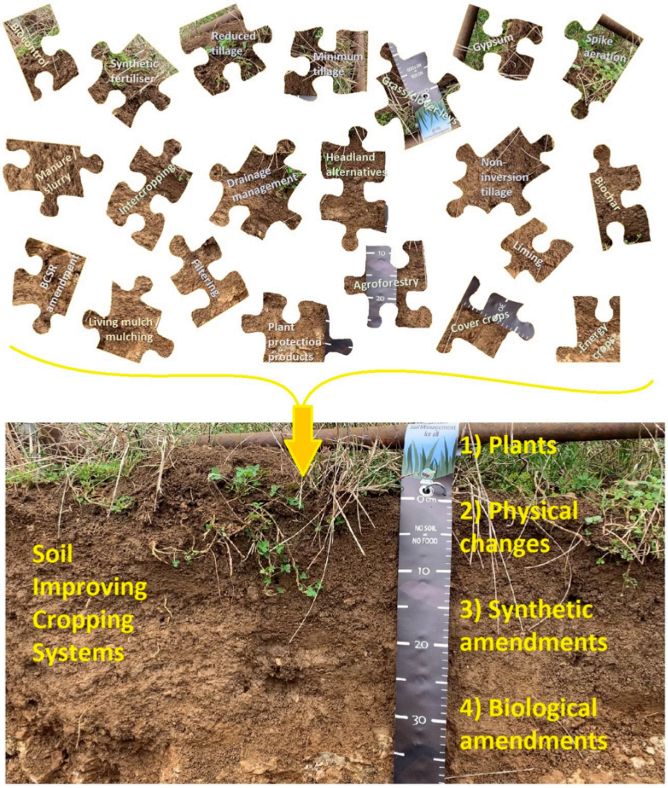

Figure 2. Soil improving cropping systems fit together like a jigsaw to improve the soil habitat for the organisms living within the soil. How they do this and by how much varies depending on the (SICS).

Box 1. Soil-borne Plant Pathogens

Soil-borne plant pathogens are part of the biodiversity within the soil, although are often not considered when soil biodiversity is discussed. These pathogens can survive a prolonged time within the soil, waiting for a host plant to grow. These organisms are various taxa including fungi, nematodes, chromists, protists and parasitic plants, and are found everywhere, both in agricultural and natural systems. All SICS that affect soil structure positively will reduce the threat of soil biodiversity loss, whilst also increasing the disease suppressiveness of the soil, reducing opportunities for opportunistic plant pathogens such as *Pythium* species to flourish. Once soil-borne plant pathogens appear at damaging levels within an agricultural system, it is necessary to control them as quickly and completely as possible; without destroying the rest of the soil biodiversity – although usually this is the unintentional consequence of soil-borne pathogen management. Commercial cultivars have been bred to be high yielding, whilst unintentionally becoming more susceptible to soil-borne pathogens, whilst years in nutrient rich conditions has also reduced crops dependency on their symbionts (Duhamel and Vandenkoornhuyse 2013). Currently, there are many projects investigating historic varieties and native relatives, to increase the naturally occurring resistance genes within a crop. Microcosm experimentation with different cultivars (including landraces) and earthworms, showed there was a large variation in responsiveness between cultivars – with a local landrace performing best with earthworms (Noguera et al. 2011).

There are a number of studies that have linked biodiversity increases with soil functions (e.g., Morriën et al. 2017, Wagg et al. 2014, Schäfer et al. 2019), however, this is rarely investigated in agricultural systems. The effect of cropping systems on the increase in abundance of a single group e.g., earthworms (Pelosi et al. 2015); although an important consideration, might be different to an increase in overall biodiversity. Furthermore, it is possible that an increase in biodiversity of one component in the food-web can lead to a decrease in diversity of another group through competition for resources or to have an add-on effect through feeding relationships (Morriën et al. 2017, Geisen et al. 2019) which makes the interpretation using only a single group of soil organisms more challenging. To monitor biodiversity, in principle all taxonomic groups need to be assessed (Griffiths et al. 2016), however, this is often impossible due to lack of expertise or prohibitively expensive. Therefore, within this review we limit ourselves to the relatively well-known groups of microbes (fungi and bacteria), meso- and macrofauna.

Overall, soil microbial biomass carbon accounts for 19 Gt globally (Crowther et al. 2019); this microbial biomass (C and N), is relatively simple to measure and can act as an indicator of management driven changes in diversity, abundance, and function (Kandeler, 2015). Arbuscular mycorrhizal fungi (AMF) form a symbiotic relationship with plants helping them to acquire nutrients and contribute to soil structure formation (Rillig and Mummey, 2006) making them one of the most studied groups. The composition of the AMF community is generally determined by plant species identity, plant diversity and soil nutrient status, especially P availability (Goldmann et al. 2020). Furthermore, soil disruption by mechanical management within agriculture (e.g., tillage) can be detrimental to both fungal hyphae and earthworm populations. Soil animals (i.e., Nematodes, Collembola, mites and earthworms) are important indicators of soil function and being further up the food chain than soil microbes, they integrate the physical, chemical, and biological properties related to their food resources; whilst due to their longer generation time (days to years) they are more stable temporally and not fluctuating as much due to nutrient flushes (Neher, 2001). Earthworms represent the largest component of animal biomass within the soil and are commonly considered to be ecosystem engineers (Blouin et al. 2013). Ecosystem engineers are organisms that affect the whole environment and either directly or indirectly have an impact on the other species inhabiting the same space (Jones et al. 1994).

It is not just the SICS that is important to understand, but the surrounding environment. Consequently, recommendations to achieve high levels of biodiversity in agroecosystems must consider specific pedo-climatic conditions or soil type. Bossio et al. (1998) gave a useful synopsis of the order of importance in evaluation of SICS: soil type > season > specific farming operation (SICS) > management system > spatial variation.

For example, earthworms are found to naturally occur at greater abundance in clay textured soils compared to sandy soil; however, compaction and waterlogging, which are also more likely to occur in clay textured soils, reduce the habitability of the environment for earthworms. Thereby introducing a SICS that reduces compaction and waterlogging risk could consequently improve earthworm abundance. Increasing earthworm abundance could also reduce the risk of compaction and waterlogging (as earthworms are ecosystem engineers and have a large impact on soil structure), leading to improvements in crop yields through soil biodiversity enhancement (*Figure 3*). Generally, all the different components of biodiversity in the soil are directly or indirectly affected by the cropping system (*Table 2*). However, each group of organisms may be affected differently, and it is thus important to understand how the mechanisms of agronomic practices affect these organisms and the functions they fulfil.

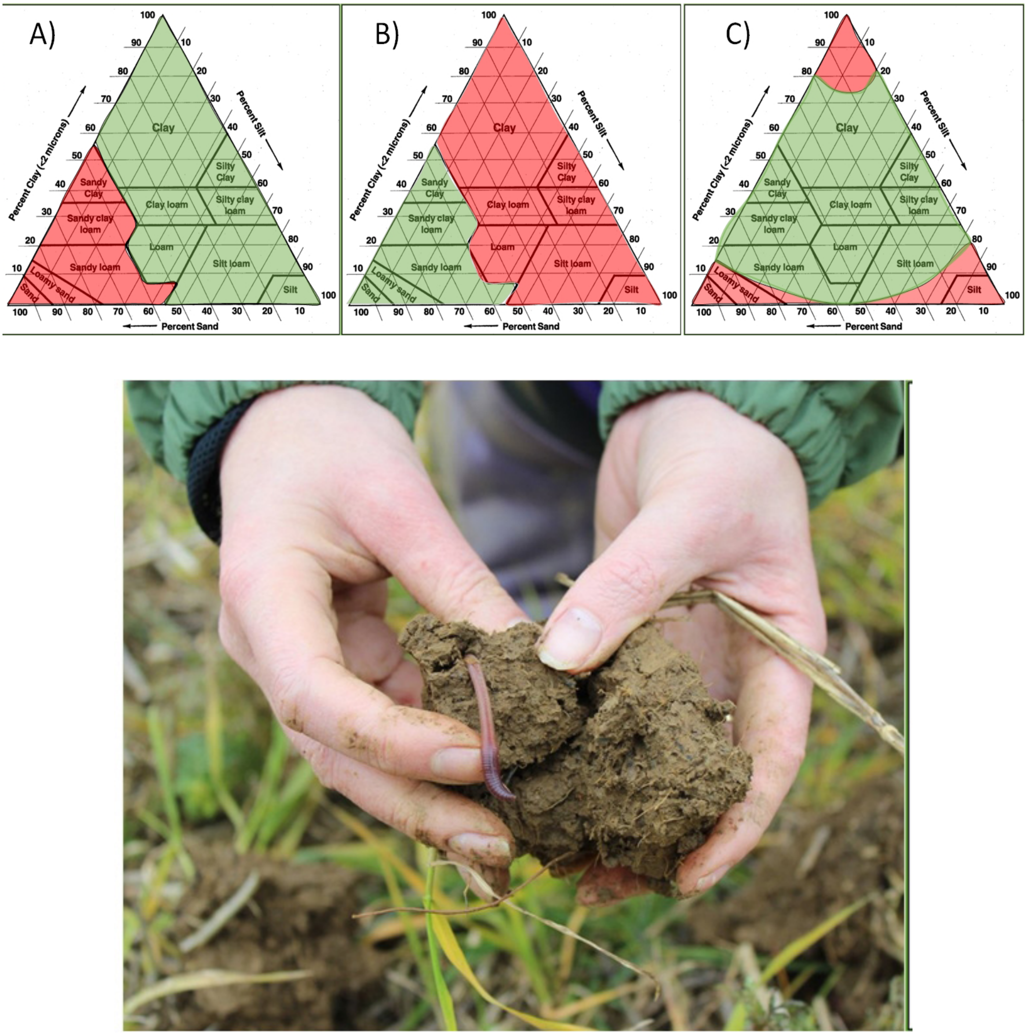

Figure 3. Conceptual diagram indicating A) abundance of earthworms in relation to soil texture; B) relative soil quality in relation to likelihood of waterlogging and compaction; and C) effect increased abundance of earthworms have on risk of waterlogging and compaction. [Green = high abundance of earthworms or low risk of waterlogging/compaction; red = low abundance of earthworms or high risk of waterlogging/compaction]. Soil sampling is shown below.

Organism	Effect on food and fibre production	How cropping system can affect organisms?
Bacteria (and Archaea)	- Increase nutrient availability - Promote plant growth - Reduce pathogens - Process/modify agrochemicals and xenobiotics - Change soil composition - Enhance soil structure - **Soil-borne pathogens**	- Increase food source (OM / amendments) - Flush of nutrients - **Change environment of soil (water/temperature)** - **Kill organism directly (pesticides)**

Organism	Effect on food and fibre production	How cropping system can affect organisms?
Fungi	- Cycling of essential nutrients - Promote plant growth - Increase N and P availability (through symbiosis) - Reduce pathogens - Process/modify agrochemicals and xenobiotics - Change soil composition - Enhance C allocation and build up OM - Decreased seedling mortality - Biocontrol against pests - Enhance soil structure (through hyphal growth and glomalins secreted by AMF) - Decomposition of plant residues (and subsequent release of nutrients). **- Soil-borne pathogens**	- Increase food source (OM / amendments / maintain plant residues / cover crops) - Flush of nutrients to provide burst of growth **- Change structure of soil (destroy hyphal network)** **- Kill organism directly (pesticides)** **- Remove host plant** **- Removal of residues of plant (food source)**
Protists	- Enhance microbial growth - Increase nutrient availability **- Soil-borne pathogens**	- Flush of nutrients - Increase food source (OM / amendments) **- Change environment of soil (water/temperature)** **- Kill organism directly (pesticides)**
Rotifers	- Contribute to nutrient cycling - Flocculation of bacteria	- Flush of nutrients **- Change environment of soil (water/temperature)** **- Anhydrobiosis (enables them to survive extended periods of desiccation)** **- Kill organism directly (pesticides)**
Tardigrades	- Enhance microbial growth	- Flush of nutrients **- Change environment of soil (water/temperature)** **- Anhydrobiosis (enables them to survive extended periods of desiccation)** **- Kill organism directly (pesticides)**
Nematodes	- Increase nutrient availability - Disperse bacteria and fungi - Reduce pathogens **- Soil-borne pathogens**	- Flush of nutrients - Increase food source (OM / amendments) **- Change environment of soil (water/temperature)** **- Change structure of soil** **- Kill organism directly (pesticides)**
Collembola	- Increase nutrient availability - Breakdown plant material, animal carcases - Faecal pellets contribute to soil microstructure and fertilisation. - Disperse microorganisms and nematodes. - Micro-ecosystem engineer (Brussaard et al. 1997) - Consumer of pathogens **- Host for parasites**	- Flush of nutrients - Increase food source (OM / amendments) **- Change environment of soil (water/temperature)** **- Change structure of soil** **- Kill organism directly (pesticides)**

Organism	Effect on food and fibre production	How cropping system can affect organisms?
Mites	- Increase nutrient availability - Breakdown plant material, animal carcases - Faecal pellets contribute to soil microstructure and fertilisation. - Disperse microorganisms and nematodes. - “Micro-ecosystem engineer” (Brussaard et al. 1997) - Consumer of pathogens - Host for parasites/parasitoids	- Flush of nutrients - Increase food source (OM / amendments) **- Change environment of soil (water/temperature)** **- Change structure of soil** **- Kill organism directly (pesticides)**
Soil dwelling immature invertebrates e.g. beetle larvae, fly larvae	- Fragmentation and decomposition of organic material - Change pH of soil passing through gut	- Flush of nutrients - Increase food source (OM / amendments) **- Change environment of soil (water/temperature)** **- Change structure of soil** **- Remove food source (plant species)** **- Kill organism directly (pesticides**
Other mesofauna (body width less than 2mm) e.g. Protura, Diplura, pseudoscorpions, beetles, spiders, Thysanoptera,	- Increase organic matter through burial of dung or carcasses - Predators of pests (pseudoscorpions, spiders etc)	- Flush of nutrients - Increase food source (OM / amendments) **- Change environment of soil (water/temperature)** **- Change structure of soil** **- Remove food source (plant species)** **- Kill organism directly (pesticides**
Myriapoda (centipedes and millipedes mainly and Pauropoda, Symphyla)	- Excreta contribute to coprogenic humus	- Flush of nutrients - Increase food source (OM / amendments) **- Change environment of soil (water/temperature)** **- Change structure of soil** **- Remove food source (plant species)** **- Kill organism directly (pesticides**
Enchytraeids	- Fragmentation and breakdown of plant litter - Enhance microbial growth, - Change soil structure (bioturbation) - Disperse of microorganisms	- Flush of nutrients - Increase food source (OM / amendments) **- Change environment of soil (water/temperature)** **- Change structure of soil** **- Remove food source (plant species)** **- Kill organism directly (pesticides**
Root herbivorous insects / pests	**- Modifies plant performance** **- Yield losses** **- Changes plant physiology** **- Transmits diseases**	- Remove food source (plant species) - Introduce host food source - Change structure of soil - Kill organism directly (pesticides)
Earthworms	- Enhance microbial growth, - Change soil structure (bioturbation) - Disperse microorganisms - Aids sporulation / germination of fungal spores. - Improves water infiltration - Ecosystem engineer	- Flush of nutrients - Increase food source (OM / amendments) **- Change environment of soil (water/temperature)** **- Change structure of soil** **- Remove food source (plant species)** **- Kill organism directly (pesticides**

Organism	Effect on food and fibre production	How cropping system can affect organisms?
Ants	- Enhance microbial growth - Disperse plant propagules - Change soil structure (bioturbation) - Increase porosity and drainage - Reduce bulk density - Ecosystem engineer	- Flush of nutrients **- Change environment of soil (water/temperature)** **- Change structure of soil** **- Remove food source (plant species)** **- Kill organism directly (pesticides)**
Termites	- Enhance macroporosity and infiltration - Change soil structure (bioturbation) - Enhance microbial growth, - Ecosystem engineer	- Flush of nutrients **- Change environment of soil (water/temperature)** **- Change structure of soil** **- Remove food source (plant species)** **- Kill organism directly (pesticides)**

Table 2. Effect of organisms on food and fibre production overview (bold text indicates negative effects).

The inclusion of a SICS offers directly observable benefits for the soil ecosystem, but also indirect effects by modifying biodiversity-related functions. This can be illustrated taking as an example the inclusion of cover crops into the crop rotation (*Figure 4*). There are several direct effects of this management technique, both aboveground, for example, an increased C input into the system or erosion reduction, as well as belowground in the root zone, including the mineralisation of sparingly available nutrient pools by root-derived enzymes, or, for some plant species, rhizobial N fixation (*Figure 4*).

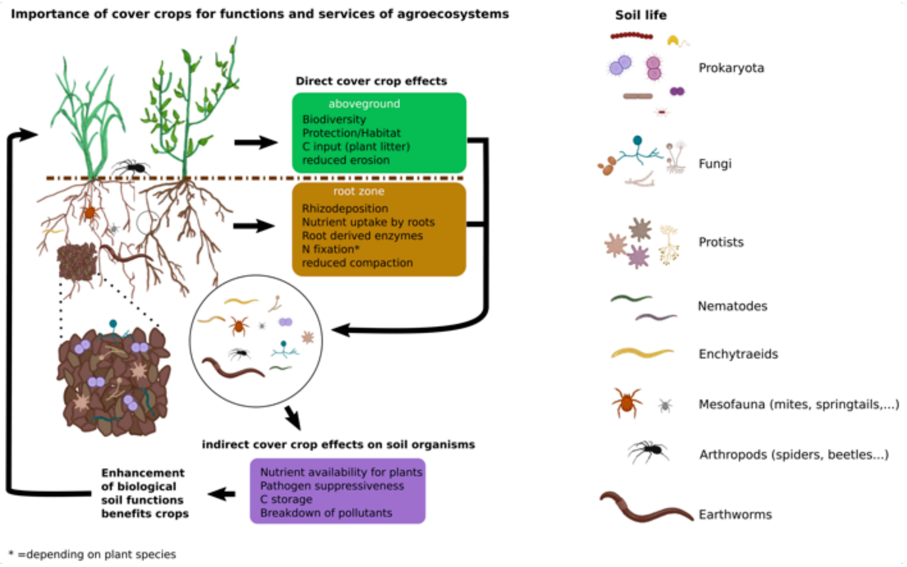

Figure 4. Importance of cover crops for functions and services of agroecosystems. Cover crops produce direct above and belowground effects which benefit the main crop, while also improving the habitat conditions for soil organisms. As soil biota are the drivers of several important ecosystem functions, the increase in biological diversity, abundance and activity enhances biological soil functions and agroecosystem services. On the right side is an overview of the different groups of organisms constituting soil life.

However, the presence of a living plant cover also improves the diversity, abundance, and activity of soil organisms, which control many processes, such as the availability of nutrients, suppression of pathogens, and C storage (the transformation of litter-derived C into soil organic matter). These enhanced biological soil functions benefit the crops grown on this field, leading to an improvement overall in the agroecosystem's performance.

There is often a discrepancy in the literature on the effects of soil biodiversity on plant productivity as soil ecologists tend to measure the effects of soil biodiversity on other ecosystem services than crop yield. Conversely, agronomists and practitioners often focus on yield measures without considering the role soil biodiversity has in maintaining soil quality and future crop yields. Therefore, there are large gaps in the literature, where these different focuses have rarely been considered, particularly in field-scale experiments. However even where yield and soil biodiversity are measured together, they are often not related directly (*Table 3*).

Location	System	Crop Performance	Soil biodiversity gain/loss	Reference
Argentina	Conventional agricultural management	Peanut-maize-sunflower rotation -> Maize grown during experimental period until end of year, before being grazed and residue ploughed under. Oat and sweet clover sown because of yield decline of maize and sunflower. ⇨ ***negative***	Collembola and pauropod reductions ⇨ ***negative***	(Bedano et al. 2006a)
		Peanut-maize-sunflower rotation -> suspected low yields of maize led to crop used as pasture for cattle during experimental period. ⇨ ***negative***	Mesostigmata and Oribatida reductions ⇨ ***negative***	(Bedano et al. 2006b)
	Good Agricultural Practice (GAPs) under no-till	GAPs (mixed crop rotation; cover crops; integrated pest, weed and disease management; nutrient restoration) in productive no till fields ⇨ ***positive***	Increases in litter and soil invertebrate abundance and modified faunal composition ⇨ ***positive***	(Bedano et al. 2016)
	Livestock	Sorghum with and without cover crops under no-till. Cover crops rye, vetch, and rye + vetch, rye + fertiliser. All cover crops with rye had similar aerial biomass whilst vetch produced significantly less ⇨ ***positive***	Cover crops increased litter and increased microbial biomass carbon and nitrogen. Soil litter disappearance is a good indicator of mineral N availability. ⇨ ***positive***	(Frasier et al. 2016)
Canada	Pasture	Grazing intensity increasing ⇨ ***neutral***	Mites less affected than Collembola; Prostigmata more abundant with grazing, Mesostigmata less abundant/ diverse with grazing. Overall diversity and abundance associated with reduced grazing intensity. ⇨ ***negative***	(Behan-Pelletier, 2003 and the references therein)
China	Maize	P-fertiliser application to maize did not affect yields due to large build up in soil prior to monitoring. ⇨ ***neutral***	Root length colonisation and arbuscular colonisation of maize roots was significantly affected by P-fertiliser application – higher rate decreased colonisation. ⇨ ***negative***	(Lui et al. 2016)

Location	System	Crop Performance	Soil biodiversity gain/loss	Reference
France	Conventional	Conventional vs no-till – Positive correlation between earthworm abundance and crop production, although contrasting effects on yields. ⇨ ***positive***	Direct drilling increases earthworm abundance and species diversity, but the beneficial effect of reduced tillage depends upon the species present and tillage intensity. ⇨ ***positive***	(Bertrand et al. 2015 and references therein)
Germany	Perennial energy crops	No difference in post-harvest C:N ratios (yields not discussed) ⇨ ***neutral***	Earthworm diversity, number and biomass significantly increased in newly introduced energy crops in comparison to maize. ⇨ ***positive***	(Emmerling, 2014)
Spain	Horticulture	Lettuce, Brussel sprouts, leaf beet and cabbage rotation; metaldehyde compared with biocontrol applications. For slug eradication, metaldehyde was only effective reduction of *Deroceras reticulatum*, whilst biocontrol only effective for *Arion ater*. ⇨ ***neutral***	Total numbers of all earthworm species, Collembola and Acari, were unaffected by treatments. ⇨ ***neutral***	(Iglesias et al. 2003)
Switzerland	Arable - Wheat	Two organic systems, one conventional with farmyard manure and one conventional with mineral fertiliser. Grain and straw yield higher in two conventional systems. ⇨ ***neutral***	Bacterivorous nematodes and earthworms most abundant in FYM receiving systems. Mineral fertiliser detrimentally affected enchytraeids and dipteran larvae. Spider abundance increased by organic management. ⇨ ***positive***	(Birkhofer et al. 2008)
USA	Arable - cover crops and tomato/corn rotation	Productivity highest after cover crops with high nitrogen content; significantly correlated with the structure of the soil fauna. ⇨ ***positive***	Cover cropped tomato/corn rotations had increased abundance of enrichment opportunist bacterial feeding nematodes ⇨ ***positive***	(DuPont et al. 2009)
UK (Scotland)	Pasture	Nitrogen fertiliser and lime addition to an upland grassland, increased above ground plant biomass and root returns to soil. ⇨ ***positive***	Nitrogen and liming reduced bacterial biomass (no change in fungal biomass). ⇨ ***negative***	(Murray et al. 2006)
UK (Wales)	Pasture	Nitrogen fertiliser application – inorganic or organic. Swards under organic management were botanically more diverse, although perennial ryegrass and white clover were predominant in all fields. ⇨ ***neutral***	Fungal PLFAs, Tardigrades, Acari and Nematodes were greater under organic management. Earthworm populations were smaller under organic management.. ⇨ ***positive***	(Yeates et al. 1997)

Table 3. Effect of agricultural management linked to crop performance and soil biodiversity at the field-scale.

Soil improving cropping systems

Several studies have shown that when the intensity of agricultural practices increases, the abundance and biodiversity of soil biota decreases (e.g. Tscharntke et al. 2005, Ponge et al. 2013, Bedano et al. 2016). Here, we highlight how agricultural practices can be harnessed to increase soil biodiversity whilst also promoting crop yield and sustainable intensification (*Figure 2*).

Crops

The biodiversity of soil organisms depends not only on the quantity and quality of food sources, but also on their spatial distribution (Müller et al. 2017, Ali et al. 2018, Pausch et al. 2018). Soil organisms are generally characterised by different life strategies: ones utilizing high pulses of (labile) OM resources (*r* strategist) and others using more recalcitrant organic compounds (*K* strategists). Consequently, crop selection, rotation or intercropping might induce a variation in root and exudate distribution within the soil profile as well as changes in the quantity and quality of OM in agroecosystems.

Crop selection and rotation

It is rare for soil to remain bare within nature, even during a fallow period within a crop rotation, plants will grow; whether these are rare arable plants or pernicious weeds is dependent on the seed bank. Soil biodiversity relies on the input of plants, as a food source and also to stabilise the soil habitat. Crop species differ in their ability to form AMF associations, together with N-fixing rhizobacteria the most studied soil microorganisms in an agricultural context, and in the benefits, they gain from this symbiosis (Kiers et al. 2011). Most agricultural crop species, with some notable exceptions (e.g., Brassicaceae, Polygonaceae and the Chenopodiaceae), are able to form AM fungal associations (Sylvia and Chellemi, 2001). However, the amount plants rely on their symbiotic mycorrhizal fungal partner is dependent on many factors, both biotic and abiotic. Since AM fungi are biotrophic, viability of AM hyphae gradually decreases in the absence of host plants such as during a fallow period. Harinikumar and Bagyaraj, (1988) in India reported 40% reduction of AM inoculum in field soil after leaving the land fallow for one season. Furthermore, long-fallow periods (>1 year) in northern Australia were associated with a decline in mycorrhizal colonization and AM sporulation in various crops (Thompson, 1987). Studies have shown that the longer the time the soil is left bare for, the larger the decrease in SOM (the only carbon source available during a fallow period); and also, the larger the detrimental effect on beneficial organisms like earthworms (Bertrand et al. 2015), however soil bacterial diversity has not been found to be affected (Hirsch et al. 2009). Leaving the soil bare for long periods of time also risks damage from other soil threats, particularly erosion.

Cover crops

Cover crops, catch crops and green manure refer to farming practices where plants are grown without removing the biomass, with the primary goal to maintain soil productivity and fertility, reducing erosion, and ensuring green cover throughout the year (Lehman et al. 2015). In general, the use of cover crops increases soil OM content and reduces soil compaction and can therefore be regarded as positive for soil biodiversity (Vukicevich et al. 2016), especially when multi-species cover crop mixtures are planted (*Figure 4*). Earthworm abundance and diversity is affected by cover crop species, for example Valckx et al. (2011) found ryegrass to be preferred over mustard, and phacelia and rapeseed residues were preferred over oats. Crotty and Stoate, (2019) found earthworm functional diversity changed dependent on cover crop mixture, with significantly more epigeic earthworms in mixtures with radish compared to bare stubble or oats and phacelia alone. Nematodes were also found to have twice the abundance in cover-cropped fields compared to fallowed (DuPont et al. 2009). In terms of increasing biodiversity, increases in biomass of one group of organisms will contribute towards an increase in total diversity, through stronger connections within the soil food web and the numerous species within that group. Overall, cover cropping definitely increases agroecosystem biodiversity and provided ecosystem services, but possible trade-offs and management complexity should not be underestimated (Daryanto et al. 2018).

Many studies consider the choice of cover crops that sustain the most diverse or most colonised AMF populations and how this subsequently affects the main crop (e.g., Hallama et al. 2019; Hannula et al. 2020), particularly in the response of P cycling (Reynolds et al. 2017) and as an additional N source (legumes). Adaption to site-specific conditions and management, including crop rotation and which cover crop species, need to be considered to maximise the benefits. The AMF-enhancing cover crops could constitute an alternative to inoculation (Cozzolino et al. 2013). Root and AMF hyphal fragments, which are important for early colonisation of the host plant, only survive for around six months in the soil (Tommerup and Abbot, 1981), therefore leaving the soil bare or with a non-mycorrhizal host can reduce colonisation rates of future crop hosts.

Intercropping

Similar to cover crops, the selection of species of plants in intercropping will affect the outcome. Intercropping with a mixture of non-mycorrhizal crops and mycorrhizal plants could help to sustain populations of AMF when the main crop is not mycorrhizal. In general, higher plant diversity leads to higher AMF diversity (Maherali and Klironomos, 2007). For example, tomato intercropped with leek showed 20% higher AM colonisation rate (for tomato) than tomato intercropped with tomato (Hage-Ahmed et al. 2013). Intercropping has also been found to support large earthworm populations through increasing the food supply throughout the year (Schmidt and Curry, 2001; Schmidt et al. 2003). These earthworm communities have even been found to be comparable to pasture and grass-legume leys with intercropping (Schmidt et al. 2001). However, besides the abovementioned positive potential, growing two or more crops simultaneously increases the complexity of management decisions (e.g., species combination or planting depth and date) and technical difficulties of harvesting and other field operations.

Grass/clover leys

Conversion from grassland to arable lowers the SOM content and stability of the environment, therefore introducing grass leys into a rotation will lead to positive SICS effects on biodiversity. For example, a two-year grass clover ley showed increases in earthworm abundance and diversity when compared to adjacent arable fields (Prendergast-Miller et al. 2021). Perennial fodder crops (e.g., grass, clover or lucerne) have been found to increase biomass and abundance of deep-burrowing (anecic) earthworms, improve soil structure and increase following crop yields (Kautz et al. 2010). Certain forage species are more favoured by certain soil fauna than others and can lead to increases in abundance and biodiversity (e.g., white clover instead of ryegrass (Crotty et al. 2015, Crotty et al. 2016)). Although, perennial ley crops in general have been found to increase earthworm numbers (Kautz et al. 2014). Native grasslands and plots where clover was grown had greater numbers of AMF compared to continuous wheat or barley plots (Menéndez et al. 2001). Cultivating clover after wheat restored AMF diversity and increased spore numbers over three years to resemble numbers in semi-natural grasslands (Oehl et al. 2003).

Agroforestry / agro-silviculture

It is well known that the perennial nature of most trees will have a profound impact on soil properties and hence, soil biodiversity, abundance, and function (Barrios et al. 2013) in any climate, although most research on the effects of agroforestry on soil biodiversity has focused on tropical regions. Biodiversity conservation is also one of the main reported ecosystem services/environmental benefits of agroforestry (Jose, 2009). However, a meta-analysis by Torralba et al. (2016) focusing on European soil biodiversity (fungi and arthropods) did not find significant enhancement within agroforestry. Most trees form symbioses with ectomycorrhizal fungi, but some also have AMF as a partner. In studies performed in the (sub)tropics it has been shown that presence of trees in plots increased sporulation, mycorrhizal colonization of the crop species and number of AMF propagules in the plant roots (e.g., da Silva Sousa et al. 2013). In the recent review of Marsden et al. (2020) agroforestry had mainly positive effects on fauna abundance and diversity, when compared to cropland, and neutral or negative when compared to forests. It would be interesting to monitor a newly started agroforestry area to assess how the soil biodiversity changes with time – moving from an arable/grassland based soil to a woodland, as to date this has not occurred within European climates.

Energy forests / Biofuel crops

In areas with limited choice of rotation crops, energy crops may be a useful addition for widening crop rotations and to generate a more diverse landscape. Where energy crops are perennial, negative effects of soil tillage are alleviated. Abundance and diversity of earthworms found in energy forests was greater than neighbouring arable fields, due to the absence of tillage, increase in OM layer and environmental buffering (Lagerlof et al. 2012). Earthworms were also found to increase in perennial energy crops in comparison to silage maize (Emmerling, 2014), and even the non-native *Miscanthus* had positive effects on earthworm communities (Felten and Emmerling, 2011). However, *Miscanthus* has been found to have a negative effect on earthworm abundance and biomass when compared to undisturbed meadow (Brami et al. 2020). Most of the plants used as biofuel crops are fast growing and benefit from forming AMF especially in low nutrient conditions or during drought. A study conducted in Canada identified that the abundance of AMF was significantly higher in the herbaceous perennial grasses (switchgrass and *Miscanthus*) than in woody species (poplar and willow) used for biofuel production (Mafa-Attoye, 2020). In this study, the addition of chemical fertilizers did not affect the colonization of AMF. However, where natural grasslands have been converted to bioenergy crops, the impact of land-use change is the main driver of biodiversity change (Desirée et al. 2014).

Physical soil environment and water

Soils provide microhabitats for soil organisms (for example, within the drilosphere, porosphere, detritusphere, aggregatusphere, rhizosphere and mineralosphere (Beare et al. 1995, Kandeler et al. 2019), but might be severely affected by soil management. Only a limited number of soil organisms are able to change the physical environment of their habitat (like earthworms burrowing and mixing the soil). Consequently, any changes in physical soil properties by anthropogenic actions may improve or reduce the functionality of a habitat for soil organisms. SICS can contribute to habitat function improvements for a diverse community of soil organisms, for example, by changing the balance between pore connectivity (substrate transport and availability of food resources), oxygen and moisture availability.

Tillage reduction

Tillage is performed to prepare the seed bed for the next crop, weed and pest management, and to mix plant residues/amendments with the soil profile. During tillage, an increased porosity "unlocks" OM, increasing its decomposability providing a temporary boost to the bacteria and fast-growing fungi that utilise OM. However, this is followed by a reduction in activity, once the most available OM is utilised (Kraus et al. 2017). This increase in porosity could also reduce the amount of moisture stored within the soil profile, reducing the soil's drought resistance and limiting soil biodiversity that rely on soil moisture for movement (bacteria, nematodes) or bodily functions (breathing through their skin). The heavy equipment used for tillage may cause subsoil compaction, which in turn can affect rooting ability and water infiltration. Compaction can lead to inundated soils during times of heavy rainfall, threatening crop yields and soil biodiversity. There is a general agreement that tillage intensity influences microbial abundance and function (Ahl et al. 1999, Kandeler et al. 1999a and b, Li et al. 2020). The major outcome of different studies showed that reduced or zero tillage changed spatial distribution of residues (*Figure 5*) leading to a re-distribution of soil microorganisms within the upper 40 cm of the soil profile. Reducing tillage has been found to lead to higher carbon and microbial biomass in topsoils compared to conventional tillage systems (Tully, and McAskill, 2020).

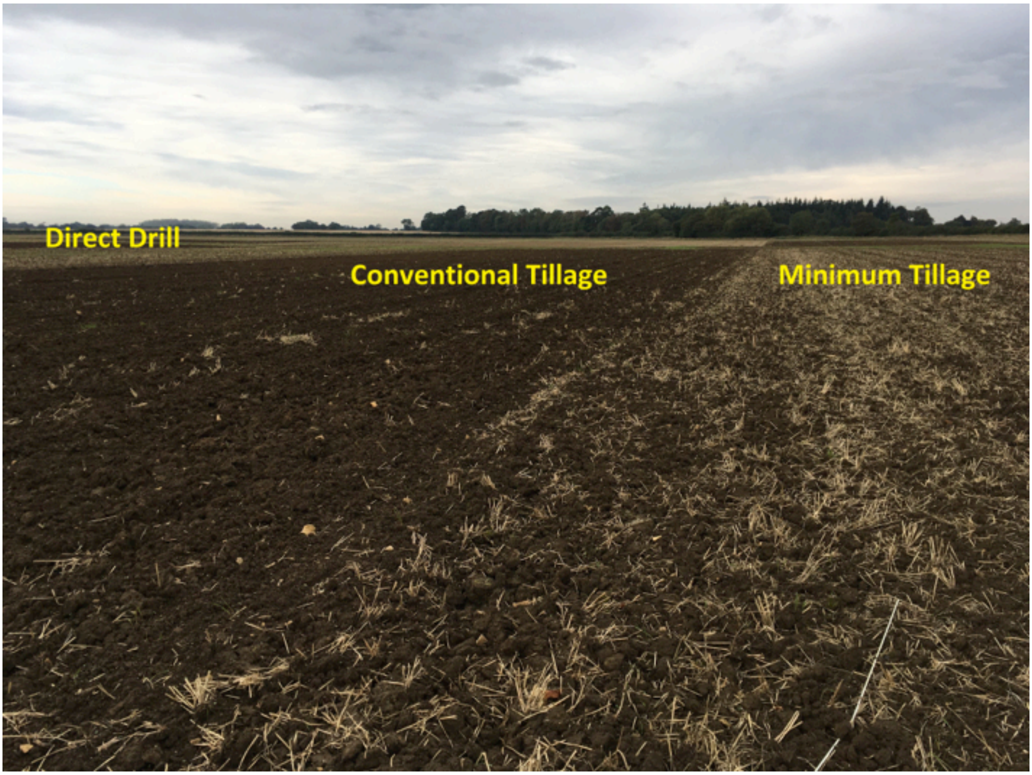

Figure 5. Photograph showing the difference in surface residue after three different kinds of tillage at the Royal Agricultural University long term experiment, Quarry Field, Harnhill, Cirencester. Tillage treatments visible in the photograph are direct drill, conventional tillage, and minimum tillage (labelled on photograph). Photograph taken by Dr Nicola Cannon (2019).

Kabir (2005) reviewed the impact of tillage practices on AMF (Arbuscular mycorrhizal fungi). Briefly, most of the studies found a reduction in the number of AMF taxa colonizing roots in systems with conventional tillage compared to reduced or no-tillage systems. There is also evidence for tillage changing community composition of AMF (Kabir, 2005; Jansa et al. 2003). Mechanisms for this include *(1)* the differences in tolerance to the tillage-induced disruption of the hyphae among the different AMF species; *(2)* changes in nutrient content of the soil; *(3)* changes in microbial activity; or *(4)* changes in weed populations in response to soil tillage (Jansa et al. 2003). The timing of tillage also influences AMF diversity – autumn tillage has been shown to cause reduced AM hyphal viability, whereas spring tillage had little effect (Kabir et al. 1997a, Kabir et al. 1997b). This is likely caused by the hyphae being detached from the host plant in the autumn, whilst in spring AMF is mostly found as spores.

All soil fauna impacted by ploughing will benefit from no-till or reduced tillage management (Orgiazzi et al. 2016). Frequent studies have shown earthworms to be negatively affected by tillage, as found in the global meta-analysis by Briones and Schmidt (2017). The detrimental effect of tillage on earthworm community composition and abundance is often dependent on the intensity (Emmerling, 2001) and frequency – the less intensively the soil is disturbed, the less harmful tillage is for earthworms (Bertrand et al. 2015). Tillage is known to be detrimental to other soil fauna including Collembola (Bedano et al. 2006a), Acari (Bedano et al. 2006b) and to a lesser extent nematodes (Fiscus and Neher, 2002), overall impacts of tillage regime on soil biota seem to be group-dependent (van Capelle et al. 2012). Tillage is commonly used within organic farming, negating some of the benefits that soil biodiversity should be deriving from organic amendments and practices. Promoting occasional reduced tillage within organic systems has been shown to have immediate positive effects on earthworm populations (Moos et al. 2016). Tillage is often used as a simple method to lessen surface soil compaction (although could be transferring the problem to the subsurface, creating a plough pan). Plants grown on compacted soil are exposed to a multi-stress environment (Colombi and Keller, 2019) limiting soil fauna's ability to access water and food, as well as changing the habitable pore space the fauna live within. Spike aeration or the use of different tine options to alleviate compaction in wheelings of arable crops (Niziolomski et al. 2016) or within pasture (Cournane et al. 2011) has the potential to improve soil structure, whilst also reducing runoff, phosphorus, and nitrogen losses (DeLaune et al. 2013). These methods have the potential to reduce compaction without disturbing the whole soil environment to the same extent as tillage. However, to date the impact of these other soil improving cropping systems on soil biodiversity loss has not been monitored.

Drainage management

Drought decreases soil water content and has been found to decrease microarthropod species richness, whilst irrigation has been found to increase microarthropod species richness (Tsiafouli et al. 2005). Drainage and irrigation have been shown to encourage multiplication of the more robust species of the Acari (Prostigmata and Astigmata) (Behan-Pelletier, 2003). Oribatida have also been found to increase in abundance and species richness with increasing soil moisture (Jakšová et al. 2020). Soil drainage has also been found to impact the community structures of actinomycetes and pseudomonads (Clegg et al. 2003).

The porosity of the soil will influence the likelihood waterlogging will occur, with a large fraction of micropores within the soil profile leading to poor aeration and a higher risk of waterlogging; whereas a high proportion of conducting continuous macropores will lead to the soil drying out more rapidly, effecting the soil habitat for the biodiversity residing within it. Water infiltration is an ecosystem service the soil provides, which can be affected by poor soil management and when the infiltration rate is low, it can lead to surface runoff, erosion, and flash flooding. Earthworms as ecosystem engineers can change the water infiltration rate of a soil through their burrowing activity and the creation of permanent burrows (Crotty, 2020). Research establishing and quantifying the relationship between earthworms and water infiltration capacity of soils has been occurring over the last fifty years (Griffiths et al. 2018). Controlling the drainage status of soil is necessary for preserving a suitable soil structure (Mueller et al. 2013) for soil biodiversity and crop growth.

Chemical amendments

All chemical amendments (pesticides, synthetic fertilisers, other chemical amendments) have an impact on soil biodiversity, irrespective of target pest or reason for applying (Karpouzas et al. 2014). The likelihood of pesticides affecting non-target soil biodiversity is dependent on mode and timing of application, as well as the climatic conditions at the time (and after) (Karpouzas et al. 2014). Due to the different ecological functions and preferences of the different taxa residing within the soil, some chemical amendments (e.g., liming or synthetic fertilisers) may promote the retention of some groups at the deficit of others. These conflicts in requirements by soil organisms indicate that more work is needed to understand which SICS might be best to reduce soil biodiversity loss.

Plant protection products (pesticides)

All pesticides have wider implications than their target pest (Crotty, 2020). For example, insecticide seed dressings or granules will also reduce soil faunal populations, including springtails, mites, beetles, fly larvae, psocopterans, nematodes and earthworms (Pisa et al. 2017). Molluscicides have been found to be deleterious to non-target soil invertebrates to a certain extent (e.g., metaldehyde (Santos et al. 2010) or ferric phosphate (Langan and Shaw, 2006; Edwards et al. 2009)). Reducing the number and amount of slug pellet applications, and following manufacturer's instructions, will reduce the risks of soil biodiversity loss.

In general, nematodes can be controlled using nematicides, while soil fungicides are much less available. Nematicides have been shown to impact all nematode trophic groups (Timper et al. 2012), this includes those nematodes that predate on plant pathogens (Hofman and Jongebloed, 1988), as well as the other nematode functional groups. This may have a lingering effect due to the changes in the soil food web of other invertebrates over the same time-period (niche filling). Herbicides have also been found to negatively affect soil fauna – for example glyphosate has been found to decrease root mycorrhization and AMF spore biomass (Zaller et al. 2014); inhibited bacterial growth (Aristilde et al. 2017); and reduced earthworm activity and reproduction (Gaupp-Berghausen et al. 2015). Miller and Jackson (1998) showed that weeds are important hosts maintaining AMF when growing non-mycorrhizal crops, forming a mycorrhizal bridge between crops. There is a significant body of literature about the negative side-effects of applying soil pesticides (Siepel, 1996, Firbank et al. 2008). Therefore, it is important to create an integrated pest management strategy, applying pesticides through "spot application" or when an outbreak is greater than the "economic threshold" of the damage occurring. In general, the use of synthetic chemical pesticides is increasingly criticized because of residues remaining in the environment and food chain, and because of the threat to biodiversity. Researchers need to help farmers by developing state-of-art automated and digitally networked technologies and find solutions so that synthetic chemical pesticide use can be abandoned in the future. At the same time, it might still be required to use mineral fertilization to guarantee high biomass yields. These cropping systems without the use of pesticides, but with mineral fertilizers represent a complete reorientation in agricultural production of food and feed.

Other chemical amendments

Soil pH has been found to be the single most significant edaphic variable to predict bacterial community composition (Fierer and Jackson, 2006) and thus can affect fungal and bacterial feeding organisms – cascading through the soil food web. Reducing the acidity of soil through liming has been found to promote earthworm abundance in the field over time (Hirth et al.

2009). However, some studies have found other soil taxa to be detrimentally affected (e.g. fungi (Murray et al. 2006, Hannula et al. 2021), Collembola (Chagnon et al. 2001); and Acari (Hagvar and Amundsen, 1981)). Liming is an appropriate measure to control *Plasmodiophora brassicae*, the causal agent of club root on brassicas. In a recent study, long term liming was the single agricultural treatment that caused the biggest change in soil fungal community structure (Hannula et al. 2021). The addition of gypsum (mined or flue gas desulphurised), is another amendment that has been found to reduce earthworm abundance and biomass in some instances (Chen et al. 2014). These conflicting results on the effects of liming on different forms of soil biodiversity, indicate more research is needed on the impact of liming and other soil amendments on soil biodiversity loss.

Industrialised animal production has led to large increases in the release of antibiotics to the soil environment. Besides potential shifts in the soil microbial community, the emergence of antibiotic resistances is a major concern for public health (Cycoń et al. 2019). Other pollutants, such as heavy metals (Thomas et al. 2020) and pesticides (Kurenbach et al. 2018) might also cause similar effects. It is also important to note, that the soil microbial community is a major reservoir for novel antibiotics (Ling et al. 2015). The use of plastics in agriculture and horticulture, as well as the use of municipal waste compost results in large amounts of plastics in the environment. Microplastics are considered an emerging threat to soil biodiversity (Tibbett et al. 2020). The effect of large amounts of chemically quite diverse plastics and their decomposition products on the soil biota is currently under investigation (Schöpfer et al. 2020, Li et al. 2021). For example, microplastic particles (smaller than 5 mm) differing in their shapes (spheres, fibres, and fragments) may harm soil organisms (Bläsing and Amelung, 2018).

Synthetic fertilisers

AMF have been described as natural biofertilisers (Berruti et al. 2016), however most farmers do not consciously rely on this biological function. Negative effects of synthetic fertiliser application on soil biodiversity can be due to the increased soil acidification. Additionally, the AMF community is usually less diverse in agricultural systems due to the over-supply of P, reducing the dependence of crop plants on the symbiotic mutualisms with fungi (Liu et al. 2016). Also, the form in which fertilisers are added has an influence on the AMF community and organic fertilisers are thought to be more favourable for AMF.

Application of fertilisers can have an effect on soil faunal populations, this can be both positive (application of a food source for soil organisms to utilise) or negative (soil acidification – changing the pH of the soil to have less hospitable conditions) (Crotty, 2020). The use of inorganic fertiliser has been found to reduce the abundance of Collembola, Oribatid mites, Enchytraeidae, and earthworms (Siepel, 1996; Yeates et al. 1997), although direct effects are limited. Earthworm abundance and diversity increases with OM input and content of soils, therefore when an inorganic mineral fertiliser source is used instead of an organic, variety numbers are reduced (Lapied et al. 2009). Nematode abundance was also found to be reduced when inorganic fertiliser and pesticide inputs where combined, compared to organic production combined with minimum tillage (Overstreet et al. 2010).

Biological amendments

Many biological amendments have large-scale benefits to soil biodiversity in general, however they can also involve the greatest changes within a farming system. For example, the change from conventional to organic agriculture is not just a change in one practice (unlike a change from ploughing to direct drilling where all other actions remain the same); this is also not going to occur across all farming systems. Organic farming is often promoted as a way to enhance the long-term sustainability of modern agriculture whilst decreasing environmental impacts (Bedoussac et al. 2015). However, changing large areas from conventional to organic farming will not maintain current yields, leading to an increase in the area farmed (Smith et al. 2018), increasing greenhouse gas emissions (Smith et al. 2019). Although other studies have found that large-scale conversion to organic farming could improve food security and sustainability on a global scale (Wilbois and Schmidt, 2019; Röös et al. 2021).

Soil biodiversity is known to increase when more natural amendments and practices occur, along with other soil improvements (increased SOM, water holding capacity, reduced erosion). However, given the current market pressures and agricultural policies, it is naïve to expect the majority of farmers to change in this way in the short term; a more practical option would consist in encouraging the use of biological amendments in conventional agricultural practice. Farmers should consider organic "farm waste" as valuable resources, and their application as biological amendments could improve soil biodiversity, whilst maintaining high yields and profitability.

Other biological amendments have been tested in microcosm experiments (e.g., biochar, mycorrhizal additions, microbial inocula), which do not represent field conditions and have not focused on the impact of these additions on native soil biodiversity – this research gap needs to be further investigated before these SICS can be invested in at the field scale (Bradáčová et al. 2019, 2020, Eltbany et al. 2019). However, a recent study by Clocchiatti et al. (2020) did show that the addition of deciduous wood sawdust and paper pulp stimulated an increase in fungal biomass within arable soil in microcosms – suggesting these biological amendments will help soil biodiversity through food addition and stabilisation of the environment, warranting further investigation.

Organic fertilisers

The application of organic fertilisers, like farmyard manure or slurry, benefit the soil biota more than mineral fertilisers, partly because the organic matter represents a source of mineralisable nutrients. However, there can be issues with the organic fertiliser containing heavy metals, antibiotics, or pathogens. The application of animal manures as nutrient sources has generally been found to increase the abundance and activity of soil biota (particularly nematodes, Collembola, Acari and earthworms) (Altieri, 1999; Birkhofer et al. 2008, Wu et al. 2013, Orgiazzi et al. 2016). Organic amendments will stimulate soil microbial activity, thereby potentially increasing the disease suppressiveness of the soil as soil-borne pathogens are out-competed. For example, long-term application of swine slurry, whilst maintaining productive crop yields, also influenced AMF and their products (glomalin) in the soil environment (Balota et al. 2016), likely through changes in soil aggregation. Utilisation of organic fertilisers has the potential to convert a farm waste into a benefit (Crotty et al. 2018), making the farming system more sustainable, as well as acting as a SICS to retain soil biodiversity.

Anaerobic digestate has a decreased pathogen load in comparison to manure/slurry, due to the digestion process, pathogen reduction increases if the process is thermophilic (Insam et al. 2015). The addition of anaerobic digestate (in the form of labile organic matter) provides a large substrate source for microorganisms and has been found to increase microbial biomass within the soil (Nkoa, 2014). Digestate does not appear to have a negative effect on earthworms exposed directly to it at low concentrations (Pivato et al. 2016), although negative effects were found at high concentrations (Rollett et al. 2020).

Biochar amendments

There have been few studies on the effect of biochar on soil organisms and even fewer in temperate soils (Mackie et al. 2015). Those that have investigated the addition of biochar have shown that microbial biomass is increased (Lehmann et al. 2011, Bamminger et al. 2016). However, AMF abundance did not increase if there was an already abundant nutrient supply (i.e., in agricultural environments). Experiments manipulating the number of earthworms and biochar showed that rice yields increased the most when both were added together (Noguera et al. 2011); however, this does not discuss how earthworm populations are affected at the field scale. Currently, the application of biochar is often combined with compost amendments to be sure that biochar with its high sorption capacity does not interfere with plant uptake of nutrients. Research is ongoing and there are concerns that the application of recalcitrant biochar entails risks, as potential negative effects would be persistent for a long time.

Mulching

Mulch is usually plant material that is partially decomposed left on the soil surface to form a cover, it can be a living mulch, green residue that has been allowed to die back or applied organic material. The mulch provides a large surface area for pesticide sorption and/or can be used by soil microorganisms as co-substrate for co-metabolic degradation of pesticides (Aslam et al. 2018). In addition, mulch reduces water infiltration rates, and soil erosion by creating an organic barrier on top of the soil surface. Organic mulch biomass is a source of C and nutrients required for soil biological activity (Orgiazzi et al. 2016). A variety of mulches can be used in-row in perennial horticultural cropping systems. These mulches have been found to increase the abundance of protozoa, bacterivorous nematodes and enrichment opportunistic nematodes in comparison to bare ground or polyethylene covering (Forge et al. 2003). Long term use of living mulch and organic fertiliser have been found to increase earthworm populations by between 1.5-2.3 times greater than conventionally fertilised populations (Pelosi et al. 2015). Mulching can also reduce the spread of some air-borne pathogens (Litterick et al. 2004) e.g., Botrytis in strawberries.

Mycorrhizal amendments

The use of commercial (laboratory grown) inoculants containing non-resident AMF is an emerging technology in field crop production, especially in systems with lower input of P-containing fertilisers. Adding AMF to soils can enhance crop yields and protect plants from biotic and abiotic stresses (Boyer et al. 2015). For example, AMF addition has been found to alleviate the stress of soil compaction in wheat and corn (Miransari et al. 2007, Miransari et al. 2008). However, carrying out an open-field, extensive inoculation treatment is often technically impractical, economically prohibitive and is advised only if the native AMF population is not present or has very low diversity, to avoid possible negative effects on the soil biota. Indeed, the effects of inoculants on native soil populations is largely unknown and needs to be understood before large scale amendments are done (Rodriguez and Sanders, 2015). Adding an AMF inoculum has been shown to be an effective and economical option to restore degraded soils with little of its native biodiversity left (Berruti et al. 2016). However, minimal research has occurred on the effect of mycorrhizal amendments on soil biodiversity in general. More research is also necessary to address inoculation-related biosafety concerns (Mitter et al. 2021).

Biocontrol

Addition of nematodes as a biocontrol agent in mollusc reduction has been shown not to affect earthworm or mesofauna abundance (Iglesias et al. 2003); although there can be some problems with efficacy in comparison to metaldehyde or iron phosphate (Rae et al. 2009). Two biocontrol agents *Streptomyces griseoviridis* and *Trichoderma* spp., are regularly used in protected cultivations against a range of fungal and chromist soil-borne pathogens. However, these products are unable to control existing high populations of pathogens. Brassicacea-based (allelopathic) management strategies, are a soil biodiversity-friendly alternative to nematicides, and have been shown to be effective at managing the top-three economically important nematode pests; root-knot (Meloidogyne), cyst (Heterodera and Globodera) and lesion (Pratylenchus) nematodes (Fourie et al. 2016). Acting as a form of biofumigation, brassica residues form toxic compounds (isothiocyanates) during their decomposition (Larkin and Griffin, 2007). Hatch crops (particularly *Solanum sisymbriifolium* and *Tagetes*) are another non-chemical method that are often used in Europe to combat potato cyst nematodes and Pratylenchus. AMF can also be considered to act as a type of biocontrol agent, as well as exchanging carbon with the host plant it can also help to defend the plant from pathogens (Johansson et al. 2004).

Discussion

Agricultural intensification (soil tillage, increased mineral fertiliser usage and crop diversity reductions (Postma-Blaauw et al. 2012)) has been shown to affect abundances of taxonomic groups with larger body sizes (earthworms, enchytraeids, microarthropods and nematodes) more negatively than smaller-sized taxonomic groups (protozoans, bacteria and fungi) (Postma-Blaauw et al. 2010). However, the effect agricultural intensification has on the whole soil food web needs to be considered, global studies of the threat to soil biodiversity show there are large areas at risk (*Figure 6*).

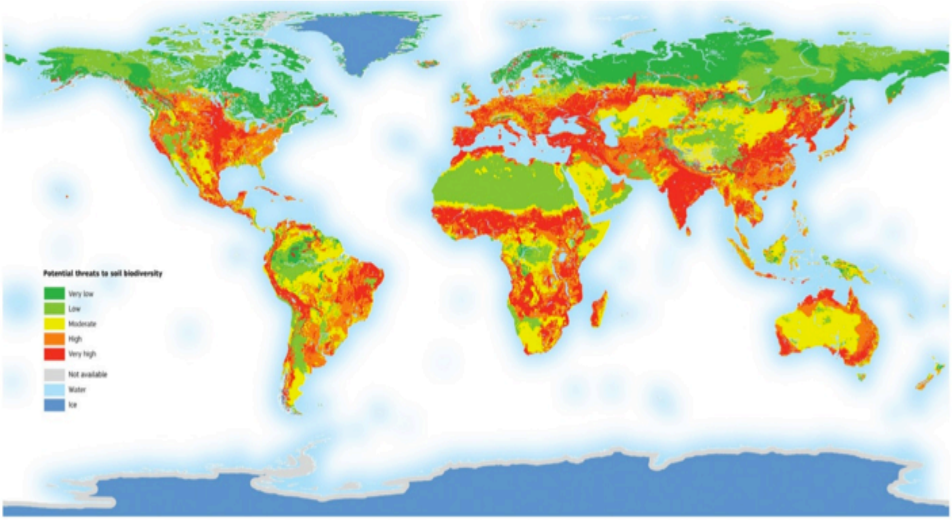

Figure 6. Map of global threats to soil biodiversity (Orgiazzi et al. 2016)

Intensively managed agricultural soils are among the poorest soils in terms of biodiversity of microbes and fauna, and the lack of diversity is likely to reduce the capacity of the soil to function (Bedano et al. 2006). Measures to extensify agricultural practises need to be taken in order to increase species diversity and the connections between them (Morriën et al. 2017) and reach a ‘healthy’ well-functioning soil, this is essential for the provision of ecosystem services. There should be a holistic approach to maintain biodiversity focusing on the inclusion of many groups of soil organisms and their interactions. From a methodological viewpoint, we can detect relative shifts, e.g., using molecular markers, but face difficulties giving absolute abundances and connecting them with ecological relevance. An urgent measure would be the incorporation of soil biodiversity measurements in national soil quality monitoring schemes. A two-way strategy combining emerging cutting-edge methods, like metabolomics (Withers et al. 2020), with “ground truth” relevant for practitioners, such as visual soil assessment and spade tests (Junge et al. 2020), as the provided summary parameters are tightly related to soil biodiversity.

Increasing soil biodiversity could also be thought of as a soil improving cropping system by itself; numerous studies (e.g., van Groenigen et al. 2014 and the references therein; Bender and van der Heijden, 2015) found that increasing soil biodiversity can increase crop yields and decrease nutrient losses, being therefore directly linked to the sustainability of agriculture. However, as seen in *Table 3* this view has not been widely considered by academics researching agricultural cropping systems. Going forward, academics researching soil biodiversity should try to measure yield within experiments assessing relevant farming techniques and report results also in similar terms as agronomists – to emphasise the importance of soil biodiversity to sustainable agriculture. Interestingly, advocates of emerging alternative agriculture movements, i.e., agroecology, conservation, or regenerative agriculture, base their claims of an increased sustainability of their systems frequently on alleged positive effects of increased biodiversity.

There are substantial knowledge gaps when it comes to practical recommendations to farmers, despite noteworthy potential synergies of the combinations of different SICS. For example, cover crop management is key for successful conservation agriculture (Mirsky et al. 2012), but limited management experience of this system results in higher risks and the possibility to make mistakes with these knowledge-intensive systems (Zikeli and Gruber, 2017). Also, the potential detrimental effects on soil biodiversity of an increased use of herbicides in response to the weed pressure in no-till systems may play a role in the mixed results of field experiments (Kremer and Means, 2009; Nguyen et al. 2016) and weed control is one of the greatest problems of herbicide-free no-till systems (Zikeli and Gruber, 2017). In practice, farmers that experiment with no-till techniques often adopt a modified direct seeding approach, using shallow non-inversion tillage in emergency situations (*Figure 5*), e.g., when the cover crop did not emerge due to unfavourable climatic conditions. This example illustrates that every management technique of a SICS comprises a wide spectrum of possibilities, making absolute recommendations difficult, but offering opportunities to fine-tune and adapt SICS to the local conditions.

Figure 7. Photograph showing example of diversity extracted from a soil core. Photograph taken by Dr Felicity Crotty, 2010, (this photograph was the winning entry in the BBSRC Science Photo Competition – Agriculture, Food, Diet and Health Category).

It is often suggested that there is a greater diversity living below-ground than there is above-ground, with soil being referred to as “the poor man’s tropical rainforest” (Giller, 1996) (*Figure 7*). It has been estimated that there are over 12.8 quadrillion (10^{12}) soil invertebrates in the top 8 cm of soil across the UK (Countryside Survey, 2007). However, it is not just the biodiversity that is important but also the interconnectance of the soil food web. The feeding relationships among the soil fauna and microbes form an intricate soil food web (Hunt et al. 1987) which ranges over multiple scales (micro, meso and macro) (Swift et al. 1979) with orders of magnitude differences in size between organisms (*Figure 8*).

Figure 8. Photograph showing difference in size of organisms living within the soil. Photograph taken by Dr Felicity Crotty, 2010, shortlisted Microflora and Mesofauna category – Global Soil Partnership and FAO, Soil Biodiversity Photo competition 2020. On the right is a pseudoscorpion (moss neobisium), a tiny predator that lives within the soil and litter layer, ensnaring a Symphypleona springtail (Collembola), part of the mesofauna, within its pincers.

Conversely, due to the large diversity of species there is also a considerable amount of functional redundancy (i.e., the same function is being performed by multiple distinct groups of organisms). This functional redundancy creates an inbuilt resilience of soil biodiversity to perturbations (Bengtsson, 2002). The diversity, abundance and complexity of the soil make understanding the interactions and effects of SICS multifaceted, with organisms interacting and affecting each other, as well as reacting differently to the different SICS. In view of the multiple ecosystem functions of soil biota, a holistic approach of increasing biodiversity through agronomic management as opposed to a reductionist approach focusing on single species or inoculation may be more appropriate (Fester and Sawers, 2011). Several groups, including mycorrhiza can be considered “umbrella” species for agroecosystems, as several management techniques that enhance these groups have other beneficial effects on other species and in the agroecosystem. Therefore, a broader approach by embracing agroecology, where farmer–scientist alliances co-create and exchange knowledge, transforming the research system of agronomists and ecologists (Levidow et al. 2014), while producing relevant results for agriculture might allow the development of more sustainable management systems. To enable better research, an interdisciplinary approach is needed, to consider soil biodiversity (ecologists) and what happens if it is lost in relation to crop yield and sustainability (agronomists).

Soil organisms can improve the soil’s capacity to function, through nutrient cycling, changing the structure of the soil – increasing porosity, reducing compaction as well as redistributing nutrients (*Table 2*; *Figure 4*). Without soil organisms, there would likely be reductions in crop yields, however lower crop yields due to reduced soil biodiversity are often masked by increased fertiliser (or other inputs) applied. Changing the focus of agricultural management to include soil biodiversity will reduce the risk of soil biodiversity loss (*Table 1*; *Figure 1*). However, it is important to consider the whole soil environment as soil type / texture will also have a large effect (*Figure 3*). In most cases, soil properties (i.e., type, texture) and climate dominate the abundance and diversity of soil biota, but constitute fixed properties, while management can be adapted to improve the provision of ecosystem services.

Within this review, we have focused on a range of organisms (mycorrhiza to earthworms; *Figure 7 and 8*), through this range of scale and lifecycle we can gauge the impact of agricultural management and have postulated potential SICS that can reduce the risk of soil biodiversity loss without compromising yields (*Figure 2*). Many of these SICS also reduce the risk of other soil threats (e.g., soil organic matter loss, soil erosion, soil compaction). These threats are all reduced if the whole agricultural management system is considered rather than just short-term yield benefits that won’t last for future farming generations. Therefore, external benefits for society, as water, soil, and biodiversity protection, as well as an increased C sequestration, which are currently not reflected in the market value of the crops, need to be considered when establishing the price of an agricultural production (Dendoncker et al. 2018).

Conclusions

Ecological studies have shown that greater plant diversity suppresses plant disease and promotes increased overall resistance and resilience of the ecosystem; however, this has not been shown in relation to soil biodiversity and therefore these theories need rigorous testing in agricultural settings at field scale (Vukicevich et al. 2016). Also, the links between different ecosystem services, especially the trade-offs and synergies with biodiversity need to be investigated. As biodiversity affects other ecosystem functions, agroecological management techniques that promote soil biodiversity and soil biological functions, indirectly affect other ecosystem services. This review has highlighted that agricultural research and soil biodiversity research needs to be brought together, so that the impact of soil biodiversity loss can be given more prominence, both to reduce biodiversity loss but also to improve the sustainability of farming systems. Selecting and adapting specific cropping systems designed to maintain or increase soil biodiversity, promoting the stabilisation of the soil environment, reducing chemical amendments, increasing biological amendments, or introducing novel technologies that optimise and reduce inputs are all potential SICS that can be utilised. This review has shown the range of SICS that can be utilised to reduce the threat of soil biodiversity loss and highlighted the research that is currently ongoing within these SICS in relation to soil biodiversity and the impact this will have on the sustainability of agricultural management.

Acknowledgements

The authors would like to thank Aad Termorahuizen for advice in the manuscript. The authors received funding from the European Union’s Horizon 2020 research and innovation programme under grant agreement No 677407 (SoilCare project) to write this book chapter.

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Chapter 13

Biochar application in an integrated approach to sustainable soil management in Taiwan: Nutrient retention and availability in excessive compost-fertilized soils

Chen-Chi Tsai and Yu-Fang Chang

Summary

In Taiwan, due to the high temperature and high humidity environment, farmers who apply chemical fertilizers (chemical or organic fertilizers) to farmland soils are prone to leaching and loss due to heavy rainfall, or rapid mineralization and decomposition due to high temperatures, reducing fertilizer efficiency and reducing crop yields. Therefore, farmers often apply excessive fertilizers in pursuit of high yields. However, long-term excessive fertilization has caused most soil acidification, degradation, and reduced soil biodiversity. In this chapter, we introduced a case study with focus on the changes of available nutrients retention and availability in excessive compost-fertilized soils as affected by biochar feedstock and rate, and soil conditions is reported. Three soils (one Oxisols and two Inceptisols) and three slow pyrolysis (> 700°C) biochars (Formosan ash (*Fraxinus formosana* Hayata), ash biochar; makino bamboo (*Phyllostachys makino* Hayata), bamboo biochar; and lead tree (*Leucaena leucocephala* (Lam.) de. Wit), lead tree biochar) were selected for incubation experiment, and the entire experimental process is presented and discussed. We hope that the co-application of biochar in the over-fertilized soil in Taiwan can reduce the decomposition rate of compost, prolong fertilizer efficiency, reduce soil nutrient leaching loss, and further restore soil biodiversity, as well as for assisting sustainable soil management, aiding the achievement of multiple sustainable development goals (SDGs), maximizing the benefits of biochar applications, and minimizing the potential environmental risk.

Introduction

As affecting by intensive agricultural development and (climate) environmental change, the steady increase in fertilizer use (inorganic and organic) in many parts of the world has resulted in severe land degradation, shrinking land area per capita, and declining soil quality (Agegnehu et al. 2016), and may threaten the continued development of agriculture (Barrow, 2012). A new doubly green revolution has been proposed (Barrow, 2012), with the potential for increasing yields, reducing negative impacts, better sustainability, and all this accessible to poorer farmers as well as commercial producers; and in this aspect, biochar could be the key input and a valuable soil amendment. The additional benefits from biochar including liming effect, soil structure improvement, nutrient retention, microbial habitat and carbon (C) sequestration may also provide productivity and financial rewards to farmers (Limwikran et al. 2018). In addition to C sequestration, the amendment of charcoal (biochar) will make a lasting contribution to soil quality (Laird, 2008), including the potential for reducing nutrient leaching loss and limiting the bioavailability of heavy metals, which in turn can reduce fertilizer needs and environmental risk (Biederman and Harpole, 2013). Biochars may influence chemical and physical properties of the entire soil, and affect soil biodiversity. Lehmann et al. (2011) indicated that the release of a variety of organic molecules from fresh biochar in the short term may in some cases be responsible for increases or decreases in abundance and activity of soil biota. Including sorption phenomena, pH and physical properties of biochars such as pore structure, surface area and mineral matter play important roles in determining how different biochars affect soil biota (Lehmann et al. 2011). Additionally, the sustainable development goals (SDGs), framed by the General Assembly of the United Nations in 2015, are a set of 17 goals and subdivided into 169 targets for tackling environmental, economic, and socio-political issues, and a deadline has been set to achieve them by 2030. Including the production, the importance and application of biochar can help in partial achievement of some of these goals for saving the world from the threats continually degrading the environment (Kumar and Bhattacharya, 2021), especially in achieving goal 3 (Good health and wellbeing), 6 (Clean water and sanitation), 11 (sustainable cities and communities), 12 (Responsible consumption and production), 13 (Climate action), and 15 (Life on land) of the SDGs.

However, nutrient responses depend on various biochar-soil combinations. The effect of biochar co-applied with compost on soil nutrient was varied, depending on the nutrient, soil, compost, or biochar; additionally, it seemed that increasing nutrient content by biochar and compost indicated that these amendments contribute more nutrients or make nutrients more available (Berek et al. 2018). Limwikran et al. (2018) indicated that application of about 10 Mg biochar ha^{-1} to agricultural soils will provide substantial amounts of plant nutrients, and the nine agricultural waste products based-biochars are likely to be quite effective sources of K and P, and conversely, biochar acts as a sink for considerable exchangeable soil Ca which may have agronomic consequences for Ca-deficient soils. The combined additions of biochar (2% by wt.) and compost (2% by wt.) significantly increased pH and EC; reduced exchangeable Al; reduced Mn and Fe in the Oxisols; increased P, K, and Ca content of the soils; and increased Ca, Mg and Fe uptake (Berek et al. 2018). In addition, the decline in the mobile contents of Al, Zn, Cu, Mn, and Cd (0.01 M $CaCl_2$ extractable) was significant in all five biochar-amended soils of 8% biochar (willow tree chips pyrolyzed at 700°C by fast pyrolysis) rate (Hailegnaw et al. 2020), indicating that biochar could efficiently reduce the mobility of Al, Zn, Cu, Mn, and Cd in soil, while the decline is mainly caused by biochar-induced changes in soil pH, CEC, DOC, and exchangeable Ca^{2+} and K^+ content of treated soils. However, there has been little work on the impact of higher biochar rate on soil nutrient retention and availability in the condition of excessive compost.

Due to the high precipitation (> 2500 mm per year) and warm temperature (average about 23~25°C per year) resulting in significant soil erosion, nutrient leaching, and rapid decomposition of soil organic matter (SOM), Taiwan's farmers often apply excessive compost (2~5%, by wt.) during intensive cultivation periods to improve soil low nutrient status, increase SOM, and lift crop productions, but in contrast frequently result in soil deterioration and negative environmental impacts (Tsai and Chang, 2020a, 2021). In the condition of excessive compost, high-temperature pyrolysis biochars have similar potential advantages in stabilizing the excessive labile organic matter from compost, and higher biochar rate (5%, wt.) has no significant decrease in soil nutrients content and has no obvious risk in increasing Cu, Pb, and Zn content, suggesting the fewer detriments to studied soils (Tsai and Chang, 2020a). Furthermore, a higher biochar rate plays a significant role in reducing the ammonium and nitrate in biochar-amended soils co-applied with excessive compost, and the immediate effects of the three studied high-temperature biochars on soil ammonium and nitrate were negative, irrespective of soil properties (Tsai and Chang, 2021); in addition, over time, the reduction amount of soil ammonium and nitrate becomes soil-specific and is primarily determined by the soil's original pH and texture. Therefore, excepting for increasing C sequestration and N retention, in the condition of excessive compost, it is imperative to delineate the response of nutrient retention and availability to different biochar feedstocks, biochar rates, and soils, in future for reducing compost application, preventing soil deterioration, and negative environmental impacts. A 400-day laboratory incubation study was conducted with three well-characterized biochars mixed with three soil types under constant laboratory incubation conditions, similar to previous study (Tsai and Chang, 2021). The current study was focus on the changes of available nutrients retention and availability in excessive compost-fertilized soils as affected by biochar feedstock and rate, and soil conditions. We hypothesized that adding biochar may retain nutrients and reduce the solubility of heavy metals (copper, lead, and zinc), thereby increasing the utilization rate of compost, reducing soil nutrient loss and heavy metal pollution, and restoring soil from degradation; and further protect and preserve the soil biodiversity. Like this, the current study results could achieve the goal 3, 6, 11, 12, and 15 of the SDGs.

Characteristics of soils, biochars and compost

The climate of Taiwan is characterized by high air temperature and heavy annual rainfall in association with complicated parent materials of soils. The agricultural soils are intensively tilled for crop production to be satisfied with the large population (about 23 million) in Taiwan. The soil degradation impacted from above natural conditions and human activities are therefore produced. The total area of cultivated soils in Taiwan is about 880,000 ha (24% of the total area), while over 80% of the cultivated soils are potentially degraded, including soil erosion on slope land, soil acidification, poor drainage, soil compaction, soil salinity, deficiency of micronutrient, water shortage of sandy soil, and heavy metal contamination. Acid rain and chemical fertilization are major factors to cause the soil acidification ($pH < 5.6$) in Taiwan. In general, more than 50% of the acid rain in Taiwan was significantly affected by North-Eastern flow and frontal passage in the dominant precipitation systems. Moreover, serious acid rain in Taiwan often occurs in the highly urbanized and industrialized areas with mean pH values of 4.5. Additionally, regarding chemical fertilization, approximate one million tons of chemical fertilizers of nitrogen, phosphorus, and potassium are applied annually on agricultural soils in Taiwan, which causes a clear increase of acidity in these soils under strong intensities of cropping due to the replacement of base cations by hydrogen ions.

The characteristics of the studied three soils and compost were analyzed and described in previous studies (Tsai and Chang, 2019). In brief, the characteristics of three studied soils, including Pingchen (Pc) soil [red earth, slightly acidic Oxisols (SAO)], Erhlin (Eh) soil [fluvo-aquic soil, mildly alkaline Inceptisols (MAI)], and Annei (An) soil [fluvo-aquic soil, slightly acid Inceptisols (SAI)], include: SAO soil was clay-textured soil with pH 6.1, MAI soil was clay loam-textured soil with pH 7.5; SAI soil was clay loam-textured soil with pH 6.5 (*Table 1*). The SAO, MAI, and SAI soils were collected from Taoyuan county, Changhua county and Tainan county, located in northern, central and southern Taiwan, respectively. According to the occupied area in rural land, these three studied soils are the top ten rural soils in Taiwan. The occupied area is 17,734, 21,158, and 12,754 ha for SAO, MAI, and SAI soil, respectively. The main raw materials (> 50 wt %) of the studied compost were poultry manure (mostly chicken) and livestock manure (mostly swine), a commercial product (organic fertilizer; Tianluo Composting Plant, Changhua, Taiwan) certified by the government and often used by farmers. As shown in *Table 1*, briefly, the characteristics of studied compost include alkaline (pH 8.41), low C : N ratio and CEC, abundant M3-P, K, Ca, and Mg content, and relatively higher M3-Zn content.

Characteristics	**Compost**	**SAO**	**MAI**	**SAI**	**A biochar**	**B biochar**	**L biochar**
pH[1]	8.41	6.1 / 5.0	7.5 / 7.2	6.5 / 6.2	10.3	10.6	9.9
Electrical Conductivity (EC) (dS m^{-1}) [1]	3.79	0.45	2.21	0.81	1.97 / 2.63	2.16 / 3.06	0.77 / 1.36
Sand (%)	NA	11	24	33	NA[3]	NA	NA
Silt (%)	NA	30	36	6.5/6.2	NA	NA	NA
Clay (%)	NA	59	39	6.5/6.2	NA	NA	NA
Soil Texture	NA	Clay	Clay loam	Clay loam	NA	NA	NA
Total C (%)	23.3	2.03	1.11 (0.81)[2]	0.94	83.0	81.8	82.5
Total N (%)	2.26	0.27	0.23	0.16	0.54	0.64	0.70
Total P (g kg^{-1})	10.2	1.16	0.98	0.77	83.0	1.86	0.55
Total K (g kg^{-1})	NA	NA	NA	NA	1.59	2.18	0.63
Total Na (g kg^{-1})	NA	NA	NA	NA	0.39	0.34	0.36
Total Ca (g kg^{-1})	NA	NA	NA	NA	4.99	0.51	7.19
Total Mg (g kg^{-1})	NA	NA	NA	NA	0.48	0.30	0.32
Total Cu (mg kg^{-1})	NA	NA	NA	NA	13.7	2.80	1.36
Total Pb (mg kg^{-1})	NA	NA	NA	NA	0.83	0.83	0.41
Total Zn (mg kg-1)	NA	NA	NA	NA	9.40	27.2	1.73
Exchangeable K (cmol(+) kg^{-1} soil)	6.43	0.32	0.29	0.21	7.22	6.02	1.91
Exchangeable Na (cmol(+) kg^{-1} soil)	1.09	0.31	0.26	037	1.40	1.17	1.26
Exchangeable Ca (cmol(+) kg^{-1} soil)	2.70	4.85	2.94	2.24	2.16	0.30	3.62
Exchangeable Mg (cmol(+) kg^{-1} soil)	2.72	0.64	0.80	0.36	0.68	0.21	0.40
Cation exchangeable Capacity (CEC) (cmol(+) kg^{-1} soil)	19.7	8.58	11.5	14.2	8.46	15.5	5.20
Base saturation (BS) (%)	69	71	37	22	100	54.0	100
Mehlich 3-P (mg kg^{-1})	6874	163	236	94.0	872	487	96.6
Mehlich 3-K (mg kg^{-1})	8911	68.4	108	94.1	2389	2886	616
Mechlich 3-Ca (g kg^{-1})	14.5	2.03	8.22	2.99	3.65	0.465	4.09
Mehlich 3-Mg (mg kg^{-1})	3972	143	344	401	422	218	278
Mehlich 3-Fe (mg kg^{-1})	396	524	589	1199	88.4	40.6	65.5
Mehlich 3-Mn (mg kg^{-1})	188	29.0	213	185	42.9	44.4	21.9
Mehlich 3-Cu (mg kg^{-1})	6.22	9.77	9.95	3.17	7.92	0.53	0.02
Mehclich 3-Pb (mg kg^{-1})	1.23	10.8	11.7	1.54	ND[3]	0.04	ND[3]
Mehlich 3-Zn (mg kg^{-1})	62.4	20.4	7.98	5.28	6.24	19.9	0.35
Elemental analysis							
C%	NA	NA	NA	NA	83.0	81.8	82.5
H%	NA	NA	NA	NA	1.49	1.60	1.50
N%	NA	NA	NA	NA	0.54	0.64	0.70
O%	NA	NA	NA	NA	154	13.3	12.0
C/N ratio	NA	NA	NA	NA	12.6	128	118
(O +N)/C atomic ratio	NA	NA	NA	NA	0.114	0.129	0.016
O/C atomic ratio	NA	NA	NA	NA	0.108	0.122	0.109
H/C atomic ratio	NA	NA	NA	NA	0.215	0.234	0.219

[1] The pH and electrical conductivity (EC) of biochar and compost were measured using 1:5 solid: solution ratio after shaking for 30 min in deionized water; Soil pH was determined in soil-to-deionized water ratio of 1:1 (g mL^{-1}) and in soil-to-1N KCl ratio of 1:1 (g mL^{-1}); Biochar EC was also measured after shaking biochar-water mixtures (1:5 solid: solution ratio) for 24 h; [2] carbonate content; [3] NA = not analyzed. (Data of compost and three soils were abstracted from Tsai and Chang (2019); Data of three biochars were abstracted from Tsai and Chang (2020a)).

Table 1. Characteristics of compost, three studied soils, and three studied biochars.

The studied biochars, including ash biochar (A) [formosan ash (*Fraxinus formosana*)], bamboo biochar (B) [makino bamboo (*Phyllostachys makino*)], and lead tree biochar (L) [lead tree (*Leucaena leucocephala* (Lam.) de. Wit)], were pyrolyzed with the stems and branches of three feedstocks in an earth kiln which was produced by the Forest Utilization Division, Taiwan Forestry Research Institute, Taipei, Taiwan. As indicated in the previous study (Tsai and Chang, 2020a), the lead tree, or white popinac, is an exotic plant; however, because its economic value has decreased over time, this plant has been left to proliferate by itself and has a strong ability to compete with other plants, and many lead trees were cut down by local governments. Bamboo grows quickly, especially makino bamboo in Taiwan, has a high strength-to-weight ratio, and as a kind of wood, is mainly composed of hemicelluloses, cellulose, and lignin, which can produce higher value-added products via pyrolysis processes. The charring for earth kilns typically requires several days and reaches temperatures up to 500 - 700 °C. The highest temperature in the kiln at the end of carbonization was above 750 °C. The biochars were homogenized and ground into a mesh of <2 mm for analysis. The particle size distribution of the studied biochar was as follows: >1 mm, 2%; 1 - 0.5 mm, 12%; < 0.5 - 0.25 mm, 13%; < 0.25 - 0.105 mm, 54%; and <0.105 mm, 19% for ash biochar; >1 mm, 17%; 1–0.5 mm, 34%; <0.5 - 0.25 mm, 16%; <0.25 - 0.105 mm, 20%; and <0.105 mm, 12% for bamboo biochar; >1 mm, 8%; 1 - 0.5 mm, 37%; <0.5 - 0.25 mm, 22%; <0.25 - 0.105 mm, 18%; and <0.105 mm, 15% for lead tree biochar. The characteristics of studied biochars, including pH, EC, CEC, elemental analysis, Fourier transform infrared spectra (FTIR), X-ray diffraction (XRD) patterns, and scanning electron microscopy (SEM) observations, were analyzed and described in previous studies (Tsai and Chang, 2020a). Compared within three biochars (*Table 1*), the A biochar has the highest content of exchangeable K, Na, and Mg, and M3-P, Mg, Fe, Mn, and Cu; the B biochar has the highest content of pH, CEC, and M3-K, Mn, and Zn, but has the lowest content of Ex. Na, Ca, and Mg, M3-Ca, Mg, and Fe; the L biochar has the highest content of Ex. Ca and M3-Ca, but has the lowest content of pH, CEC, Ex. K, M3-P, -K, Mn, Cu, and Zn. Additionally, the three high-temperature-pyrolysis biochars have higher aromatic structure contents, fewer polar functional groups, and lower labile C (water-extractable organic C) (Tsai and Chang, 2020a, 2021), as evidenced by the low CEC of the three biochars.

Soil-Biochar incubation test and data analysis

The incubation experiment has described in the previous study (Tsai and Chang, 2021). In short, the incubation experiment for each soil comprised seven treatments with five replicates (n = 105), including (1) natural soil + 5% compost (Control); (2) soil + 5% compost + 2% A biochar (A2); (3) soil + 5% compost + 5% A biochar (A5); (4) soil + 5% compost + 2% B biochar (B2); (5) soil + 5% compost + 5% B biochar (B5); (6) soil + 5% compost + 2% L biochar (L2); and (7) soil + 5% compost + 5% L biochar (L5). Then 5 wt % compost was to test excessive application, like previous studies (Tsai and Chang, 2019, 2020a, 2020b, 2020c, 2021). We must confess that application of these high rates (5%, by wt.) of biochar may be problematic in practices, e.g., difficult for operational incorporation in the field. Ten sets of incubation vessels were prepared for ten sampling times. Twenty-five grams of mixed soil sample was placed in 30-mL plastic containers, which were subsequently put into 500 mL plastic jars. The jars were sealed and incubated at 25 °C. Soil moisture content was adjusted to 60% of field capacity before the incubation and was maintained throughout the experiment using repeated weighing. Destructive sampling of soil samples was conducted at 1, 3, 7, 28, 56, 84, 140, 196, 294, and 400 days for measurements of soil Mehlich-3 (M3) extractive nutrients (P, K, Ca, Mg, Fe, Mn, Cu, Pb, and Zn). The analysis methods were as described in the previous study (Tsai and Chang, 2019, 2020b, 2020c).

Statistical analyses (the calculation of means and standard deviations, and differences of means) were performed using the Statistical Analysis System (SAS) 9.4 package (SAS Institute Inc., SAS Campus Drive, Cary, NC, USA). Arithmetic means of the available nutrients were calculated from the results at each consecutive measurement date. A repeated measure multivariate analysis of variance (MANOVA) was used to test the effects of biochar feedstocks, soils, addition rates, and their interactions on available nutrients in each incubation period. The feedstocks, addition rates and soils served as between-subject factors, and incubation time served as the within-subject factor. The repeated measure MANOVA was carried out using the general linear model (GLM) procedure. Three-way analysis of variance (ANOVA) was used to examine differences in the selected soil parameters of biochar-amended soil incubation test among soil, biochar and rate. The results were analyzed by analysis of variance (one-way ANOVA) to test the effects of each treatment. Significantly different means were compared via least significant difference (LSD), based on a t-test with a 5% probability level. The values presented in the graphs and the text are means ± 1 standard deviation (SD). The Pearson correlation coefficient (*r*) was calculated using SAS 9.4 software, including the mean value of soil pH and C/N (abstracted from Tsai and Chang (2021)). According to previous studies (Tsai and Chang, 2021), three-way ANOVA was analyzed in the first and 57th weeks due to the obvious changes in the first week. The sum content of available nutrients at the first week include day 1, 3, and 7 destructive soil samples analysis, and at the 57th weeks include 10 times' destructive soil samples analysis.

Changes of available P, K, Ca, and Mg

In the first week, available P showed an insignificant decrease for all biochar treatments compared to the control in the SAO soil, on the contrary, the MAI and SAI soil mostly showed insignificant increases (*Figure 1a*, *Table 2*). After 57th weeks incubation, the A5 treatment in the SAO soil showed a significant increase (+5%) and the L5 treatment showed a significant decrease (-6%). In the MAI and SAI soil, the sum content of M3-P mostly declined, and a significant decrease (-5%) has occurred in the L treatment (L2 and L5). The change of available K in three soils compared with the control all showed significantly increasing trends with increasing biochar rate at the first week and 57th weeks (*Figure 1b*, *Table 2*). The Ca content at the first week was an insignificant difference between treatments insignificant difference in the SAO and SAI soil, but a significant decrease in L2 (-5%) and L5 (-6%) treatments of the MAI soil (*Figure 1c*, *Table 2*). In the 57th weeks, the B5 treatment of the SAO soil and the MAI soil showed a significant decrease and no significant difference between treatments in the SAI soil. The highest Ca content in the control and biochar-amended MAI soil would be due to the highest Ca content of the raw MAI soil. However, the L biochar with higher Ca content only showed an insignificant increase in the MAI and SAI soil, about a 2% increase compared to the control. The relative percentage of the sum of P, K, and Ca in the first week compared to the final sum at 57th weeks was between 29% to 32% for three soils, with a relatively higher percentage for P and similar between K and Ca.

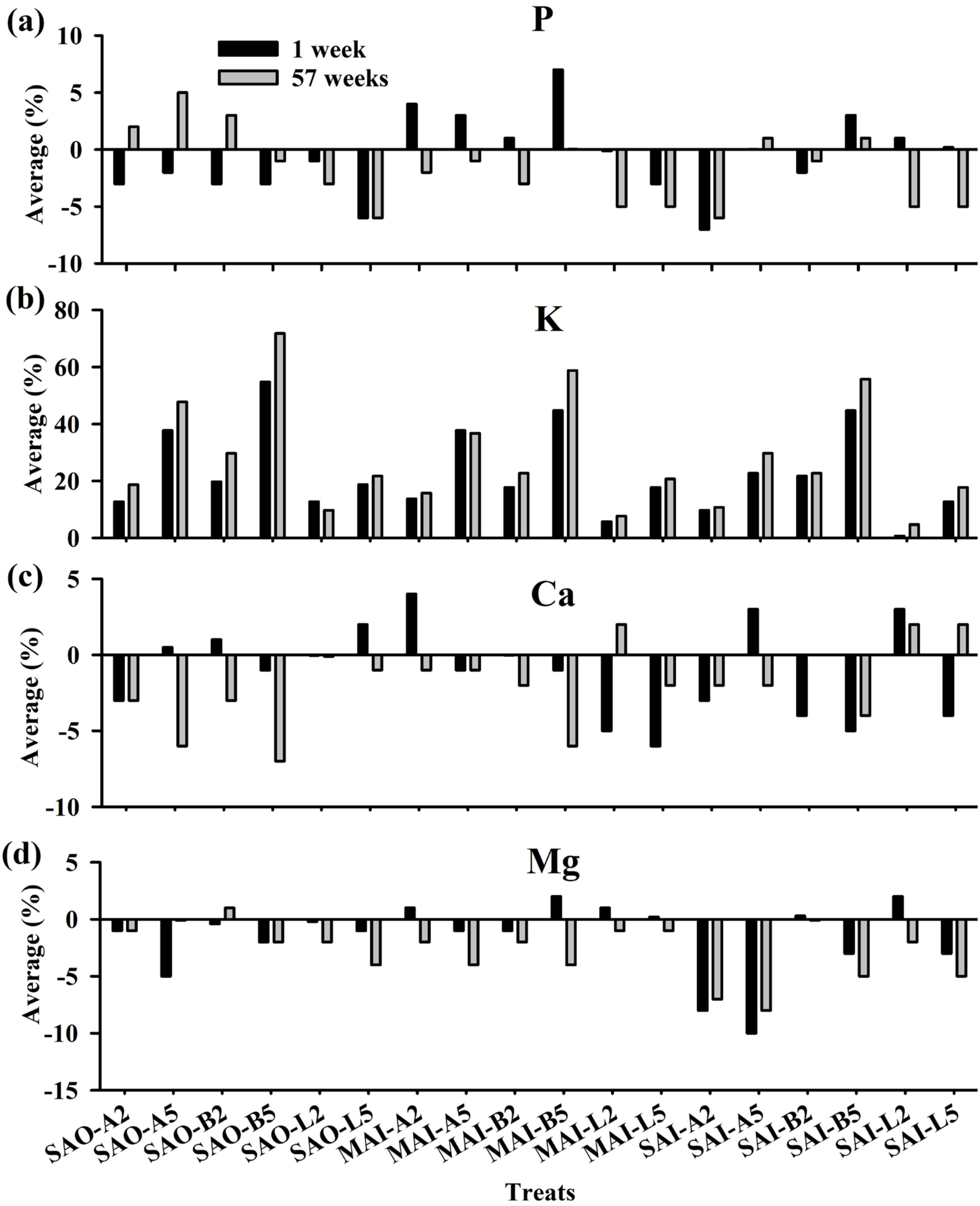

Figure 1. Average percentage (%) of sum relative value of Mehlich 3-extractive (a) P, (b) K, (c) Ca, and (d) Mg in three studied soils at the first week and the 57th weeks. The first week value is the sum of destructive soil samples analysis at day 1, 3 and 7, and the 57th weeks value is the sum of totally ten times' destructive soil samples analysis. Percentage expressed as the difference value (1 week and 57th weeks, respectively) between biochar amended treatments and un-amended control treatment for each soil. The data are mean value (n = 5).

Soil	Treats[1]	P		K		Ca	
		1 week (g kg^{-1})[2]	57th weeks (g kg^{-1})[2]	1 week (g kg^{-1})[2]	57th weeks (g kg^{-1})[2]	1 week (g kg^{-1})[2]	57th weeks (g kg^{-1})[2]
SAO	Control	1.27±0.07 c[3]	3.68±0.13 ef	0.97±0.04 j	3.10±0.06 m	6.84±0.25 hij	25.0±0.66 defg
	A2	1.24±0.08 c	3.75±0.05 de	1.10±0.07 i	3.71±0.07 k	6.65±0.52 j	24.1±0.62 fgh
	A5	1.24±0.04 c	3.88±0.13 d	1.34±0.04 de	4.61±0.07 f	6.87±0.14 hij	23.6±0.94 gh
	B2	1.23±0.07 c	3.78±0.13 de	1.17±0.04 gh	4.02±0.07 i	6.93±0.17 ghij	24.1±0.41 fgh
	B5	1.23±0.04 c	3.66±0.09 ef	1.50±0.08 b	5.35±0.08 c	6.80±0.16 ij	23.2±0.54 h
	L2	1.25±0.09 c	3.58±0.16 fg	1.10±0.05 i	3.40±0.010 i	6.83±0.31 hij	25.0±1.34 defg
	L5	1.19±0.08 c	3.46±0.10 g	1.16±0.04 ghi	3.79±0.08 jk	6.98±0.22 ghij	24.7±0.38 defg
MAI	Control	1.55±0.08 b	5.11±0.14 a	1.11±0.07 hi	3.86±0.10 j	12.3±0.51 ab	41.9±1.62 ab
	A2	1.61±0.04 ab	5.03±0.09 ab	1.27±0.02 f	4.45±0.04 g	12.8±0.51 a	41.3±0.65 b
	A5	1.59±0.06 ab	5.04±0.15 a	1.53±0.07 b	5.30±0.10 c	12.2±0.42 b	41.3±0.72 b
	B2	1.56±0.08 b	4.95±0.09 abc	1.31±0.09 ef	4.75±0.08 e	12.3±0.35 ab	41.2±0.65 b
	B5	1.66±0.14 a	5.11±0.26 a	1.61±0.06 a	6.14±0.19 a	12.2±0.30 b	39.3±0.30 c
	L2	1.55 ±0.09 b	4.84±0.13 c	1.18±0.02 g	4.17±0.04 h	11.7±0.21 c	42.9±1.97 a
	L5	1.51±0.10 b	4.86±0.10 bc	1.30±0.06 ef	4.66±0.07 ef	11.5±0.55 c	41.2±2.56 b
SAI	Control	1.02±0.09 d	3.43±0.16 g	1.14±0.05 ghi	3.82±0.06 j	7.63±0.30 def	25.3±0.65 def
	A2	0.95±0.05 d	3.21±0.07 i	1.25±0.04 f	4.24±0.08 h	7.39±0.62 efg	24.7±0.90 defg
	A5	1.02±0.04 d	3.47±0.16 g	1.41±0.03 c	4.99±0.14 d	7.87±0.26 d	24.9±1.17 defg
	B2	1.00±0.44 d	3.40±0.07 gh	1.39±0.06 cd	4.72±0.07 e	7.33±0.26 fgh	25.4±0.55 def
	B5	1.05±0.06 d	3.46±0.09 g	1.66±0.04 a	5.97±0.13 b	7.25±0.36 fghi	24.3±0.42 efgh
	L2	1.02±0.08 d	3.24±0.09 hi	1.16±0.03 ghi	4.03±0.07 i	7.84±0.21 de	25.8±0.81 de
	L5	1.02±0.06 d	3.25±0.13 hi	1.29±0.02 ef	4.50±0.07 g	7.29±0.29 fghi	25.9±1.18 d

[1]: **A** = ash biochar, **B** = bamboo biochar, **L** = lead tree biochar, 2 = 2% biochar addition, 5 = 5% biochar addition; [2]: The first week value is the sum of destructive soil samples analysis at day 1, 3 and 7, and the 57th weeks value is the sum of totally ten times' destructive soil sample analysis; [3]:Mean± standard deviation (SD); Means (n =5) compared within a column followed by a different lowercase letter are significantly different at $p < 0.05$ using a one-way ANOVA.

Table 2. Significant test of Mehlich-3 extractable P, K, and Ca at the 1 week and the 57th weeks.

In the SAO and MAI soil, the Mg content at the first week showed insignificant differences between treatments, but in the SAI soil, the A2 and A5 treatments were significantly lower than the control (*Figure 1d*, *Table 3*). At the 57th weeks, the Mg content also did not show significant reduction with biochar rate increasing in the SAO and MAI soil, but the L5 treatment of the SAO soil and the A5 and B5 treatment of the MAI soil were significantly lower than the control, reducing 4%, respectively. In the SAI soil, the Mg content showed significant reduction with biochar rate increasing, especially for the A2 and A5 treatment significantly lower than the control, reducing 7% and 8%, respectively. However, the A biochar containing the highest available Mg content did not significantly increase the Mg content of three biochar-amended soils at the first week and after incubation.

Changes of available Fe, Mn, Cu, Pb, and Zn

No significant difference (increase or decrease) of Fe and Mn content between treatments could be found at the first week and the 57th weeks in the SAO and MAI soil (*Figure 2a, 2b; Table 3*). However, the significant decline of Fe content in the SAI soil has occurred in the B5 treatment at the first week and in the A2, A5, and B5 treatment at the 57th weeks. The Mn content also showed a significant decrease in the B5 and L2 treatment at the first week and the B5 treatment at the 57th weeks. The addition of A biochar containing the highest Fe content has resulted in an insignificant increase of Fe content with an increasing rate in the SAO soil, but a significant decrease in the SAI soil. The addition of B biochar containing the highest Mn content has resulted in inconsistent changes of Mn content in three soils. The Fe and Mn content within three soils were consistent with the Fe and Mn content of raw soils, that is, for the Fe content with the decreasing order SAI > MAI >SAO, and the Mn content with MAI > SAI > SAO. The relative percentage of the sum of Mg, Fe, and Mn in the first week compared to the final sum at 57th weeks was between 27% to 32% for three soils, with a relatively higher percentage for Fe, followed by Mg, and Mn.

Soil	Treats[1]	Mg		Fe		Mn	
		1 week ($g\ kg^{-1}$)[2]	57th weeks ($g\ kg^{-1}$)[2]	1 week ($g\ kg^{-1}$)[2]	57th weeks ($g\ kg^{-1}$)[2]	1 week ($g\ kg^{-1}$)[2]	57th weeks ($g\ kg^{-1}$)[2]
SAO	Control	1.21±0.04 e[3]	4.13±0.06 i	0.80±0.05 fgh	2.48±0.06 efg	98.7±3.7 f	493±24 d
	A2	1.19±0.12 e	4.11±0.11 ij	0.78±0.04 gh	2.53±0.03 def	97.8±3.2 f	503±23 d
	A5	1.15±0.03 e	4.13±0.09 i	0.81±0.05 fgh	2.60±0.09 de	98.4±2.6 f	502±23 d
	B2	1.21±0.03 e	4.17±0.12 i	0.80±0.03 fgh	2.48±0.10 efg	99±3.1 f	519±12 d
	B5	1.18±0.05 e	4.03±0.07 ij	0.78±0.04 h	2.40±0.10 g	98.4±3.6 f	498±5.9 d
	L2	1.21±0.03 e	4.04±0.06 ij	0.79±0.04 fgh	2.44±0.06 fg	96.8±2.8 f	496±13 d
	L5	1.20±0.05 e	3.98±0.06 j	0.79±0.04 fgh	2.44±0.10 fg	96.4±3.6 f	493±19 d
MAI	Control	1.63±0.06 d	5.72±0.12 def	0.83±0.04 efgh	2.60±0.06 de	479±18 ab	1629±33 a
	A2	1.64±0.02 d	5.62±0.11 efgh	0.86±0.04 defg	2.63±0.06 d	489±12 ab	1645±29 a
	A5	1.61±0.07 d	5.48±0.10 h	0.85±0.02 defgh	2.57±0.06 de	476±19 ab	1621±33 a
	B2	1.61±0.08 d	5.60±0.09 fgh	0.86±0.05 defg	2.62±0.10 d	494±25 a	1646±41 a
	B5	1.67±0.15 cd	5.52±0.21 gh	0.91±0.09 cde	2.62±0.15 d	505±34 a	1651±68 a
	L2	1.65±0.05 d	5.69±0.08 def	0.86±0.01 defg	2.64±0.06 d	481±8.0 ab	1652±26 a
	L5	1.63±0.10 d	5.65±0.13 efg	0.86±0.05 def	2.63±0.10 d	498±40 a	1676±74 a
SAI	Control	1.81±0.07 ab	6.26±0.14 a	1.00±0.09 ab	3.31±0.13 a	422±46 d	1291±50 b
	A2	1.66±0.07 cd	5.81±0.10 cd	0.95±0.04 bc	3.16±0.13 c	410±27 de	1275±60 bc
	A5	1.63±0.06 d	5.77±0.20 de	0.94±0.04 bc	3.12±0.14 c	417±48 d	1261±74 bc
	B2	1.81±0.04 ab	6.26±0.14 a	1.01±0.04 ab	3.33±0.06 a	430±35 cd	1225±44 c
	B5	1.75±0.09 bc	5.92±0.17 bc	0.92±0.07 cd	3.16±0.11 bc	379±22 e	1225±44 c
	L2	1.85±0.05 a	6.15±0.08 a	1.04±0.03 a	3.31±0.07 a	458±31 bc	1308±38 b
	L5	1.74±0.05 bc	5.97±0.09 b	1.00±0.06 ab	3.27±0.04 ab	414±44 d	1288±38 b

[1]: **A** = ash biochar, **B** = bamboo biochar, **L** = lead tree biochar, 2 = 2% biochar addition, 5 = 5% biochar addition; [2]: The first week value is the sum of destructive soil samples analysis at day 1, 3 and 7, and the 57th weeks value is the sum of totally ten times' destructive soil samples analysis; [3]: Mean±standard deviation (SD); Means (n =5) compared within a column followed by a different lowercase letter are significantly different at $p < 0.05$ using a one- way ANOVA.

Table 3. Significant test of Mehlich-3 extractable Mg, Fe, and Mn at the 1 week and the 57th weeks.

The addition of the A biochar containing the highest Cu content has resulted in the insignificant increase of soil Cu content with increased biochar rate at the first week for three soils, but the significant increase at the 57th weeks with increased biochar rate (*Figure 2c, Table 4*).

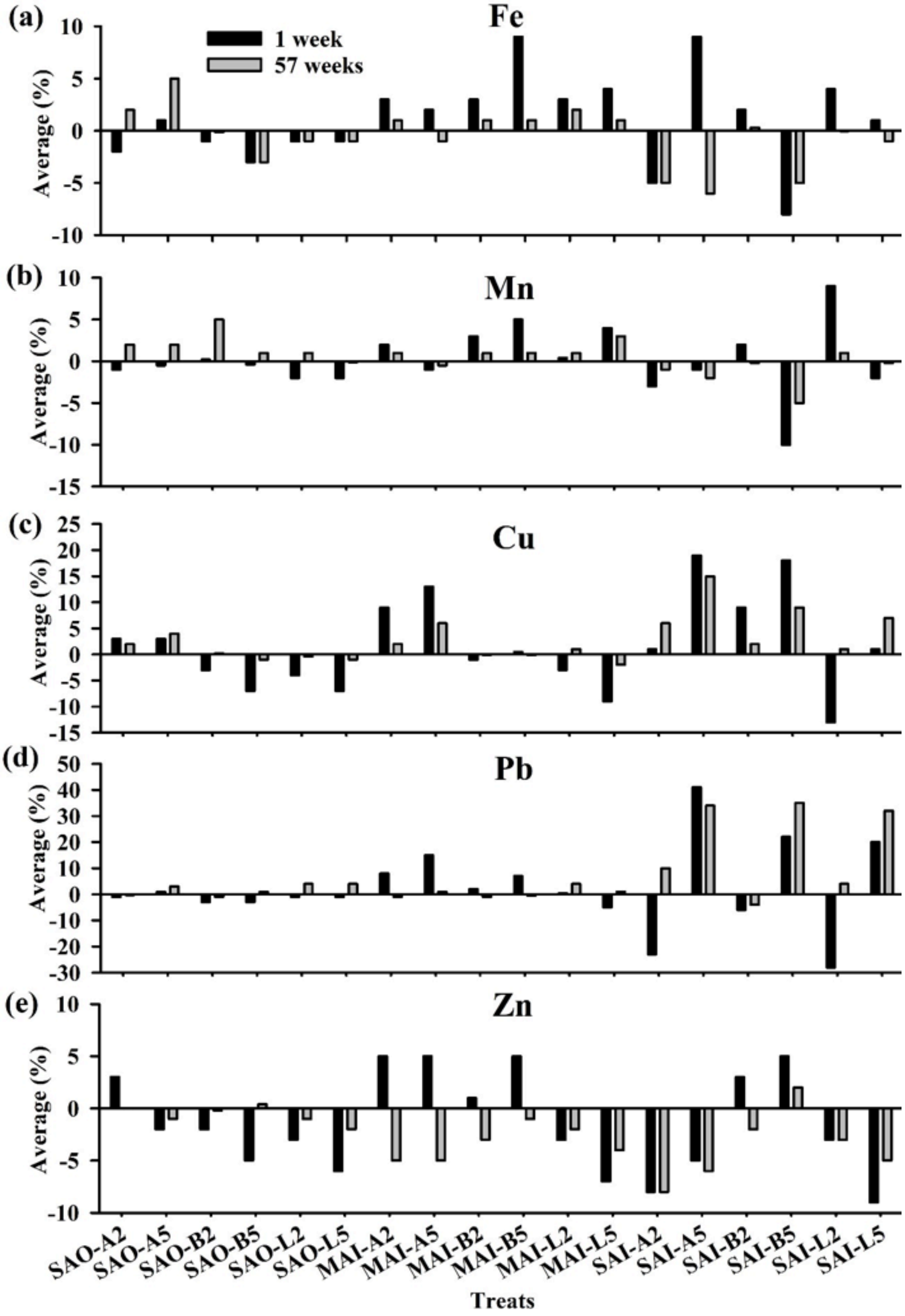

Figure 2. Average percentage (%) of sum relative value of Mehlich 3-extractive (a) Fe, (b) Mn, (c) Cu, (d) Pb, and (e) Zn in three studied soils at the first week and the 57th weeks. The first week value is the sum of destructive soil samples analysis at day 1, 3 and 7, and the 57th weeks value is the sum of totally ten times' destructive soil samples analysis. Percentage expressed as the difference value (1 week and 57th weeks, respectively) between biochar amended treatments and un-amended control treatment for each soil. The data are mean value (n = 5).

The addition of B and L biochar in the SAO indicated the insignificant reduction of the Cu content between 2% and 5% addition rate, but significantly lower than the control. Similar changes could be found in the MAI soil, but only the L5 treatment showed a significant reduction. The mixed-soil Cu content at the first week in the SAI soil was significantly lower than the SAO and MAI soil, about one-fifth, and the Cu content insignificantly increased with increased biochar rate. After the incubation (57th weeks), the A5 and B5 treatment of the SAI soil both showed a significant increment compared to the control and the 2% treatment. The Pb content of the compost and three biochars was lower (1.23 mg kg^{-1} $compost^{-1}$) and very lower (0.04 mg kg^{-1} $biochar^{-1}$~not detected), respectively (*Table 1*). The Pb content of the biochar-amended soil could be expected to have no significant effect by biochar, as evidenced by the mostly insignificant difference between treatments at the first week and 57th weeks in three soils (*Figure 2d, Table 4*). The Zn content in the compost, three soils and three biochars was relatively higher within Cu, Pb, and Zn (*Table 1*). Thus, the sum Zn content (*Table 4*) was higher than Cu and Pb. In the MAI and SAI soil at the first week and the SAO soil at the 57th weeks, there were insignificant differences between treatments (*Figure 2e*). The L biochar treatment showed similar changes in three soils, that is, an insignificant decline with the increase of biochar rate. The changes of A and B biochar treatments were inconsistent at the first week and 57th week in three soils. However, after incubation, the change in Zn content compared to the control almost showed a decrease. The relative percentage of the sum of Cu, Pb, and Zn in the first weeks compared to the final sum at 57th weeks was between 17% to 29% for three soils, with a relatively higher percentage for Zn, followed by Cu, and Pb.

Soil	Treats[1]	Cu		Pb		Zn	
		1 week ($g\ kg^{-1}$)[2]	57th weeks ($g\ kg^{-1}$)[2]	1 week ($g\ kg^{-1}$)[2]	57th weeks ($g\ kg^{-1}$)[2]	1 week ($g\ kg^{-1}$)[2]	57th weeks ($g\ kg^{-1}$)[2]
SAO	Control	20.2±0.88 bcd[3]	69.6±0.95 ef	19.7±0.87 e	78.6±2.43 e	86.4±2.96 b	286±8.1 a
	A2	20.8±0.58 bc	71.1±0.72 cd	19.5±0.89 e	78.4±0.74 e	89.6±5.55 a	286±8.0 a
	A5	20.9±0.56 b	72.5±0.73 b	20.0±0.63 e	80.7±1.47 cde	84.8±1.94 bc	284±2.9 a
	B2	19.6±0.59 cde	69.8±0.91 def	19.2±1.20 e	77.9±1.55 e	84.8±2.40 bc	286±2.7 a
	B5	18.8±0.93 e	69.1±1.26 f	19.1±1.10 e	79.1±2.55 de	81.8±4.33 c	287±6.8 a
	L2	19.4±0.26 de	69.3±0.51 f	19.6±0.56 e	82.1±1.84 c	83.4±2.71 bcj	284±5.4 a
	L5	18.8±0.61 e	69.0±0.65 f	19.6±1.15 e	81.6±0.58 cd	81.4±3.64 c	280±3.1 a
MAI	Control	20.8±1.36 bc	70.9±1.88 cde	21.5±1.59 d	91.1±4.37 b	45.4±1.82 def	176±8.0 b
	A2	22.7±0.31 a	72.6±0.61 b	23.3±0.23 b	89.9±0.78 b	47.8±1.25 d	167±2.0 c
	A5	23.6±0.22 a	74.8±0.43 a	24.7±0.54 a	92.0±0.58 b	47.8±1.49 d	166±4.2 c
	B2	20.6±1.32 bcd	70.8±0.82 cde	21.9±1.82 cd	90.7±1.16 b	46.0±1.87 de	170±1.6 bc
	B5	20.9±0.37 b	70.8±1.35 cde	22.9±0.66 bc	90.7±2.26 b	47.2±1.29 de	174±2.9 bc
	L2	20.2±0.42 bcd	71.5±0.86 bc	21.6±0.90 d	94.9±1.10 a	44.2±1.95 ef	173±3.3 bc
	L5	18.9±2.89 e	69.2±2.68 f	20.5±3.15 de	92.4±3.49 b	42.4±3.78 f	169±3.1 bc
SAI	Control	4.16±0.64 fg	22.4±0.84 i	0.19±0.07 f	7.34±0.98 f	35.9±2.32 ghi	119±6.2 de
	A2	4.20±0.59 fg	23.7±0.60 hi	0.18±0.05 f	8.12±0.55 f	32.8±1.71 i	110±5.4 f
	A5	4.98±0.69 f	25.8±0.90 g	0.28±0.08 f	9.88±0.80 f	33.8±1.11 hi	112±4.7 f
	B2	4.54±0.49 fg	22.9±0.51 i	0.20±0.06 f	7.04±0.47 f	36.8±1.51 gh	116±2.5 def
	B5	4.90±0.38 f	24.5±0.43 gh	0.24±0.04 f	9.88±0.77 f	37.6±1.33 g	122±3.1 d
	L2	3.62±0.40 g	22.5±0.33 i	0.16±0.04 f	7.64±0.63 f	34.8±1.61 ghi	116±2.2 def
	L5	4.18±0.76 fg	23.9±1.16 hi	0.24±0.09 f	9.68±0.86 f	32.8±1.35 i	113±4.5 ef

[1]: **A** = ash biochar, **B** = bamboo biochar, **L** = lead tree biochar, 2 = 2% biochar addition, 5 = 5% biochar addition; [2]: The first week value is the sum of destructive soil samples analysis at day 1, 3 and 7, and the 57th weeks value is the sum of totally ten times' destructive soil samples analysis; [3]: : Mean±
standard deviation (SD); Means (n =5) compared within a column followed by a different lowercase letter are significantly different at $p < 0.05$ using a one- way ANOVA.

Table 4. Significant test of Mehlich-3 extractable Cu, Pb, and Zn at the 1 week and the 57th weeks.

Biochar effects on available P retention and availability

The available P content was the only significant correlation ($p < 0.0001$) with biochar factor and incubation time, and almost all interactions of time, soil, biochar, and rate (*Table 5*). At the first week of biochar application on excessive compost-fertilized soil, soil condition plays an important role, and additionally, with time different biochar feedstocks and rates would gradually alter the soil conditions but the change magnitude is soil specific and smaller (*Table 6*).

Source of variation	df[1]	P	K	Ca	Mg	Fe	Mn	Cu	Pb	Zn
Between subject effect										
Soil	2	ns[2]	ns	***[3]	***	***	***	***	***	***
Biochar	2	***	***	***	***	ns	ns	***	***	***
Rate	2	ns	ns	***	***	ns	ns	***	***	***
Soil×Biochar	4	ns	ns	*	***	ns	ns	ns	***	ns
Biochar×Rate	4	ns	ns	ns	ns	ns	ns	***	***	***
Soil×Rate	4	ns	ns	ns	***	ns	ns	*	ns	ns
Within subject effect										
Time	9	***	***	***	***	***	***	***	***	***
Time×Soil	18	***	***	***	***	***	***	***	***	***
Time×Biochar	18	***	***	***	***	***	***	***	***	***
Time×Rate	18	**	**	ns	ns	ns	ns	ns	ns	ns
Time×Soil×Biochar	36	***	***	***	***	***	***	***	***	***
Time×Soil×Rate	36	ns	**	***	*	ns	ns	ns	***	***
Time×Biochar×Rate	36	***	***	*	**	ns	*	ns	***	ns

[1]: df = degree of freedom; [2]: ns = not significant; [3]: * significant at $p < 0.01$, ** $p < 0.001$, *** $p < 0.0001$.

Table 5. Significance (P value) of repeated-measures MANOVA results on selected soil parameters in different woody biochar (Biochar), soil type (Soil) and biochar application rates (Rate) after 400 days of soil incubation.

Source		Soil	Biochar	Rate	Soil x Biochar	Biochar x Rate	Soil x Rate	Soil x Biochar x Rate
df		2	2	2	4	4	4	8
P	1 w	***	ns	ns	ns	ns	ns	ns
	57th w	***	***	*	ns	**	ns	ns
K	1 w	***	***	***	**	***	ns	*
	57th w	***	***	***	ns	***	**	*
Ca	1 w	***	***	ns	***	ns	ns	ns
	57th w	***	***	***	ns	ns	ns	ns
Mg	1 w	***	***	*	*	ns	*	ns
	57th w	***	***	***	***	ns	***	*
Fe	1 w	***	*	ns	*	ns	*	ns
	57th w	***	ns	ns	**	ns	ns	*
Mn	1 w	***	ns	ns	*	ns	ns	ns
	57th w	***	ns	ns	ns	ns	ns	ns
Cu	1 w	***	***	ns	*	*	ns	**
	57th w	***	***	***	nc	***	*	**
Pb	1 w	***	ns	ns	ns	ns	ns	*
	57th w	***	***	***	***	***	ns	*
Zn	1 w	***	**	ns	***	ns	ns	ns
	57th w	***	***	***	ns	***	ns	ns

[1]:df = degree of freedom; 1 w = 1 week; 57th w = 57 weeks; ns = not significant ($p > 0.01$); * $p < 0.01$; ** $p < 0.001$; *** $p < 0.0001$.

Table 6. Summary of three-way ANOVA results (p value) on soil Mehlich-3 extractable nutrients (P, K, Ca, Mg, Fe, Al, Mn, Cu, Pb and Zn) in three soils (Soil), three biochars (Biochar) and three biochar application rates (Rate) at the first week (1 w) and the 57 weeks (57th w) of soil incubation[1].

Soil P availability was affected after biochar addition due to soil pH change (Murphy and Stevens, 2010), P sorption capacity change (Laird et al. 2010), and the available P content of the soil (with or without application of P fertilizer) and the initial sorption capacity of the soil and the controlling sorption mechanism (by Fe, Al, clay content vs. by $CaCO_3$) (Bornø et al. 2018). After biochar addition, the amount of P precipitated with Al and Fe decline after increasing pH in acidic soils (Parfitt, 1978; Atkinson et al. 2010, Bornø et al. 2018), due to the increase of active alkaline metal (Ca^{2+}, Mg^{2+} and K^{+}) oxides resulting in the decrease of active Al^{3+} solubility (DeLuca et al. 2015). In neutral or alkaline soils, the reduction of P availability could be attributed to the added metals enhanced P fixation (DeLuca et al. 2015). This study results showed that the soil pH has no significant correlation with available P content in three soils (*Table 7*), which could be attributed to the smaller magnitude of soil pH changes, increasing 0.01-0.26 pH unit at the end of incubation (Tsai and Chang, 2021). According to Yang et al. (2016), biochar with high pH and CEC being more effective in increasing soil P availability, the relatively high pH and CEC of B biochar in this study could have the potential in increasing soil P availability but not evidenced in *Table 2*. Compared to the B biochar, the A biochar has similar high pH, lower CEC but higher available P content, suggesting that the A biochar was more effective in increasing soil P availability, as clearly evidenced in *Table 2*. Besides, the application of 2-8 % maize straw biochar significantly increased both Olsen-P and soil microbial biomass (SMB)-P in Red earth and Fluvo-aquic soil, with greater effect by increasing application rates (Zhai et al. 2015); in addition, the increase of soil Olsen-P and SMB-P were less in acidic Red earth soil than alkaline Fluvo-aquic soil under the same maize straw biochar application, this is probably because less P is available in Red earth for microbial organisms due to a stronger sorption capacity. On the contrary with the study of Zhai et al. (2015), the increase of available P in acidic SAO soil (Red earth soil) in this study was not less than alkaline MAI soil (Fluvo-aquic soil) and acidic SAI soil (Fluvo-aquic soil), which could be attributed to a large amount of inorganic P added from excessive compost. It needs to consider both soil and biochar properties for agricultural use of biochar in regards to P availability, because of the contradictory effects of the biochars on P dynamics in different soils (Bornø et al. 2018).

The P content of the control and biochar treatments in SAO soil showed significantly positive correlations with Mg, Fe, Mn, Cu. and Zn, and it was Mg, Fe and Zn in MAI soil (*Table 7*). No significant correlation occurred between P and other variables in the SAI soil. In this study, Mg and Fe seem more important for P availability. P ions could precipitate with free ions in the solution released from the biochar such as Ca^{2+}, Mg^{2+}, or co-precipitate with mixed mineral complexes (Al-Si-Fe-Ca) on biochar surfaces (Shepherd et al. 2017). After application at the high biochar application rate (20 g biochar kg^{-1}) without N or P additions, the decrease in available P in 10 days was attributed to the result of P sorption to biochar surfaces (Nelson et al. 2011). Chintala et al. (2014) proposed that the incorporation of biochars to acidic soil at 40 g kg^{-1} increased the equilibrium solution P concentration (reduced the sorption) and increased available sorbed P; in calcareous soil, application of alkaline biochars significantly increased the sorption of P and decreased the availability of sorbed P. In this study, the decrease in available P at the first week and 57th weeks after application at the 2% and 5% biochar application rate could be a result of P sorption to biochar surfaces, especially obvious in the L biochar treatments. Similar effects of biochar application in acidic Inceptisols (Xu et al. 2014) and in acidic Luvisols and Lixisols (Bornø et al. 2018), where a low P loading increased P availability, and a high P loading decreased P availability compared to the control soil.

Soil	Variables	pH	C/N	P	K	Ca	Mg	Fe	Mn	Cu	Pb	Zn
SAO	pH	1.00	0.67[1]/0.58[2]	ns[3]	ns	ns	ns	ns	ns	ns	ns/0.64	ns
	C/N		1.00	ns	0.70/0.86	ns	ns	ns	ns	ns	ns	ns
	P			1.00	ns	ns	0.63/0.70	0.69/0.66	0.79/ns	ns/0.63	ns	0.59/ns
	K				1.00	ns/-0.67	ns	ns	ns	ns	ns	ns
	Ca					1.00	ns	ns	ns	ns	ns	ns
	Mg						1.00	ns	ns	ns	ns	ns
	Fe							1.00	0.86/ns	ns/0.72	ns	0.55/ns
	Mn								1.00	ns	ns	0.66/ns
	Cu									1.00	0.66/ns	0.75/ns
	Pb										1.00	ns

Soil	Variables	pH	C/N	P	K	Ca	Mg	Fe	Mn	Cu	Pb	Zn
MAI	pH	1.00	0.66/ns	ns[3]	0.69/ns	ns	ns	ns	ns	ns	ns	ns
	C/N		1.00	ns	0.75/0.85	ns	0.76/-0.55	ns	ns	ns	ns	ns
	P			1.00	ns	ns	0.76/ns	0.66/ns	ns	ns	ns	0.71/ns
	K				1.00	ns	ns	ns	ns	ns	ns	ns
	Ca					1.00	ns	ns	ns	ns	ns	ns
	Mg						1.00	0.60/0.59	ns	ns	ns	ns
	Fe							1.00	0.85/0.89	ns	ns	ns
	Mn								1.00	ns	ns	ns
	Cu									1.00	0.93/ns	0.70/ns
	Pb										1.00	0.73/ns
	Zn											1.00

Soil	Variables	pH	C/N	P	K	Ca	Mg	Fe	Mn	Cu	Pb	Zn
SAI	pH	1.00	0.81/0.70	ns	ns	ns	-0.67/ns	ns	ns	ns	ns	ns
	C/N		1.00	ns	0.75/0.86	ns	ns	ns	ns	ns	ns	ns
	P			1.00	ns	ns	ns	ns	ns	ns	ns	ns
	K				1.00	ns	ns	ns	ns	ns/0.60	ns/0.60	ns
	Ca					1.00	ns	ns	ns	ns	ns	ns
	Mg						1.00	0.57/0.75	ns	ns/-0.70	ns/-0.59	ns
	Fe							1.00	0.81/0.73	-0.68/-0.66	ns	ns
	Mn								1.00	-0.83/ns	ns	ns
	Cu									1.00	0.68/0.83	ns
	Pb										1.00	ns
	Zn											1.00

[1]: Pearson correlation coefficients between measured parameters the 1 week; [2] Pearson correlation coefficients between measured parameters the 57 weeks; [3] ns = no significance at the first week and at the 57 weeks; numbers in the table indicate significant correlations by SAS analysis ($p < 0.0001$).

Table 7. Pearson correlation coefficients between measured parameters at the 1 week and the 57th weeks within treatments of three soils.

And further, high pH, relatively high content of elements such as Ca, Mg, Fe and Al and the presence of carbonates, especially calcite, could explain the high P sorption capacity of wood biochar (Bornø et al. 2018). The X-ray diffraction (XRD) patterns of L biochar suggested that L biochar contains more calcite relative to A biochar but non for B biochar (Tsai and Chang (2020a), see *Figure S1*), indicating the high P adsorption of L biochar. The results could explain the significant P content reduction of L biochar treatments (L2 and L5) in three soils. Compared with the control, the content of P in the soil MAI and SAI in the first week increased temporarily, while the content of P at the 57th weeks in the SAO soil increased slightly, indicating that the biochar may have the potential to inhibit phosphorus adsorption or precipitation reactions after excessive compost additions. Some temporary increase in P availability in the biochar and P-amended soil, Nelson et al. (2011) demonstrated that low-molecular-weight organic acids have been shown to decrease P adsorption and increase P availability through the complexation of cations, such as Ca, Al, and Fe; therefore, biochar could have adsorbed cations, as evidenced by the decrease of cations (like Ca and Mg) concentration, resulting in delayed P adsorption or precipitation in the biochar-amended soil. This theory also can be supported in the current study, as evidenced by the decreased Ca and Mg concentrations in the biochar-amended soil (*Table 2, Table 3*). Besides, at the end of the C mineralization incubation test, the available Al content in three soils mostly showed an insignificant decrease with biochar rate increasing, except for the A5 treatment (Tsai and Chang, 2020a); as compared to the control, the significant decline of Al content in SAI and MAI soil suggested the increase of available P, but insignificant.

Biochar effects on available K, Ca, and Mg retention and availability

Similar to available P, available K content showed only a significant correlation ($p < 0.0001$) with biochar factor and incubation time, and all interactions of time, soil, biochar, and rate (*Table 5*). Including soil, biochar, and rate are all important factors and significant correlations with available K content ($p < 0.0001$) at the first week as well as at the 57th weeks (*Table 6*). In three biochar-amended soils, the K content all showed significantly positive correlations with C/N at the 1 week and the 57th weeks (*Table 7*). In addition, the K content showed a significantly negative correlation with Ca at the 57th weeks in SAO soil, no significant correlations in MAI soil, and significantly positive correlations with Cu and Pb at the 57th weeks in SAI soil. The significantly negative correlations between available K and Ca at the 57th weeks in SAO soil suggested that considerable soil Ca was adsorbed by biochar due to the exchange of Ca from soil induced by the large amount of K released to the soil from biochar (Limwikran et al. 2018). Several physicochemical factors that can influence the availability of biochar-derived K nutrient to plants, including solubility of K compounds into water and/or high ionic strength solution, the extent and rate of dissolution of these compounds from biochar to soils, and properties of the soils such as texture, exchange capacity, pH, and water content Jindo et al. (2020). The available K content in three biochar-amended soils as compared to the control shows a significant increase by 10~72% in SAO soil, 8~59% in MAI soil, and 5~56% in SAI soil, especially for K-rich B and A biochar and at the higher biochar rate addition (*Table 1*). Biochar application could result in increased soil K content and plant K tissue concentration (Biederman and Stanley, 2013). In addition, increased concentration of K in biochar, together with Mg and Ca, functions as a liming agent to neutralize acid soils (Jindo et al. 2020). The previous study results (Tsai and Chang, 2021) indicated that soil pH significantly increased with the increasing biochar rate only in the SAO soil, but mostly showed insignificant increases in the MAI and SAI soils; and, compared with the control, there was a greater pH increase in the first week, but the pH gradually declined at the end of incubation test, indicating a reduction in the liming effect of biochar. Besides, the A5 treatment in the SAO soil, and the L5 treatment in the three soils, showed obvious pH increases (3% to 4%) at the end of the incubation test (Tsai and Chang, 2021). The decreased Ca and Mg concentrations (*Table 2, Table 3*) can support the reduction in the liming effect of biochar at the end of the incubation test. As shown in *Table 1*, A biochar contains a high concentration of K, Ca, and Mg, and L biochar contain the highest Ca concentration, indicating that even in the condition of excessive compost the A biochar and L biochar have the higher pH neutralization capacity in biochar-amended soil, especially for the acidic soil. Additionally, in the current study, considerable soil Ca was declined (e.q., adsorbed by biochar) at the first week and at the end of the incubation test, which could due to the exchange of Ca from soil induced by the large amount of K released to the soil from biochar (Limwikran et al. 2018). Similar to Ca, the less considerable Mg decrease may also have interactions between K and Mg.

For available Ca and Mg, soil, biochar, rate and incubation time are all important factors (*Table 5*), but the interactions of soil, biochar, and the rate at the 57th weeks for Ca content are insignificance (*Table 6*). In addition, available Ca content only show significantly negative correlations with available K at the 57th weeks in SAO soil, but no significant correlations in MAI and SAI soil (*Table 7*). The available Mg content showed significantly positive correlations with available P in SAO soil; negative with C/N, positive with P and Fe in MAI soil; and negative with pH and C/N, positive with Fe, and negative with Cu and Pb in SAI soil. Ngatia et al. (2017) suggested that high biochar pyrolysis temperature favoured the accumulation of thermally stable C dominated by aromatic C as well as promoted increased alkalinity due to high concentration of Ca and Mg promoting P sorption and suppressing P availability. In this study, Mg seems more important than Ca for short-term and long-term P availability in SAO soil and short-term in MAI soil. The Mg retention would be resulted from the long-term increasing C mineralization and/or N mineralization of organic materials in MAI and SAI soil, and from short-term soil pH decreased in SAI soil.

Biochar effects on available Fe, Mn, Cu, Pb, and Zn retention and availability

Soil factor and incubation time were significant correlations with available Fe and Mn, while biochar, rate and most of the interactions showed insignificant correlations (*Table 5 and 6*). On the contrary, the available Cu, Pb, and Zn content were significantly affected by the soil, the biochar, the rate, the incubation duration, and almost all the possible interactions between these factors ($p < 0.01$-0.0001) (*Table 5*), but the rate at the first week showed insignificant effects ($p > 0.01$) on Cu Pb, and Zn contents (*Table 6*), and additionally the biochar has no significant effect on Pb. The studied biochars did not contain high Fe, Mn, Cu, Pb, and Zn concentration, except for higher Cu in A biochar and Zn in A and B biochar as compared to the compost and studied soils (*Table 1*). Thus, the insignificant effects of biochars addition (feedstock and rate) on the contents of those elements in biochar-amended soils could be expected, and the study results supported the expectation (*Table 3 and 4*). The previous study also indicated that the changes of F, Mn, Cu, Pb, and Zn ranged from –10% to +10%, and were variable in the three soils and less significant between biochar treatments and the control (Tsai and Chang, 2020a). In alkaline soils, the soil available Fe and Mn (diethylenetriaminepentaacetic acid, DTPA–extractable) increased with increasing biochar application rate (0, 1, 2, and 10% by weight), but did not affect soil Zn or Cu (Ippolito et al. 2014). In addition, at a higher biochar rate (10%, by wt.), co-applied with 2% manure, significantly increased DTPA-extractable Fe and Mn; and a synergistic effect between biochar and manure may have caused the increase in extractable soil Zn at the higher (2% and 10%) as compared to the lower (0% and 1%) biochar application rates; and treatment responses were not observed for extractable soil Cu (Ippolito et al. 2016). As compared to the unamended soil, the combined additions of biochar (2% by wt.) and compost (2% by wt.) significantly reduced Mn (-36%) and Fe (-14%) in the Oxisols; and increased P, K, and Ca content of the Ultisols and Oxisols (Berek et al. 2018). Typically, the soil Fe and Mn are complexed by soil organic matter in neutral and alkaline soils, the fast pyrolysis biochar addition may have also supplied labile organic C or primed decomposition of natural soil organic matter, leading to an increase in Fe and Mn in the soil solution (Ippolito et al. 2014). The slow pyrolysis biochars used in this study did not supply much labile C and prim decomposition of natural soil organic matter, but some available Fe and Mn. However, the excessive compost addition did have the potential for supplying much more labile C, primming natural soil organic matter decomposition, and directly adding higher available Fe and Mn. The increased effects can be found in MAI soil in the first week and continued to 57th weeks (*Table 3*). Besides, the decreases in available Fe and Mn were likely due to mineral forms changing from more to less available over time, and the slight increases in Zn and Cu concentrations may have been due to the slight decrease in soil pH over time (Ippolito et al. 2014).

Furthermore, the decline in the available content of Mn in biochar-amended soils could be attributed to the increment in soil pH (McBride, 1982; Idrees et al. 2018, Hailegnaw et al. 2020), and the exchange of Mn from soil exchange sites with exchangeable Ca^{2+} and K^+ originated from biochar also could contribute to the decline (Hailegnaw et al. 2020). In the current study, the available Mn content was only significantly positive correlated with available P and Fe at the first week within biochar treatments of the SAO soil (*Table 6*). However, it was only significantly positive correlated with available Fe at the first week and the 57th weeks within biochar treatments of the MAI and SOI soil. The effects of biochar-induced pH increment and ion exchanges are not obvious in this study. As soil factor is the most important factor (*Table 5 and 6*), this could confirm the less ability of biochar to reduce the availability of Fe and Mn irrespective of soils in the condition of excessive compost.

Metal adsorption to biochar could be one of the mechanisms by which metals are immobilized (Beesley et al. 2011). The effect of biochar on metal immobilization in soil may be influenced by the feedstock materials, pyrolysis conditions, and application rates (Yang et al. 2016). Several pieces of research indicated that an increase in soil pH would promote heavy metal adsorption and precipitation, thereby reducing their bioavailability and mobility in soil (Zhang et al. 2013, Lu et al. 2014, Yang et al. 2016). However, soil pH only showed significantly positive correlations with available Pb at the 57th weeks in SAO soil, and no significant correlation with Cu, Pb, and Zn in MAI and SAI soil (*Table 7*). The results indicated that the biochar-induced pH change has no significant effect on available Cu, Pb, and Zn in biochar-amended soils. The major reason could be attributed to the decomposition of excessive compost in biochar-amended soils resulting in soil acidification and offset the liming effects of alkaline biochar on biochar-amended soils. Moreover, increasing available P induced by biochar led to a reduced extractability of heavy metals, potentially by precipitation and complexation with phosphate (Ahmad et al. 2014). As shown in *Table 6*, available P was significantly positive correlations with Cu and Zn in SAO soil, Zn in MAI soil, and no significant correlation in SAI soil, in contrast with Ahmad et al. (2014). As compared within three studied soils (*Table 1*), the raw SAI soil has the highest CEC, highest available Mg and Fe content, and higher Mn content. The significant negative correlations between Cu and Mg, Fe, Mn, and Pb and Mg (*Table 7*), suggested that the release of Mg, Fe and Mn into biochar-amended soil will decrease the content of Cu and Pb, especially for Cu, in SAI soil. Thus, considerable Cu and Pb increased in SAI soil due to the large amount of exchangeable Mg, Fe, and Mn from soil and/or biochar have occupied the surface exchange site on soil and/or biochar.

The study of Qian et al. (2016) suggested that the formation of OH^-, CO_3^{2-} and Si species on biochars and the release of these species played the predominant role on Zn immobilization with the formation of various Zn precipitates/minerals, such as hemimorphite ($Zn_4(H_2O)(Si_2O_7)(OH)_2$), hydrozincite ($Zn_5(CO_3)_2(OH)_6$), and colloid $Zn(OH)_2$. The previous study (Tsai and Chang (2020a), see *Figure S1*) indicated that the L biochar contains more calcite relative to

A biochar but non for B biochar based on the XRD diffraction (XRD) patterns. As described above, we expect the formation of hydrozincite in L and A biochar treatments, which results in the higher decline of available Zn content in L and A treatments (2% and 5%) in three biochar-amended soils (*Table 4*).

Conclusions

The present study investigated the use of biochar as a potential amendment to retain excessive nutrient (P, K, Ca, Mg, Fe, Mn, Cu, Pb, and Zn) availability in three excessive compost fertilized soils. Our study results indicate that in the condition of excessive compost-fertilized, as compared to the effect on available K the primary and most consistent effects of biochar on the retention and availability of available nutrients (P, Ca, Mg, Fe, Mn, Cu, Pb, and Zn) are generally a smaller magnitude, and less consistent across soils, feedstock, and rate, but consistent across time. Soil conditions are the most important factor at the first week, and additionally, different biochar feedstocks and rates would gradually alter the soil conditions with time but the change magnitude is soil specific and smaller. Biochars with higher calcite content, like L biochar and A biochar in this study, have higher P and Zn sorption capacity, that is, these biochars have greater potential for reducing the environmental pollution. Based on the above conclusions, we accepted our hypothesis that biochar applications would cause a decreasing effect on the release of available nutrients and heavy metals, except for K. Our previous and current studies probe the interactions between biochar rate and excessive compost as an opportunity for C sequestration, N conservation and, simultaneously, keeping P and Cu, Pb and Zn in over-fertilized soils. In the condition of excessive compost, biochar application leads to efficient compost fertilizer use, and reduce soil nutrients and heavy metals pollution, and in further, can be a benefit for achieving the goal 3, 6, 11, 12, and 15 of the SDGs. For validating the laboratory findings, additional short-term or long-term field research should be investigated on different soil types and different agro-ecosystems to evaluate their effects in soil–crop systems. In addition, the potential of biochar to recover degrading soils and in further to preserve soil biodiversity is promising in excessive compost-fertilized soils in Taiwan.

Acknowledgments

This research was funded by National Science Council of the Republic of China, Contract number NSC-102-2313-B-197-002. Special thanks to Dr. G.S. Hwang, Forest Utilization Division, Taiwan Forestry Research Institute for supplying the ash, bamboo, and lead tree biochar.

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Chapter 14

Soil biodiversity under different uses and managements in Venezuela

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Introduction

Soil biodiversity can be described in many ways, including: *i)* ecosystem diversity encompassing the variety of soil habitats, *ii)* species diversity, understood as the variety and abundance of different types of organisms that inhabit a soil, which is similar to the concept of taxonomic diversity, *iii)* genetic diversity, which is the combination of different genes found within a population of a single species, and the pattern of variation observed within different populations of the same species, *iv)* phenotypic diversity, which is based on any and/or all morphological, biochemical or physiological aspects of the organisms in the soil, and is the result of genes and environmental factors, and finally, *v)* functional diversity, the variety of functions performed by soil biota, such as nitrification and litter degradation (Orgiazzi et al. 2016).

The contributions of soil organisms can be grouped into three categories. First, soil microorganisms (i.e., bacteria, fungi) and microfauna (i.e., protozoa and nematodes) that transform organic and inorganic compounds into accessible forms as part of their metabolism through complex biochemical processes, critical for ecosystem services. Second, soil organisms are part of a vast food web that cycles energy and nutrients from microscopic forms, through the soil megafauna to organisms living on the soil surface. An important part of the food web is represented by mesofauna, such as collembola and mites, which accelerate the decomposition of litter and improve the cycle and the availability of nutrients (especially nitrogen), and prey on the smallest organisms in the soil. Finally, the macrofauna and megafauna of the soil, such as earthworms, ants, termites, and some mammals, ecosystem engineers that modify the porosity of the soil, the transport of water and gases, bind the particles of soil in stable aggregates that reduce erosion (FAO et al. 2020) and improve the soil physical conditions.

In Venezuela, as in many other countries, the use of land for agricultural purposes has resulted, in many cases, in soil degradation, including the gradual deterioration of its structure, a decrease in organic matter content, loss of nutrients by erosion and leaching, acidification associated with the increasing input of nitrogen through nitrogen fertilizers, thus producing a reduction in the biodiversity and biomass of soil organisms (Rojas-Ordaz, 1995). Likewise, tillage is one of the practices that contributes to completely modify the soil environment where microorganisms live, affecting the microbiological and chemical properties of the soil, modifying the soil organic matter, the structural stability, and the water retention capacity (Gupta et al. 1994, Young and Ritz, 2000).

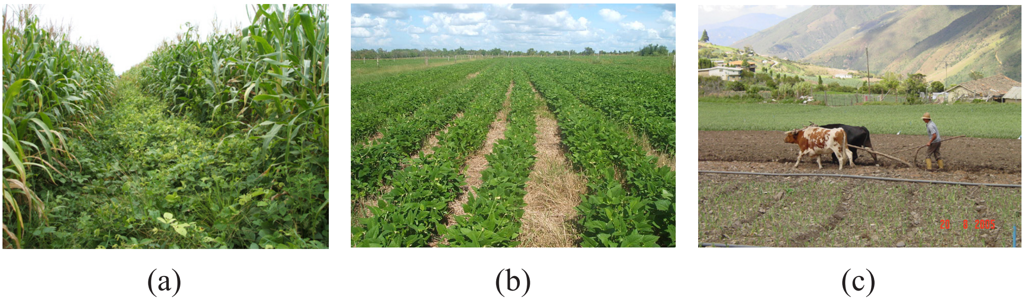
The soil ecosystem can also be altered by the application of pesticides, since the increase in the use of chemicals to enhance productivity can lead to a decrease in soil biota, especially the effects of pesticide interactions with soil microorganisms and negative effects of xenobiotics on the total population of microorganisms (Rojas-Ordaz, 1995). Likewise, xenobiotics can be resistant to additional degradation and be a threat to higher organisms (Liu et al. 1989).

In Venezuela, the role of microorganisms in soil formation and fertility had not been considered until 1995, highlighting at that time the research related to the nitrogen fixation by rhizobia, the organisms' pests and pathogens of animals and crops, some studies on mycorrhizal fungi and enzymatic activity as indicators of biological activity in the soil, as well as a few studies on the effects of pesticides on soil biota. Similarly, the effects of management practices on the physical and chemical properties of soils have been studied; however, from the microbiological point of view research was scarce (Rojas-Ordaz, 1995).

More recent studies have been carried out related to the impact of agronomic management practices for the sustainability of soil biodiversity in annual and permanent cropping systems, pastures, forest systems or agroforestry, as well as in the management and recovery of soils degraded or affected by non-agricultural activities in Venezuela, which are summarized in this work.

Soil management practices under short-cycle crops

Mechanized annual crops of cereals, grain legumes, oilseeds and textiles produced in the sub-humid zones of the lower tropic (western, central, and eastern plains of Venezuela), vegetables grown in low altitude and dry zones of the tropic (arid and semi-arid regions) and produced in high altitude, humid and cold zones of montane area (Andean region of Venezuela) are considered within this category. Some examples are shown in *Figure 1*.



(a) (b) (c)

Figure 1. Soil management practices in annual crops **(a)** Maize crop and *Centrosema macrocarpum* as a cover crop in the central plains, **(b)** *Vigna unguiculata* under zero tillage in the central plains, **(c)** Farmer ploughing the soil using oxen in the Andean region.

Traditional management of these agroecosystems is characterized by the indiscriminate, and in some cases, excessive use of soluble fertilizers and agrochemicals; the same applies to conventional tillage for soil conditioning and the establishment of monocultures, which has caused serious problems of soil degradation and the loss of soil biodiversity, with decreased productivity.

In recent years, different soil management strategies have been evaluated to make these production systems more sustainable, such as crop rotation, conservation tillage, crop diversification or polyculture, amendment, and nutrient management (organic, inorganic, and biological fertilization) and integrated pest management, among others. Due to their contribution to the increase of soil organic matter (SOM), many of these strategies improve conditions for soil organisms and therefore their biodiversity (González and Acosta, 2007, FAO et al. 2020).

Crop rotation has shown positive effects in maintaining high levels of SOM, especially when including legumes in the rotation, although the SOM accumulation rate is highly dependent on soil characteristics (structure, texture, mineralogy) and regional climatic conditions. Espinoza et al. (2007), in evaluations with the rotation maize (*Zea mays* L.) - cotton (*Gossypium hirsutum*) and maize - bean (*Vigna unguiculata*), observed changes in SOM related to the quantity and quality of plant residues returned to the soil, independently of the tillage system used. López et al. (2006) achieved a positive effect on soil properties and crop yields when rotating sorghum (*Sorghum bicolor* (L.) Möench) - pigeonpea (*Cajanus cajan* (L.) Millsp.).

Studies conducted by Zamora et al. (2005) with tomato (*Solanum lycopersicum* L.) - onion (*Allium cepa*) rotation, and by Mogollón et al. (2015a) with tomato – red pepper (*Capsicum annuum* L.) - onion rotation in soils affected by salts, showed that rotation improved the SOM in comparison with monoculture, which was reflected in the soil biological properties and their long-term quality. In the Andean region, Márquez et al. (2019) found that the production of native potatoes (*Solanum tuberosum* spp. Andigenum) managed under rotation with other vegetables, long fallows, and the use of organic amendments, was ecologically more sustainable than the production of introduced potatoes (*Solanum tuberosum* spp. Tuberosum) in monoculture and with inorganic fertilization.

Regarding tillage, the incorporation of crop residues in conventional tillage (CT) or its maintenance on the soil surface with reduced tillage (RT) and no tillage (NT) systems produced large changes in SOM, including the soil biological and biochemical activity. Experiments in different areas of the country have been conducted on tillage systems with crops, such as maize and sorghum, in monoculture or rotation with other crops. The results show that zero tillage or direct sowing (DS) allowed a higher activity of some enzymes at the surface level due to the increase in SOM content, depending on the residue quality and the previous management of the agroecosystem (Contreras et al. 2001, España et al. 2002, García and Rivero, 2012). In reduced tillage (RT) and no tillage (NT) systems, the maintenance of residues on the surface influenced the microorganisms' populations, regardless of their quality (Espinoza et al. 2007) and affected C and N fractions of the SOM (Espinoza, 2010). In soils susceptible to degradation, the associated crops have been proposed to improve the sustainability of production systems. In savannas dedicated to extensive livestock, management systems with maize associated with pastures of different qualities (grasses and legumes) were evaluated, to standardize the forage supply and make efficient use of the available forage throughout the year (Berroterán, 2000). Hernández-Hernández et al. (2004) and Lozano et al. (2018) achieved with this production system an increase in SOM and biological activity in a relatively short time (3 years). In semi-arid areas, Muñoz et al. (2016) found that under the association maize - squash (*Cucurbita moschata*) - beans - cassava (*Manihot esculenta* Crantz), there were better physical, chemical, and biological soil conditions, compared to the monocultures of melon (*Cucumis melo*) and *Aloe vera.*

In salt-affected soils, the effect of the application of organic amendments and conditioners (vermicompost, goat manure, aloe residues, residual sewage sludge, cacti extract, and biopolymers) on the soil biological and biochemical properties, was studied (Acosta and Paolini, 2006, Torres et al. 2015, Mogollón et al. 2016). The results showed that SOM, biological activity, and enzymatic activity responded positively to the vermicompost, even at low doses. Other properties related to salts affectation were also improved, since the pH values, exchangeable sodium and electrical conductivity decreased. The rest of the evaluated materials had a certain favourable effect on the soil biological activity.

Annual crops and beneficial microorganisms

The rational use of nitrogen and phosphate fertilizers in combination with agroecological management favours the biogeochemical cycling of nutrients (Toro et al. 2008; España et al. 2006), which allows plants to manifest mechanisms as exudation of organic acids, activation of enzymes such as acid phosphatase (López et al. 2007), changes in rhizospheric soil pH and the affinity for symbiotic microorganisms effective in plant nutrition such as arbuscular mycorrhizal fungi (AMF) (Toro and Andrade, 2021) and plant growth-promoting rhizobacteria (PGPR) (Vessey, 2003), both known as beneficial microorganisms (Dellagi et al. 2020) and/or biofertilizers. These microorganisms, properly selected for their effectiveness and strain-soil-plant compatibility (Rodríguez and López, 2009), are a viable source for the sustainable management of agroecosystems.

Regarding nutrient management and their effect on the soil biological and biochemical properties, research has been carried out in different areas of the country on the incorporation of crop residues or green manures, application of organic amendments, use of low solubility sources (rock phosphate, RP), and biological fertilization (AMF and/or PGPR).

The use of green manures from *Phaseolus lanatus*, *Crotalaria juncea* and *Penisetum purpureum* in maize, and C. *juncea* and *Vigna radiata* in radish (*Raphanus sativus*) cultivation, produced positive effects on SOM, on the number of microorganisms and their activity, as well as on the crops yield. The effect was related to the type and age of the green manure (Contreras et al. 1995, Fernández and Ortega, 2002). On the other hand, the combination of green manures from *C. juncea* (legume) and *S. bicolor* (grass), and different nutrient sources (soluble and low solubility) using sorghum as an indicator crop, did not show short-term changes (3 years) in AMF richness, but favoured the presence of phosphate solubilizing bacteria (PSB) in the sorghum rhizosphere (Toro et al. 2008).

It is known that organic amendments (manure, compost, vermicompost and sewage sludge, among others) improve soil structure, provide nutrients, and have a strong influence on the soil microflora, especially in low fertility soils. Contreras et al. (2006) and Matheus et al. (2014), evaluated the effect of adding different materials (poultry manure, goat manure, cattle manure, vermicompost and biofertilizers) on soil properties and crop response. The results showed that regardless of the soil used, C mineralization and the crop response were higher with the poultry manure. The differences in crop response found between the amendments were attributed to their composition.

In a savanna soil, maize system in direct sowing (DS) on cover crops of *Brachiaria dictyoneura* (BD) and *Centrosema macrocarpum* (CM) with different phosphoric fertilization sources: soluble fertilizers, rock phosphate (RP) and biological fertilization by AMF (BF), were evaluated. The results showed that cover crops and fertilization treatments affected SOM fractions, total and mineral N contents, and available P, mainly under CM cover crop and BF. BF maintained the previous levels of AMF diversity, although it modified the community composition, increased the percentage of root colonization and improved soil properties. The application of high doses of P notably reduced the AMF diversity, regardless of the source (Hernández-Hernández et al. 2004, Alguacil et al. 2010, Lozano et al. 2012, Rivero et al. 2012).

The property of plants to associate with nitrogen-fixing symbiotic bacteria is remarkable in soils of low fertility, such as Oxisols and Ultisols, frequent soil orders in Venezuelan savannas, highlighting the rhizobia-legume symbiosis (Ramírez-Bahena et al. 2016; Mora et al. 2017). López et al. (2010) isolated and characterized native rhizobia associated with legumes and free-living nitrogen fixers associated with maize, testing their nitrogen fixation capacity to select efficient strains, and producing biological resources for the formulation of biofertilizers.

In studies carried out with beneficial microorganisms in different soils and crops, López et al. (2008) and Rodríguez and López (2009) found that native nitrogen-fixing bacteria (NFB) (*Azotobacter* and *Rhizobium*) and PSB (*Bacillus megaterium* var. Phosphaticum) strains, showed potential to be used as biological fertilizers for maize and beans in low to very low fertility soils. On the other hand, Jiménez et al. (2017) found that AMF (*Scutellospora fulgida*, *Funneliformis mosseae, Glomus manihotis*) and cattle manure positively influenced the initial growth of chili plants (*Capsicum frutescens*), and that *F. mosseae* favoured mycorrhizal colonization, vegetative growth, and pigmentation of chili.

In soils of different fertility, it was found that total microbial population densities (bacterial and fungal) and PGPR in the rhizosphere of wild and crop plants, varied with soil conditions such as pH and SOM content, and with the crop species (Reyes and Valery, 2007). The use of PGPR strains alone or in consortium showed potential to increase in both production and grain quality of the maize only with NK fertilization. A certain discriminative power of the strains evaluated was also found due to the soil fertility conditions (Valery and Reyes, 2013).

Consistent response of PGPR (*Trichoderma koningi, Penicillium sp., Enterobacter sp., Bacillus megaterium*) as biocontrollers of the pathogen *Alternaria alternata* was found in tomato cultivation. The double effect of *T. koningi* as a growth promoter and resistance inducer against this pathogen shows its importance for the sustainable management of the crop (Alcedo and Reyes, 2018). The promotion of plant growth because of exo-radical interactions could be due not only to N_2 atmospheric fixation, phosphates dissolution or growth hormones production, but also to the suppression of pathogens through different mechanisms such as the release of siderophores, antibiotics, hydrolytic enzymes and hydrocyanic acid (Peña and Reyes, 2007).

It has been proven that intensive agricultural systems (soluble fertilizers, pesticides, and mechanization), negatively affect the reserves of C and soil microorganisms, mainly NFB and PSB were less sensitive to land uses and extreme conditions (Mogollón et al. 2010, Torres et al. 2012, Jaurixje et al. 2013, Mendoza et al. 2014). In semi-arid areas under intensive use, decreases of 50 to 86% in SOC reserves and of 36 to 68% in C-BM have been achieved (Mogollón et al. 2015b). Some practices and biodiversity indicators evaluated in the main production systems in Venezuelan eco-territorial units are presented in *Table 1*.

In studies of savannas of Guárico state in which conservationist managements were integrated, beneficial microorganisms were isolated, particularly rock phosphate solubilizing bacteria. Several species of the genera *Bacillus* sp., *Corynebacterium* sp., (Hernández-Hernández et al. 2015), the new species *Pseudomonas guariconensis* (Toro et al. 2013) and *Burkholderia cepacia*, were described. On the latter, its growth-promoting capacity was studied on maize (Mora and Toro, 2007). Likewise, its interaction with arbuscular mycorrhizae was tested in maize, showing that *B. cepacia* behaved as a mycorrhiza helper bacteria (Frey-Klett et al., 2007), promoting the activity of mycorrhiza and favouring the growth and development of maize (Mora et al. 2019).

Cabrales et al. (2019), compared the efficiency of mycorrhizal fungi native of Guárico savannas in the growth and yield of maize, finding that the consortium of native fungi and the fungus *Cetraspora pellucida* improved phosphorous nutrition of maize, achieving 100% yields with a 25% reduction in phosphorous fertilization. Reyes et al. (2002), studied the effectiveness of *Penicillium rugulosum* in the solubilization of phosphate rock, finding that its inoculation stimulated the growth of maize. These studies suggest the use of arbuscular mycorrhizae and plant growth-promoting bacteria for the sustainable management of production systems for maize; this practice may be suitable for other annual crops as well.

Eco-territorial Units	Main production system	Sustainable management practice	Evaluated indicators
Sub-humid zones of the lower tropics	Mechanized annual crops. Extensive or semi-extensive cattle livestock. Agroforestry plantations. Commercial fruit trees. Low altitude vegetables. Subsistence farming.	Conservation tillage. Crop rotation. Associated crops. Green manures. Organic amendments. Low solubility fertilizers. Biological fertilizers.	Total organic carbon (TOC), total nitrogen (TN), available phosphorus (P), potential C and N mineralization, C and N from microbial biomass (C-MB, N-MB), basal respiration, CO_2 evolution, enzymes activity (urease, dehydrogenase, protease, and acid phosphatase), Glomalin, C and N in the aggregates, % length of colonized roots arbuscular mycorrhizal fungi (AMF), spore density and AMF diversity, microbial populations (bacteria and fungi), N° nodules, nodule activity.
Humid and cold zones of the lower montane and montane areas	High altitude vegetables. Roots and tubers. Temperate climate fruit trees. Intensive cattle livestock for milk.	Organic amendments. Fallow. Crop rotation. Low solubility fertilizers. Biological fertilizers.	TOC, TN, P, C-MB, N-MB, basal respiration, enzyme activity (urease, dehydrogenase, and acid phosphatase), total microbial communities and hydroxyapatite solvents, AMF colonization, dark septate fungi (DSF), spore density and N° propagate infective, antagonism or biocontrol tests.
Dry zones of the low tropics	Irrigated low altitude vegetables. Drought tolerant crop plantations. Irrigated warm climate fruit trees. Subsistence farming. Extensive livestock with sheep and goats.	Crop rotation. Associated crops. Organic amendments. Biological fertilizers.	TOC, TN, P, C-MB, N-MB, hydrolysis of fluorescein diacetate (FDA), enzyme activity (urease, alkaline phosphatase, dehydrogenase, β-glucosidase), basal respiration, metabolic ratio (qCO_2), symbiotic N-fixing strains and free life, pH, electrical conductivity, exchangeable sodium percentage.

Table 1. . Management practices and indicators of soil biodiversity evaluated in different eco-territorial units of Venezuela, according to INIA (2016).

The use of plant residues constitutes a source of energy for the microbiota, activating specific microbial communities; their C/N ratio affects the quality of the residue. In a study carried out in a Vertisol from Guárico using molecular techniques (DGGE) (*Figure 2*), España et al. (2011a), found that *Streptomyces* sp. and *Arthrobacter* sp. were the prevailing bacteria in the decomposition of soybean residues. *Pseudonocardia* sp. and *Saccharopolyspora* sp. prevailed in maize residues decomposition. *Penicillium* sp. and *Aspergillus* sp. were the efficient fungi species in the decomposition of maize residues and *Fusarium* sp. and *Mortierella* sp. for soybean residues (España et al. 2011b).

The studies described the importance of beneficial microorganisms in promoting the growth and yield of annual crops, as well as the role played by soil microorganisms in the decomposition of plant residues, by improving the availability of nutrients in the soil. Its knowledge would lead us to the formulation of biofertilizers and products to facilitate the decomposition process in these agroecosystems, thus favouring their sustainable management.

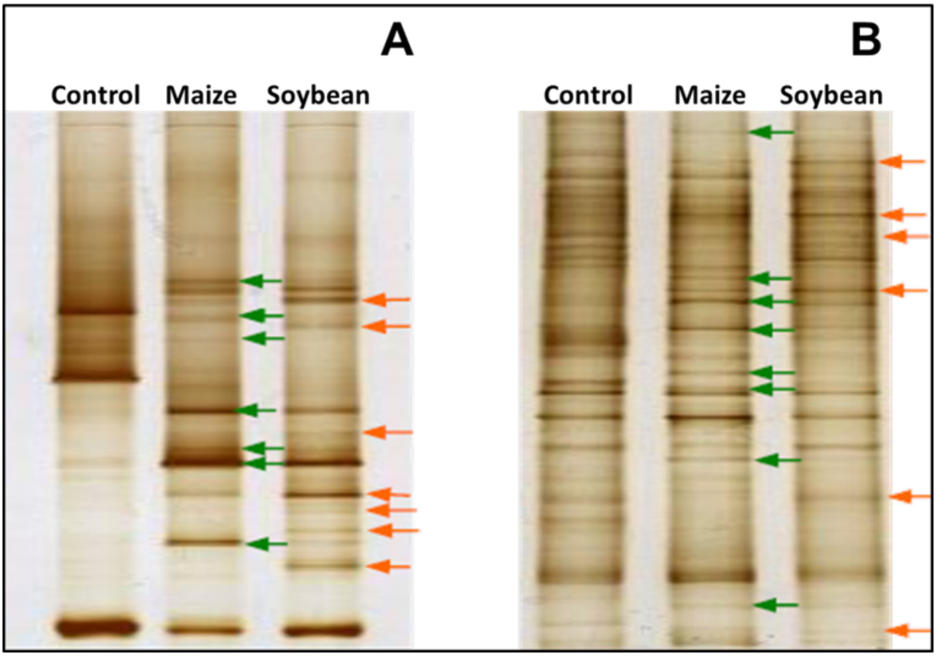


Figure 2. Structure of the soil microbial communities obtained 15 days after the incorporation of plant residues with different qualities (maize and soybean). **A**. Denaturing gradient gel electrophoresis (DGGE) profiles for fungal communities (18 S rRNA). **B**. DGGE profiles for bacterial communities (16S rRNA). The arrows indicate specific communities associated with each type of residue, absent in the control (adapted from España et al. 2011a and 2011b).

Permanent crops

Permanent crops have growth cycles of more than one year, are sown once for long periods and do not need to be replanted after each harvest as they can be harvested over several seasons, such as cocoa and coffee (FAO, 2021). In Venezuela, the main permanent crops are coffee, cocoa, bananas, plantains, pineapple, tobacco, citrus, and sugar cane (*Figure 3*). First, we will focus on coffee and cocoa as they are managed under agroforestry.

(a) (b) (c)

Figure 3. Representative permanent crops of Venezuela (a) Cacao plantation in the north-central region, (b) Banana plantation in the western region, (c) Orange plantation in the north-central region.

Permanent crops under agroforestry management

In Venezuela, coffee (*Coffea arabica* L.) is usually cultivated in vertical layers multi-crop system that could include a series of crops such as citrus, plantain, papaya trees, among others shade trees in the same plot. These agroforestry systems allow the efficient cycling of the nutrients provided by the litter and are considered low input management, in contrast to the traditional management that involves fertilization, and with the organic management that avoids chemical inputs in favour of grain quality. In a comparative study of these three managements in Andean plantations of Venezuela, Paolini (2017) found that farms with organic agriculture presented the highest values of total organic carbon, water-soluble carbon, microbial basal respiration, and biomass, compared to those with conventional management. The results suggest that the addition of fertilizers and pesticides negatively affected the functioning of microorganisms, making them more inefficient in the use of carbon and energy than conventional management. Likewise, Pardo-Plaza et al. (2019) working in a coffee agroecosystem in the north-central zone of Venezuela found that the soil microbiota showed sensitivity to changes due to climatic seasonality and coffee variety evaluated.

Permanent cultures can be treated in the nursery with agents that benefit their establishment in the field, such as beneficial microorganisms, as well as with pathogens controllers (Lozada, 2011). Colmenárez-Betancourt and Pineda (2006) achieved low rates of Anthracnose in coffee plants that had been mycorrhized at the seedling stage.

Meloidogyne exigua is a nematode well distributed in all coffee growing areas of Venezuela and causes great agronomical problems such as chlorosis, dwarf shoot development, leaf loss, root damage and decline in crop production of coffee plants. In nursery conditions, Alban et al. (2013) found that the early inoculation of mycorrhizae in coffee plantlets, allowed plants to regain the energy lost by the parasitic interaction. Arbuscular Mycorrhiza (AM) may help coffee plants with lignification of the plant cell wall cuticle. As the cuticle thickens it is more difficult for nematodes to penetrate and enter plant roots. Therefore, AM help coffee plants to uptake and transport nutrients, improving their nutritional status and stabilizing nematode attacks. It is suggested that symbiotic interactions help neutralize parasitic interactions. Gómez and Pulido (2019), in studies with coffee plantlets in the nursery, observed that the co-inoculation of the fungus *Trichoderma harzianum* and arbuscular mycorrhiza improved the phytosanitary quality of the seedlings in nursery stages.

In Venezuela, cocoa (*Theobroma cacao* L.) is a traditional crop, which is grown under the shade of some trees, mainly legumes of the *genus Erythrina*. The management of this crop is done with low input technology and farmers usually do not apply agrochemicals. The cultivation of cocoa in Venezuela reaches up to 68.76% of the economic activity in the north coastal region. The inhabitants of the zone recognize that the use of mycorrhizae increases production and reduces the problems that affect cocoa development and health (Bolívar et al. 2009). In greenhouse experiments, Cuenca et al. (1990) inoculated different species of mycorrhizal fungi, obtaining promising results in the promotion of cocoa growth and improvements in Cu and Zn nutrition thanks to mycorrhizal inoculation. A survey made by Cuenca and Meneses (1996) in sixteen plantations of cacao in northern Venezuela showed that AMF colonization was present in all of them. Percent colonized root length varied between 14% and 69% and a negative correlation was found between % AM colonization in cacao roots and available soil P that can be explained by the well-known depressive effect of P on AM infection. Fifteen species of AMF were described in the sites. Diversity of AMF communities did not vary with plantation age and none of the measured indexes (Richness, Diversity index, Shannon-Wiener index, or Simpson's dominance concentration) varied significantly with the age of plantations. However, there was a significant negative correlation between richness, diversity (calculated according to Whittaker) and available P in soils. Also, the Shannon-Wiener index was positively correlated with soil organic matter content and might reflect that AM has some saprophytic capacity.

Rivas and Pavone (2010) managed to control in vitro the pathogen *Crinipellis perniciosa*, that causes the Witch's Broom disease which significantly affets cocoa, with native strains of *Trichoderma*, isolated from the rhizosphere of cocoa plants of the central-north region of Venezuela; these results indicate the potential of using these isolates in the biological control of the phytopathogen *C. perniciosa*.

Regarding other permanent crops, González and Cuenca (2008) found that the inoculation of *Musa* AAB cv. Harton with foreign mycorrhizal fungi improved the growth and production of fruits of plantain, compared to native fungi, particularly the fungus *Scutelospora heterogama*. The authors recommended the use of this species instead of natives as an alternative to increase plantain production.

Delgado et al. (2010) developed a procedure for the determination of a soil quality and health index for banana plantations in Venezuela. Sixty chemical, physical, and microbiological indicators were measured in six banana farms selected in Trujillo and Aragua states. The test was carried out to verify the convenience and functionality of the various indicators used. The farms were selected from a wide range of contrasts on management practices and productivity levels (export farms and local production farms) and were divided into different areas based on productivity (bad/good). The best explanatory variables related to production were selected based on multivariate data analysis techniques. Eleven variables were selected as determinants of banana productivity levels and related to the soil health index. Five of the eleven variables selected were biological: microbial respiration, total organic carbon biomass, number of free-living nematodes and bacteriophages and populations of *Trichoderma*. This highlights the importance of soil biodiversity in the lands with Musaceae.

González et al. (2021) evaluated areas cultivated with plantain in the south of Maracaibo Lake through soil microbial activity. They selected lots of high and low vigour plantain plants (HV and LV) and determined microbial carbon, basal soil respiration and microbial and metabolic ratios. These authors found that the vigour parameters: number of hands per bunch; pseudostem circumference and height of sons were significantly higher in HV in comparison to LV, while, for microbial carbon and basal soil respiration, no statistically significant differences were found between batches of HV, but were high for tropical soils, and suggest a better quality of the degrading organic substrate for the HV lot. The high microbial activity was closely related to the soil texture and, in turn, this positively influenced the biometric parameters of the plants.

Soil management practices under pastures

The Llanos ecoregion in Venezuela occupies approximately 241,476 km^2, which is equivalent to almost a third of the Venezuelan territory. The savannas are one of the most widespread ecosystems, with low fertility soils, a markedly seasonal climate, limited to the development of extensive livestock (López-Hernández et al. 2008). The native vegetation of savannas is dominated by grasses that are usually used for cattle raising. To improve the forage supply in the Central Plains, in quantity and quality of biomass produced, the native grasses of the savannas have been replaced by introduced grasses, such as *Brachiaria dictyoneura* and *Centrosema macrocarpum*, used as cover crops, together with conservation tillage systems, improving the soil properties (*Figure 4*). Some of them are improved when the cover crop is grass and others when it is a legume (Lozano et al. 2010).

In savanna soils of the Venezuelan Amazon, favourable changes in soil fertility have also been found with the association of cultivated pastures *Urochloa dyctioneura*, *Stylosanthes capitata* and *Centrosema macrocarpum*, in comparison with natural vegetation, highlighting a higher net colloidal charge density associated to the highest carbon sequestration in the first 10 cm of about 74 g(C) m^{-2} y^{-1} (Ojeda et al. 2009). The replacement of natural vegetation in the savannas of the eastern plains by *Brachiaria decumbens* has shown impacts on the exchange of CO_2 at seasonal and annual scales, finding that the annual measurements of the net production of the ecosystem indicated that the field of *B. decumbens* constituted a strong carbon sink of 216 g (C) m^{-2} y^{-1}, while in the herbaceous savanna was only a weak sink 36 g (C) m^{-2} y^{-1}. This high net ecosystem production reflects the adaptation of *B. decumbens* to lowland conditions and its potential in helping to mitigate greenhouse gas emissions (San José et al. 2014).

(a) (b) (c)

Figure 4. Soil management practices under pastures **(a)** Natural vegetation in Venezuelan savannas, **(b)** Improved maize-livestock system in the central plains, **(c)** Livestock in the central plains.

Regarding the effects on microbial activity in soils in the eastern plains, Gómez and Paolini (2011) found that the introduction of adapted forage grasses, such as *Brachiaria brizantha* and *Andropogon gayanus* promoted significant favourable changes in total organic carbon and water-soluble carbon, as well as in the measurements of substrate-induced respiration, basal respiration, the dehydrogenase activity, the fluorescein diacetate hydrolysis and the arginine ammonification, in comparison with the natural savanna and the grazed natural savanna.

To evaluate the effects of vegetation burning (a common practice in *Trachypogon* savannas) and grazing, some soil quality parameters were compared between the savanna protected from burning and grazing for 25 years and the unprotected savanna. Significant positive effects were found in the protected savanna in the physical, chemical, and biological variables evaluated (lower bulk density, higher water storage capacity, as well as a higher content of exchangeable bases, organic carbon, total N, organic P and total P). The highest values of microbial C and microbial N are also highlighted (López-Hernández et al. 2008).

Results of evaluations carried out by Espinoza (2004) in soils of the Central Plains, which include the incorporation of fresh residues of grasses (sorghum), legumes (*Indigofera lespedicioides*) and wild vegetation (*Trachypogon* sp., *Axonopus* sp.) showed that the incorporation of legume residues increased carbon mineralization and stimulated microbial activity. Likewise, legumes residues contribute to increasing macroaggregates and weakens fractions of C, which suggests a conservation of the entrances of C into the soil. Similar results were found by Lobo et al. (2013) with the incorporation of cover crops (*Centrosema macrocarpum*, *Brachiaria dictyoneura* and *Brachiaria decumbens*), where an increase in macroaggregates was evidenced.

In paramo conditions (Venezuelan Andes), Morales and Sarmiento (2002) found that the cultivation of the natural paramo has a negative effect on the community of soil macroinvertebrates, reducing its density, richness, and diversity. After six years of succession, only the density is totally recovered. Characteristic morphotypes of each successional phase were found, that could be indicators of environmental quality and/or perturbation. A positive correlation between the number of soil fauna morphotypes and plant species and between soil fauna and plant diversity was found.

Biological N_2-fixation seems to be an option to maintain the productivity of improved pastures in savanna soils. Studies carried out in this sense to determine the biological N_2-fixation by three legumes of tropical pastures (*Centrosema pubescens*, *Stylosanthes hamata* and *Pueraria phaseoloides*) cultivated in monoculture and associated with the *Brachiaria humidicola* grass, using the ^{15}N isotope dilution technique, allowed to demonstrate that *B. humidicola* as monoculture had the highest shoot biomass production among all treatments, compared with the associations. Likewise, monoculture legumes had a very significant proportion of N derived from biological fixation, with *C. pubescens* as the best N_2-fixing legume. A higher contribution of biological fixation was also observed in legumes when they were mixed with the grass. A clear transfer of N from the legume to the companion grass was not observed, since ^{15}N in *B. humidicola* was always higher in mixed swards (Gil et al. 1997). The effect of inoculation with Rhizobium strains and nitrogen fertilization on the biological fixation of N_2 by *Pachecoa venezuelensis* (forage shrubby legume) have also been evaluated in soils of the eastern savannas, demonstrating the existence of native rhizobia in the soils studied, which can effectively nodulate the legume, while nitrogen exerted a restrictive effect on the chlorophyll content and foliar nitrogen (Marcano et al. 2001).

The low productivity of pastures in the savannas is mainly since they are sown or are established in acid soils, with low natural fertility, low content of organic matter, with P being one of the most deficient nutrients. This has led to studies on the effects of natural and modified rock phosphates on the production and quality of introduced pastures, the available P in the soil, the animal load capacity in pastures and the production of meat and milk from farms in various states of the country (Casanova, 2007). The fractionation of P in *Trachypogon* savanna soils revealed that the relatively high P content in microorganisms suggests the important role of this pool as a source of labile P and as a catalyzer of P transformations. Although available P forms are scarce, microorganisms and plants can satisfy their P requirements from the organic labile and moderately labile fractions, which account for 24% of total P in these soils (Hernández-Valencia and López-Hernández, 1999). In studies carried out in the rhizosphere of *Trachypogon plumosus* Nees (López-Gutiérrez et al. 2001), it was found that colonization by arbuscular mycorrhizae and seasonal changes in both microbial activity and acid phosphomonoesterase activity could be crucial mechanisms for the availability and uptake of P in savanna ecosystems.

In soils of La Gran Sabana, southeast of Venezuela, where the arable layer was removed, soils were replanted with *Brachiaria decumbens*, in addition to the application of lime and fertilizers, an increase in the number of mycorrhizal arbuscular fungi spores was shown after replanting (Cuenca and Lovera, 1992). However, the diversity of these fungi did not recover to the level of natural ecosystems even 7 years after the disturbance occurred (Cuenca et al. 1998).

Studies carried out by Mora et al. (2013) revealed that arbuscular mycorrhizae (AM) associations are relevant in savanna soils since the native plants present a high symbiosis affinity; moreover, AM colonization and propagules are found in an important number of plant species. On the other hand, the presence of phosphate solubilizing bacteria (PSB) and high root colonization by AM suggest a synergic effect in the plant uptake of phosphorus. Likewise, the nitrogen fixation by the different mechanisms existing in savannas appears as an option to supply N to the plant system. N and P supplies are likely acting together or in a synergistic form in the soil-plant system, therefore they constitute an important nutritional mechanism of biological origin.

Studies carried out by Morales-Márquez et al. (2018) in savanna soils to characterize the macroinvertebrate community in the natural savannah dominated by the *Trachypogon vestitus* grass (its structure and diversity, its time variation, and its relationship with the physical and chemical properties of the soil), revealed that Coleoptera, Hymenoptera and Isoptera were the most dominants orders. The relationship with soil properties differed according to families: Lampyridae, Aphodiidae and Formicidae had a positive correlation with soil macroporosity (radius of the pores > 15 µm); Tenthredinidae and Tenebrionidae had a positive correlation with the microporosity (pores < 15 µm). The community in general, correlated positively with soil porosity and negatively with bulk density.

Agroforestry and forest ecosystems

The type of vegetation cover can substantially modify the characteristics of the soils and consequently, the diversity and abundance of the biota found therein. In agroforestry and forestry systems, trees extract nutrients from the deeper horizons of the soil and deposit them in the superficial horizons through the litter, favouring their recycling and reducing their losses. Additionally, the contributions of organic matter increase the humus content, thereby increasing the cation exchange capacity and reducing the loss of nutrients. In some cases, the organic matter added to the soil by the trees moderates extreme changes in pH, and therefore the availability of essential nutrients and toxic compounds. Since nitrogen, phosphorus and sulfur are mainly found in organic form, their content can influence nutrient availability through mineralization. In another sense, the type of biota that inhabits soils can influence nutrient cycling, carbon transformation and storage, soil structure, and intra and interspecific biological regulation. Microbial activity tends to increase under trees, due to the higher content of organic matter, water availability and less extreme temperatures, a separate mention deserves the association of trees with nitrogen-fixing organisms and mycorrhizae that improve the availability of nutrients for plants. In Venezuela, studies on the soil biota of agroforestry and forest systems have been scarce and focused on their diversity and especially on the effects of different types of management on nutrient cycling.

López-Hernández et al. (2009) found that the transformation of a savanna ecosystem of the Venezuelan Amazon, with acid soils, poor in nutrients and excessively drained in an agroforestry system fertilized with compost, increased the abundance and biomass of earthworms, being the most important genera *Onycochaeta*, *Goiascolex* and *Pontoscolex*. Additionally, total N, available P, and exchangeable Ca, Mg and K, microbial C, N and P and the activity of the enzymes acid phosphatase and urease were higher in the agroforestry system, which is not only a product of the contributions of organic matter through compost, but also the activity of earthworms and other soil biota to decompose plant residues and improve the nutritional status of soils. According to the results of Netuzhilin et al. (1999) ants were the most numerous and diverse group in the savanna studied. Besides the ants, Colleoptera, Gryllidae and Araneida were the most abundant macroarthropods collected. The organic management in the agroforestry system has led to some changes in the macroarthropod community structure. Total individuals were lower in the native savanna and Diplopoda, Chilopoda and Isopoda appeared in the agroforestry system.

Medina et al. (2011) experimented for several years to evaluate the effect of the number of years, the season, the association modality, the grazing frequency, the pruning strategy, and the depth in the behaviour of the edaphic macrofauna in a grass monoculture system and associated with *Morus alba* and *Leucaena leucocephala*. During consecutive samplings for three years after the establishment of the woody plants, an increase in the richness of orders was observed, from 5 to 12 families and in the number of individuals from 10 individuals m^{-2} of soil in the pasture to 30 individuals m^{-2} of soil within silvopastoral systems. In addition, a higher density of edaphic individuals per unit area was observed in the first 10 cm of the soil, compared to the deeper strata (20-30 cm). The edaphic macrofauna was significantly affected by climatic periods and grazing frequency. The highest number of organisms was observed during the rainy season and with the grazing more widely spaced (every 120 days). The pruning strategy did not cause important variations in the number of individuals studied. In all cases, arthropods and annelids were the most represented phyllum; as well as diplopods, insects and arachnids for classes. Regarding the orders, the coleopterans, the hemiptera, the haplotaxids and the orthoptera stood out as the most significant. In this regard, in the study area subjected to the highest grazing frequency, detritivore organisms (Collembola), such as other entomophages were identified, due to the greater deposition of feces by cattle. Also, the presence of coleopterans of the Scarabaeidae family should be noted, since they are buriers of excreta in silvopastoral systems; a highly significant function as they participate in the recycling of nutrients and favour the decrease of the parasite load in grasslands.

The transformation of any natural ecosystem into a forest system leads to an important soil modification, not only in the total carbon pool, but also in the carbon associated with the microbial biomass. Zabala and Gómez (2010) studied the changes in fungal and bacterial biomass in pine plantations (*Pinus caribaea* var. *hondurensis*) established in a tropical savanna, to determine if these parameters are sensitive indicators of changes in the carbon content. The quantification of fungal and bacterial biomass was carried out by using cyloheximide as fungal inhibitor, and streptomycin and chloranphenicol as specific bacterial inhibitors. This research reveals that this land-use change exerted a significant effect on soil microbial biomass and shows that in pine plantations the fungal component is dominant, in contrast to the native savanna, in which the bacterial biomass dominates. On the other hand, the substitution of native savanna by pine plantation promotes a major soil carbon sequestration.

These studies indicate the usefulness of microbial parameters in the diagnosis of the quality of agronomic management in crops. Likewise, they highlight the convenience of the use of beneficial microorganisms and controllers of plant diseases since they minimize the dependence of use of chemical fertilizers and biocides and the impact they generate on the environment. It is convenient to deepen studies to make the use of beneficial microorganisms and soil biology a widespread practice in agriculture in Venezuela.

Soils contaminated with hydrocarbons

Oil production is one of the main economic activities in the country and there have been more than 15,000 oil pits and numerous spills that have produced soil contamination (Hernández-Valencia et al. 2017). In addition to collecting the remaining hydrocarbon, bioremediation is the most widely used practice to manage soil contamination by oil and is carried out with the stimulation of indigenous microbial populations (Infante et al. 2010). In contaminated soils, several organisms have been identified with the ability to degrade or transform hydrocarbons, among which are bacteria such as *Yersinia rohdei*, *Pantoea agglomerans*, *Sphingobacterium thalpophilum*, *Actinobacillus capsulatus*, and fungi belonging to the genera *Fusarium*, *Penicillium*, *Trichoderma*, *Aspergillus*, *Neosartorya*, *Pseudallescheria*, *Cladosporium*, *Pestalotiopsis*, *Phoma and Paecillomyces* (Naranjo et al. 2007, Díaz et al. 2013, Medina et al. 2014). In general, the degradation of hydrocarbons is more efficient in consortia of different species of microorganisms that allow a greater capacity to degrade the different petroleum compounds, compared to the use of a single species with more limited capacities.

As an example of the above, Zamora et al. (2017) assessed the changes after 120 days in the petroleum hydrocarbon content and the activity of microbial communities in soil contaminated with oil crude. Three treatments were established: a) fertilized soil (F: contaminated soil with organic and inorganic fertilization), unfertilized soil (WF: contaminated soil without fertilization) and abiotic control (AC: contaminated soil fertilized and sterilized). The content of oils and greases, basal respiration and use of specific organic substrates by microorganisms were determined. They found that the decrease in the content of oils and greases followed the order F>WF>AC. Differences in patterns of substrate utilization by microorganisms in treatments fertilized and unfertilized were also found with a decrease in functional diversity in treatment without fertilizer. They conclude that the functional structure of the microbial community was more stable in fertilized soil, and consequently more efficient in degrading the oil crude contaminant. This study also highlights the importance of fertilization as a source of nutrients (N and P) in the production of biomass of microorganisms during the degradation of the hydrocarbon.

As defined by Vangronsveld et al. (2009), phytoremediation is the use of plants and associated microorganisms to remove, contain, inactivate, or degrade harmful environmental contaminants, and can enhance the degrading activity of microorganisms, as found by Mager and Hernández-Valencia (2013). They observed that the content of microbial carbon and the activity of the dehydrogenase enzyme were higher in soils treated with plants, in comparison whit soils that did not have plants. Unfortunately, most of these tests have been carried out under greenhouse conditions.

Considering the possible role of arbuscular mycorrhiza (AM) in phytoremediation, Infante et al. (2013) studied plants and their phytoremediation potential in zones contaminated by hydrocarbons, describing 28 species, most of them typical of disturbed habitats. These plants also presented the AM association, and in the case of legumes, they also presented nodules typical of rhizobial symbiosis. The presence of AM and rhizobia nodules suggests that both symbiotic associations may favour the establishment of plants in soils contaminated by hydrocarbons.

In Venezuelan central east savannas, Toro et al. (2017) found that most plants growing in oil-contaminated soils had the AM symbiosis. They identified the morphotypes of Glomeromycota fungi, finding 86.21% with characteristics of *Glomus* sp., 10.34% belonged to the genus *Acaulospora* and 3.45% to the genus *Enthrophospora*. In greenhouse trials in which native mycorrhizal fungi were inoculated to *Brachiaria brizantha* growing on soil contaminated with 4% of the same hydrocarbon, it was found that up to 79.75% of the hydrocarbon was degraded in the rhizosphere of mycorrhized plants (measured as total petroleum hydrocarbons, TPH).

Gamarra (2014) observed that a greater number of microorganisms with the potential to degrade hydrocarbons grew in the rhizospheres of mycorrhized plants. The effect of specific microbial groups growing in mycorrhized rhizospheres has been described by Coninx et al. (2017) and suggests cooperation between microorganisms for the degradation of the contaminant. The results indicate the potential use of native mycorrhizae and bacteria with oil degrading potential for the rehabilitation of soils contaminated with hydrocarbons.

A vision of the future on soil microbiology and molecular techniques

Soil is a complex ecosystem, inhabited by millions of microorganisms that make up the soil microbiome (Nannipieri, 2014). Their function is known as many of these microorganisms, but their microscopic size and complex morphological differentiation provided the vision that the edaphic microbial world was a great black box whose functioning was known but not all its microscopic members. Traditionally we have studied microbial activity through the determination of the microbial biomass, the quantification of processes such as denitrification, edaphic respiration, biological nitrogen fixation or enzymatic activities, but the real knowledge of the number of microorganisms remained in the quantification of the viable organisms through the Most Probable Number or counts on selective media prepared in the laboratory. Over time, it was appreciated that these counts underestimated the real number of microorganisms in the soil: not all of them grow on selective media, so many of these microorganisms were left uncounted (Schloter et al. 2018). It is estimated that at least 99% of the prokaryotes present in the soil are unable to grow in culture media, which is why most of the prokaryotic diversity of the soil is underestimated (Daniel, 2005).

Since the 1980s, an important advance in the knowledge of soil microbial diversity has been produced by incorporating new study techniques that do not require prior cultivation of microorganisms, based on the analysis of molecular markers such as phospholipid fatty acids (PLFA) and nucleic acids (DNA and RNA). These markers are present in all cells, can be extracted directly from soil samples without the need for previous cultures and allow different groups of microorganisms to be differentiated. Using these markers, not only the composition of microbial communities can be determined, but the abundance of specific microorganisms can also be quantified.

After the important technological development undergone by molecular biology in recent years, the use of nucleic acids applied to the study of microbial communities has generated a large amount of information. Its extraction has been achieved efficiently thanks to physical, chemical and enzymatic processes and the elimination of complex substances (i.e., humic acids) that are extracted together with nucleic acids and interfere with subsequent molecular analyzes (Thies, 2008). Currently, commercial methods or kits for DNA extraction from soils are available, which are highly efficient in most cases.

However, there are still a good number of questions about microbial aspects of the soil and particularly in tropical areas, for example, the changes that occur in these communities in response to different environmental factors and/or disturbances and the functional role of the different types of microorganisms that make up the community. The advances achieved today with the application of genome sequencing and analysis techniques contribute to elucidating several of these unknowns. The study of the genomes of all microorganisms present in a community or metagenomics has been successfully applied in soil microbial communities and in the characterization of non-culturable microorganisms (Hirsch et al. 2010), which contributes to determining their role in the edaphic ecosystem. Figure 5 shows a scheme of the methodological approach to be carried out for the study of the soil metagenome, according to Daniel (2005).

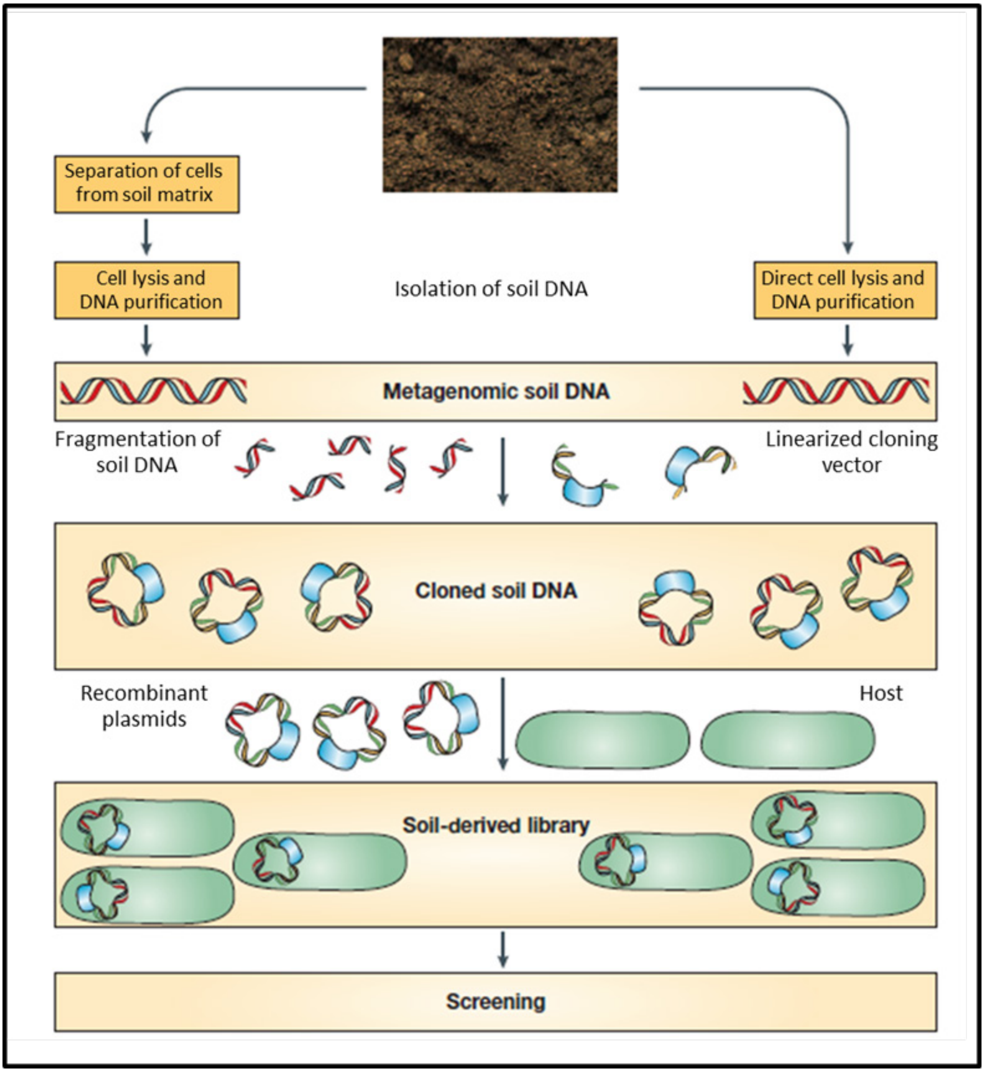


Figure 5. Essential steps to explore and study the genomic diversity of soil microbial communities by metagenomics. Soil DNA is extracted by separating the cells from the soil particles, followed by cell lysis and DNA extraction or by direct lysis of the cells. The recovered DNA is fragmented and ligated with a vector of choice. The recombinant vector is then introduced into a suitable bacterial cloning host. Then, using different strategies, the clones with the genes of interest are identified (adapted from Daniel, 2005).

Among other disciplines related to genomics, metabolomics stands out, which has focused on the detection of new biocatalysts or lytic enzymes and antimicrobial bioactive compounds (Nannipieri et al. 2014). Other omic techniques such as proteomics, study on a large scale the proteins of living organisms and the proteome or complete set of proteins of an organism or system; metatranscriptomics defined as the analysis of the expression of the genes of the microbial community in a particular environment; volatilomics, which studies volatile organic compounds emitted by organisms, and proteogenomics, which uses a combination of proteomics, genomics, and transcriptomics to help identify peptides. Molecular approaches also provide improved tools for seeking new inoculant consortia that may provide benefit in cropping systems or remediate contaminated soils, among other applications, as suggested by Thies (2008).

The omic techniques are promising, but still require revisions to be definitively and extensively applied (Nannipieri et al. 2014). The possibilities generated by omic techniques and other molecular scopes are wide, and important advances are expected in the understanding of the functioning of the edaphic microbiome communities, much of which remains unexplored in tropical systems.

These techniques are used today for studies of soil biology in many countries, but few or none are available in Venezuela yet. The few studies that apply molecular techniques cited in this review are done in collaboration with foreign laboratories. It is necessary to train personnel and acquire the necessary equipment for its implementation, which will contribute to the knowledge of soil biology of Venezuelan soils.

Acknowledgment

The authors thank Ms Auxi Toro García for reviewing the english language of this manuscript.

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Chapter 15

Understanding the multifunctionality of soil biodiversity patterns across pedodiversity

Stefano Mocali and Edoardo A.C. Costantini

Introduction

Soils contribute achieving of sustainable development goals of Agenda 2030 through the many ecosystem services they provide. The main soil ecosystem service is the ability to provide biomass, in the form of food for humans and animals, wood and fibre. Soil organisms recycle dead organic matter (O.M.) into a mineral form that plants can use for cell formation and vegetative growth. The value of net primary production (NPP) reaches, globally, 550 Gt per year (Le Quéré et al. 2018), but differs considerably according to the climate and the type of soil. In Amazonian soils, for example, NPP is on average 12.8 ± 0.9 Mg C ha^{-1} $year^{-1}$ (average ± standard error) but varies between 9.3 ± 1.3 Mg C ha^{-1} $year^{-1}$ and 17.0 ± 1.4 Mg C ha^{-1} $year^{-1}$, respectively in poor sandy soil and a very fertile site of Terra Preta, with on average 64 ± 3% of the total NPP aboveground and 36 ± 3% belowground (Aragão et al. 2009). The ability of soil to receive and recycle the biomass produced by plants and animals also makes it the main terrestrial reservoir of organic carbon, with the absorption of about 2,400 Gt per year, about five times higher than that of plants and animals (560 Gt) and three times that present in the atmosphere (800 Gt) (Le Quéré et al. 2018). It is therefore understood how soils can be so important to mitigate climate change, removing about 25% of the equivalent carbon emitted every year through the use of fossil fuels in the world.

Among their diverse abilities, soils are also capable of hosting great biodiversity. Soil biodiversity plays an important role in supporting the sustainable productivity of ecosystems and regulating multiple other ecosystem services, including nutrients cycling, organic matter decomposition, and pathogen control in natural terrestrial ecosystems (Han et al. 2021, Bardgett and Van Der Putten, 2014). An agricultural soil usually contains living biomass in the order of 3,000 kg (fresh weight) per hectare. This is the equivalent of 4 cows, 60 sheeps or 35 farmers (Bloem et al. 2003). Although the estimates are still very uncertain, recently FAO has reported that at least a quarter of global biodiversity is present in soil and that around 40% of animal species spend at least part of their life within the soil (FAO et al. 2020). Other authors estimate the share of animal biodiversity present in the soil to be 23% (Decaëns et al. 2006). Even if such values are just raw estimations, they give an idea of the huge amount of this “unseen majority” living belowground (van der Heijden et al. 2008). Soil biota is fundamental to human life itself (Blum et al. 2019) and there is unequivocal evidence that biodiversity loss reduces the efficiency by which ecological communities capture biologically essential resources, produce biomass, decompose, and biologically recycle essential nutrients (Cardinale et al. 2012). Remarkably, a decline of soil and environmental biodiversity may directly affect the human commensal microbiota, thus increasing the occurrence of allergies and other chronic inflammatory diseases among urban populations worldwide (Hanski et al. 2012, Rook, 2013).

In agriculture, soil biodiversity is responsible for essential ecosystem functions and services, such as plant productivity, nutrient cycling, organic matter decomposition, pollutant degradation and pathogen control, which are valued at trillions of US dollars annually (Delgado-Baquerizo et al. 2020). However, the linkages between diverse soil organisms and ecosystem functions remain unclear and the roles played by biodiverse soil organisms (bacteria, fungi, protists, and invertebrates) for multiple types of ecosystem functions (ecosystem multifunctionality) remain largely unresolved. Rigorous assessment of the role of soil biodiversity in regulating multifunctionality is urgently needed to better understand the potential consequences of soil biodiversity losses for the maintenance of multiple ecosystem functions and services that are critical for human wellbeing and global ecosystem sustainability. Similarly, the variability of the relationships between biodiversity and soil functioning and ecosystem services across the European edaphoclimatic and management gradient is yet not well understood (Delgado-Baquerizo et al. 2020).

A largely unexplained aspect of soil biodiversity is the space-time pattern at different scales. The large number of paradigms and theories proposed attests its scientific interest, while it also underlines the complexity of the ecological processes involved. Most of the available surveys of soil biodiversity overlooked soil complexity and pedological features that might play a key role in determining soil biodiversity composition and (multi)functionality across different spatial-temporal scales.

In this chapter, we provide a synthesis of our knowledge about soil biodiversity patterns, addressing *i)* how does soil biodiversity respond to environmental gradients at specific scales of space and time; *ii)* which soil factors are driving soil biodiversity patterns and at what scales do they operate; *iii)* how pedodiversity and biodiversity might be linked. This work is also motivated by the applied issue of the conservation and management of below-ground biodiversity, for which a clear understanding of the factors that drive soil communities represents a necessary prerequisite.

Biodiversity and ecosystem multifunctionality

We have still limited knowledge of the relationship between microbial diversity and ecosystem functioning, particularly in soil. In fact, despite the availability of soil biological indicators and the increasing number of soil ecology studies, major gaps and/or geographic and taxonomic biases exist in our understanding of soil biodiversity (Guerra et al. 2021). A lack of matching data for soil biodiversity and multiple ecosystem functions was observed in current global datasets and no consistent relationships between soil species diversity and soil functions have been found to date, implying that more species do not necessarily provide more services. Recent studies demonstrated that just the same few bacterial taxa can be responsible for most of the major services, like carbon cycling, in diverse ecosystems across the planet (Rannik and Kõlli, 2018), and that several species can perform the same function (Allison and Martiny, 2008). Indeed, the services provided by soil biodiversity should not be considered in isolation, but rather as different facets of a set of highly associated functions performed by soil biota. Such holistic knowledge is currently lacking, and we do not yet have an exact understanding of the potential interlinkages among services. Therefore, our understanding of biological communities should focus on the interspecific characteristics of the complex communities rather than on the individual/species level characteristics, such as species richness and abundance.

Over the past decades, ecological network studies (e.g., food webs, mutualistic and host-parasite networks) for macro-organisms along environmental gradients have been carried out, showing high variation in ecological networks in contrasting environments (Pellissier et al. 2018). Nevertheless, how microbial co-occurrence network characteristics (e.g., topologies) change across biogeographic gradients were rarely studied (Tu et al. 2020).

Biodiversity patterns and indicators

Biodiversity and its driving forces have been studied for decades in ecology, biogeography, and conservation biology. However, most of such effort has been focused on aboveground terrestrial organisms (e.g., birds and plants) and processes (e.g., food production), thus overlooking the majority of terrestrial biodiversity (Troudet et al. 2017). Remarkably, below and aboveground diversity do not necessarily follow similar ecological patterns, which suggests that protecting aboveground biodiversity may not sufficiently reduce threats to soil biodiversity and that even when the focus is on restoring wild areas or increasing carbon sequestration —both seen as positive outcomes of nature conservation—such practices might not have the same positive effects on soil organisms and their associated functions (Cameron et al. 2019). Several initiatives (i.e., Orgiazzi et al. 2016, Pavlů et al. 2021) have identified the most commonly used indicators for monitoring biodiversity. For example, EU projects like ENVASSO and ECOFINDERS were carried out to propose and recommend a set of suitable indicators for monitoring soil biodiversity and ecosystem services for EU agricultural sites (Griffith et al. 2016).

The results showed that the diversity of the microbial and faunal groups was very sensitive to soil management but responded differently to the changes in land-use intensity and that their ranking of biodiversity varied between sites. The authors concluded that for effective monitoring of biodiversity, all taxonomic groups would need to be addressed because changes in the biodiversity of one group cannot be used to infer changes in other taxonomic groups. These bioindicators will also require standardization and their deployment will need to be cost-effective and policy-relevant to be efficiently applied throughout Europe. Recently, a new soil ecological indicator monitoring system, based on essential biodiversity variables (EBVs) linked to global targets, has been proposed, to provide relevant information to specific policy targets and policies (Guerra et al. 2021). More specifically, EBVs for soil systems were proposed as a holistic system approach, where soil organisms are intertwined with relevant soil chemical, physical, and functional properties, contributing to overall societal well-being. Soils are not seen just as key elements for agriculture and forestry, but also for climate change and nature conservation sectors. For example, chemical and physical variables such as litter decomposition or soil aggregation, respectively, are considered EBVs as they contribute to determining soil health, thus contributing to overall nature conservation policy, as well as earthworm biodiversity. Such initiative (SOILBON) is just an example to provide a roadmap for researchers and policymakers on the priorities for data collection at the global scale and on how to integrate such information into policy design (https://geobon.org/bons/thematic-bon/soil-bon).

However, several conceptual and methodological constraints still need to be addressed and overcome. Overall, the interaction of species with their environment and neighboring species and/or their movement in space and time provides different species distribution and interactions across soils (*biodiversity patterns*). For example, traits linked to how carbon and energy are processed and transformed in biomass can describe separate niches.

The canonical examples are r and K strategists, where carbon and energy are either primarily invested in reproduction, or invested in tolerating biotic and/or abiotic stressors, respectively (Grime, 1977). Copiotrophs are considered as microorganisms with relatively high growth rates that have relatively poor growth efficiency (as carbon incorporated to biomass per unit resource), relatively high cell maintenance energy costs, dependence on relatively high concentrations of organic carbon in their environment.

They demonstrate rapid population blooms upon the addition of organic matter and do not tolerate abiotic stress very much (Finn et al. 2021). Oligotrophs are considered as the inverse: low growth rate, high growth efficiency, low cell maintenance energy requirements, high substrate uptake affinity, slow growth yet at a consistent rate, and are resilient to abiotic stress. Remarkably, members within a taxon tend to have similar life strategies and niches and possess common traits compared to members of other taxa (Philippot et al. 2010). While ecological coherence of taxa has been largely considered, the traits that differ between terrestrial prokaryote taxonomic groups, and whether these traits describe the niches they occupy, remain open questions.

Spatial and temporal patterns of soil biodiversity

Many paradigms and theories have been proposed to explain spatial and temporal patterns of biodiversity at different scales. Soil communities appear weakly structured by competition, although competitive constraints may account for assembly rules within specific taxa. The main factor constraining local soil biodiversity is the compact and heterogeneous nature of soils, which provides an unrivalled potential for niche partitioning, thus allowing high levels of local biodiversity. This heterogeneity is increased by the impact of ecosystem engineers that generate resource patchiness at a range of spatial-temporal scales. Recently, some authors proposed a conceptual framework based on dividing soil (S) into two spatial compartments (Thakur et al. 2020) (*Figure 1*): soil hotspots (S’) and soil microsites (S’’). Soil microsites are nested in soil hotspots and soil hotspots are nested in the soil. These compartments represent the size at which soil biodiversity can be studied. At S’ spatial scale, both bulk and rhizosphere soil are present, and all size groups of soil organisms can be sampled, even the litter layer. Within S’ compartments, the biodiversity of intermediate-sized soil organisms, such as soil meso- and microfauna are mainly studied. Often the study of soil microarthropods and their resources are carried out at this spatial scale. Within S’ hotspots, soil microsites (S’’) represent another spatial compartment, and this is the finest spatial scale at which biodiversity can be studied in the soil. Soil microsites could range from a tip of a plant root to even a single aggregate (µm to mm) in the soil. The study of soil biodiversity at this spatial scale focuses primarily on the diversity of microorganisms and their primary predators.

The soil system is polystructural and multivariate in space and time. Thus, how we measure diversity will depend on our intended application of the results, scale, etc. For example, it was shown that “hot-spots” of high specific denitrification activity were associated with particulate organic C material in the soil, thus supporting the concept that the patchy dispersion of particulate organic material is a major factor influencing the variability of natural denitrification rates (Parkin, 1987). Now this concept is very well established, and soils are considered as the most heterogeneous parts of the biosphere, with an extremely high differentiation of properties and processes within nano- to macroscales. Remarkably, the spatial and temporal heterogeneity of input of labile organics by plants creates microbial hotspots over short periods of time – the ‘hot moments’ (Kuzyakov and Blagodatskaya, 2015).

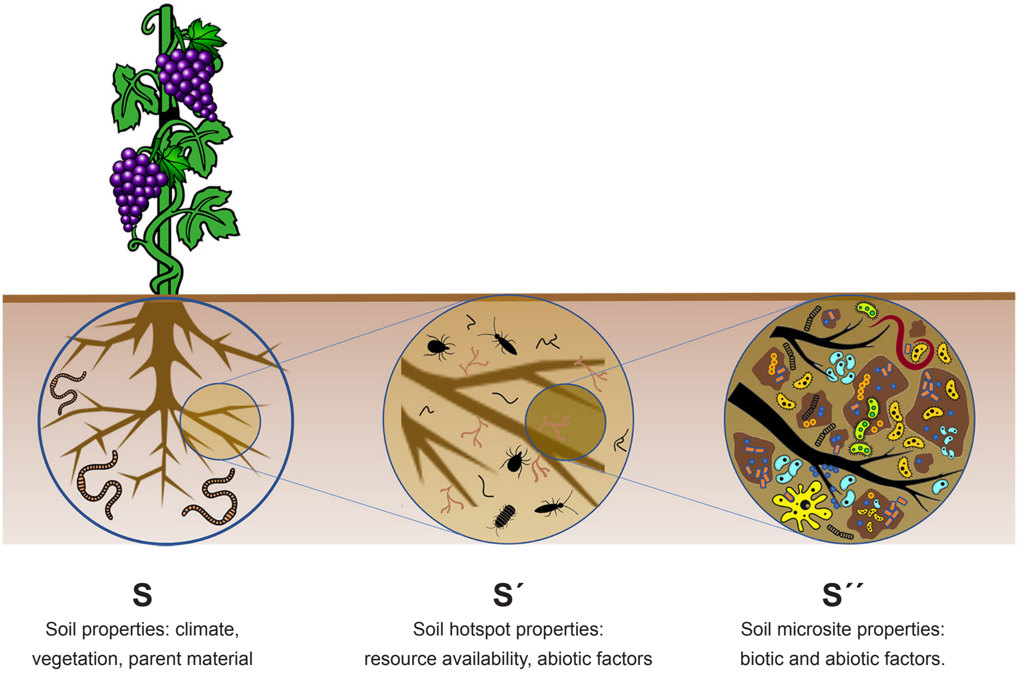

Figure 1. Illustration of spatial compartments in the soil for studying soil biodiversity from micro- to macro-organisms. The properties of each compartment that potentially affects the respective biodiversity pattern are listed below the compartments. Soil micro-aggregates are colored light brown in the S″ compartment; all organisms in S″ are either microorganisms or their predators (e.g., nematodes and protists). Note that microorganisms can colonize micro-aggregates as illustrated in S″ (modified from Thakur et al. 2020).

Microbial hotspots are defined as small volumes of soil with much faster process rates and much more intensive interactions compared to the average soil conditions. For example, microbial hotspots may be found in the rhizosphere, detritusphere, and on aggregate surfaces, but hotspots are frequently of mixed origin. Hot moments are short-term events or sequences of events inducing accelerated process rates as compared to the average rates. Thus, hotspots and hot moments are defined by dynamic characteristics, i.e., by process rates. The fraction of active microorganisms in hotspots is $2e^{20}$ times higher than in the bulk soil, and their specific activities (i.e., respiration, microbial growth, mineralization potential, enzyme activities, RNA/DNA ratio) may also be much higher.

Hot moments induce succession in microbial communities and intense intra- and interspecific competition affecting C use efficiency, microbial growth, and turnover. Consequently, spatial and temporal hotspots are mainly responsible for the ecologically relevant processes in soil and understanding how they affect soil taxonomical and functional soil diversity, as well as ecosystem services, is a priority.

Soil aggregates and biodiversity multifunctionality

There are many experimental and observational studies providing evidence that the relationship between biodiversity and ecosystem functioning is more linear than saturating (Wagg et al. 2014, Mora et al. 2014). Many studies have been conducted to establish the relationships between soil biodiversity and ecosystem functioning (Delgado-Baquerizo et al. 2016, Le Bagousse-Pingue et al. 2019). However, most of these studies focused on broader scales, while there is limited information on the variations in soil multifunctionality at the micrometer-scale (e.g., soil aggregates). And whether soil aggregate size is shaping multifunctionality is still an open question.

It is well known that stable soil aggregates are important drivers of soil fertility and microbial diversity, while are highly vulnerable to land degradation. Soil aggregates are the basic components of soil architecture and differences in pore size, oxygen potential, moisture content, organic matter, and predation pressure provide microscale heterogeneous habitats for distinct microorganisms. There are small (<2 mm), strongly bound clusters of minerals and organic carbon forms that persist through mechanical disruptions and wetting events. Their spatial structure is important for biogeochemical cycling, and we cannot reliably predict soil biological activities and variability by studying bulk soils alone. Thus, to fully understand soil functional diversity patterns, it is necessary to understand the micrometer-scale interactions that occur between soil particles and their microbial inhabitants. In a recent review, some authors suggested that aggregate-based approaches are a critical next step for developing a predictive understanding of how geochemical and community interactions govern microbial community structure and nutrient cycling in soil (Wilpiszeski et al. 2019). Instead, large composite samples are commonly favored to obtain an overview of microbial diversity at the scale of a plot or a field. This neglects the fine-scale heterogeneity of soil structure and thus loses much information on patterns of community assembly. A recent study demonstrated that DNA extracted from individual soil aggregates provides new insights into how microbial communities are assembled (Szoboszlay and Tebbe, 2021). Most bacterial cells occur inside aggregates rather than on their surfaces (Ranjard et al. 2000), and biogeochemical cycles are considered to mainly occur within aggregates (Wilpiszeski et al. 2019). Soil aggregates have been regarded as “massively concurrent evolutionary incubators” (Rillig et al. 2017) or as “microbial villages” (Wilpiszeski et al. 2019) that represent small communities separated by distance and physical barriers and connected only periodically, for example, during wetting events.

Pedodiversity and biodiversity

Another issue to be considered is that soil biodiversity differs by soil types and land use peculiarities and is different in crop-, grass- and forestlands. Moreover, the diversity of soil taxa in the soil cover composition (pedodiversity) and the distribution pattern of soils in landscape depend on the diversity of soil parent materials and geodiversity, landscape topography, land use, and the climatic conditions of the region (Costantini and L’Abate, 2016). Some studies showed how geodiversity had a great influence on both pedodiversity and plant cover diversity (Krasilnikov et al. 2007; Kasparinskis and Nikodemus, 2012), stating that the basis of the pedodiversity of any territory is its geodiversity (Serrano and Ruiz-Flano, 2007; Ibáñez and Bockheim, 2013) (*Figure 2*).

Geodiversity has been defined as “the natural range of geological, geomorphological and soil features, assemblages, systems and processes” (IUCN-WCPA strategic plan 2005-2012). Thus, geodiversity has a decisive role in the development of both soil cover and the biodiversity of its humus cover, as outlined by a recent paper, where a pedocentric approach to the formation of the biodiversity of a plant association and edaphon was proposed (Rannik and Kõlli, 2018). The first pedologist who tried to analyze and quantify the pedological diversity of a territory was V.M. Fridland (1974, 1976), but the first paper that formally explained the main tools for the analysis of soil diversity or ‘pedodiversity’ was published many years later (Ibáñez et al. 1995).

Although biodiversity and pedodiversity can be expected to be closely related at different scales and from different points of view, few studies have been conducted to test these relationships (Ibáñez and Feoli, 2013).

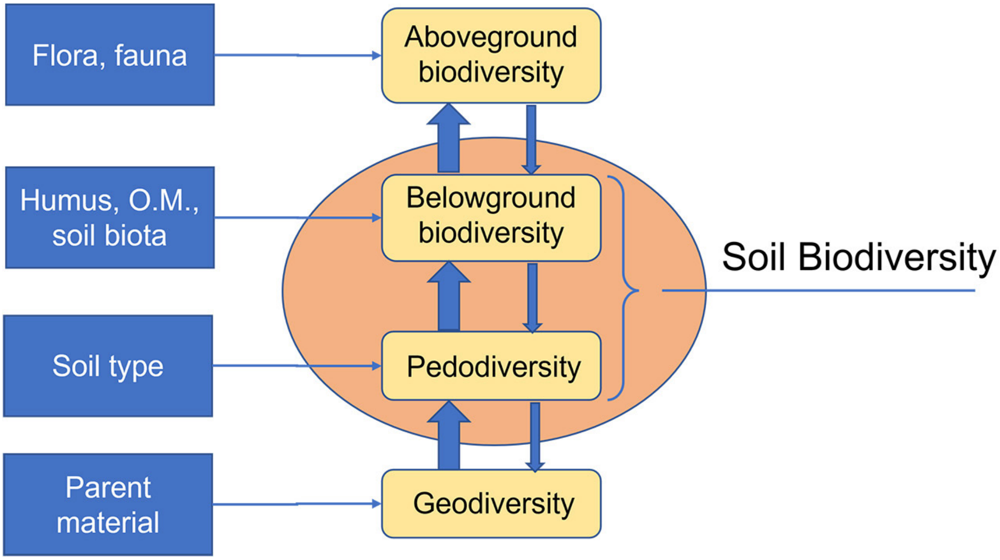

Figure 2. The influence of geodiversity on pedodiversity and through it on belowground and aboveground biodiversity.

Soil self-organization as a factor of biodiversity

To provide high ecosystem services, soils must be healthy and well-functioning. As mentioned above, the soil is a complex dynamic system, an ecological machine where processes take place for both short and long or very long times. It is precisely the dynamics and the products of these processes that determine the morphological and functional characteristics of the soil. Soil formation requires more or less long times, of the order of hundreds or thousands of years when the parent material is a rock that is difficult to alter, while it can take a few years for incoherent sediments (Costantini et al. 2015, Mardhiah et al. 2014). Soil formation is a self-organized process of gradual accumulation of solid products of the functioning of the soil, it is finite over time and tends to reach a stationary state (Targulian and Krasilnikov, 2007). The processes that occur in the soil and between soils lead to their self-organization which can be observed at different levels, from the micrometric to the geographical scale (Phillips, 1995). In the context of the field investigation, soil self-organization is understood by studying the profile and arrangement of its mineral and organic components in specific and distinctive forms, particularly the horizons and pedogenetic features (Jahn et al. 2006) (*Figure 3*).

Figure 3. Soil profile exhibiting distinct vertical spatial patterns. Below the O horizon where the plant debris accumulates (few centimeters thick), there is the plough layer, rich in humus (Ap horizon, up to about 30 cm), followed by the well-structured cambic B horizon (25 cm thick) and the reddish BC horizon, until 70 cm. The parent material is made of light brown fluvial sands. The groundwater fluctuates at about 110 cm depth. Every horizon offers different spatial and temporal boundary conditions to soil biodiversity and its functionality.

There are marked shifts in microbial communities and abiotic conditions with soil depth. Although most studies have focused exclusively on the microorganisms found in surface soil horizons or layers, communities found in the litter layer (or O-horizon) are often distinct from those found in underlying mineral soil horizons (A and B horizons) and deeper saprolite (C horizons). Key soil properties — such as pH, organic carbon concentration, salinity, texture, and available nitrogen concentration — can vary substantially across these distinct soil environments.

The shape and size of aggregates and porosity are among the main soil characteristics, which are particularly important because they indicate both the processes of soil formation and the functionality of the soil itself. The self-organization of the soil is a phenomenon that normally occurs in nature as a consequence of the factors of pedogenesis, but human activity can profoundly modify the self-organization processes of the soil, up to be the dominant factor. In this case, the soils are classified as Anthrosols (IUSS, 2015). The soil is a dynamic and open system that tends to find a balance according to the environmental conditions in which it is found. The process of soil self-organization tends to reach a state of dynamic equilibrium over time with the flows of matter and energy in its environment, in which the entropy of the system is minimized (Addiscott, 2010; Targulian and Krasilnikov, 2007). Soil self-organization is part of the natural heritage, but it also has an economic value, because the processes of soil self-organization and those that determine the functionality and produce ecosystem services are parallel (Robinson et al. 2009). Therefore, soil degradation can also be defined as the process that leads to the decline of the level of self-organization of its constituents due to natural causes or, more often, to human activities.

As previously mentioned, the way organisms are distributed in soil and behave in nature can vary widely depending on the spatial characteristics in which they are found; for this reason, biotic communities can differ considerably according to the type of environment and, in the case of telluric organisms, the type of soil.

A complex and tortuous soil structure increases shelters for predated organisms, reduces competition, increases the number of species, and facilitates nutrient retention. Soil structure, defined as a distribution of aggregate/porous space of different sizes, can therefore be an important control of soil biological communities, which are highly structured in their size distribution. Soil aggregates are often more important than age and vegetation type in differentiating major microbial taxa. In particular, macroaggregates may have larger α diversities and higher community microbial stability, accompanied by low microbial respiration rates (Sun et al. 2021).

An increase in the angularity of the aggregates causes a decrease in the growth of bacteria and fungi, but the fungi decrease more than the bacteria. As the sharpness of the angle increases in an elongated porous space, the degradation of organic matter decreases (Arellano-Caicedo et al. 2021). This phenomenon, together with the ability of clays to bind with organic matter, explains why soils with dynamic clays and well-developed angular and prismatic polyhedral aggregates can turn black and capture organic carbon for very long times.

A significant example of how the different types of soil self-organization reflect environmental conditions that correspond to the differentiation of soil organisms is that of humus profiles (Zanella et al. 2018). In the passage of Mor-type humus forms to Moder and then to Mull, the existence of a gradient of increasing soil biodiversity with increasing humification of organic matter is highlighted. Open ecosystems such as meadows, pastures and agricultural fields, with Mull-type humus, see the prevalence of larger-sized invertebrates such as earthworms, ants, millipedes, termites, etc., and great biodiversity. On the other hand, in the Mor-type humus forms, typically encountered in closed ecosystems such as alpine forests, on siliceous substrates and in cold and humid climates, specialized groups prevail, tolerant to acids, such as enchytraeidae, mites and springtails and lower overall biodiversity. They are therefore more specialized ecosystems and sensitive to variations. Indeed, there is growing evidence that biodiversity increases the stability of ecosystem functions over time (Cardinale et al. 2012).

Pedodiversity and management of below-ground biodiversity

The soil-organisms complex can be viewed as a self-organizing system capable of adapting to prevailing conditions (Young and Crawford, 2004). Therefore, by ignoring the interaction between physical and biological processes in soil, we might be missing its most essential feature.

Pedodiversity shapes the habitat variety for soil organisms and should be carefully considered in designing conservation plans and management of below-ground biodiversity. It has been reported that during nature restoration on abandoned arable land a compositional shift in soil biota, preceded by tightening of the belowground networks, corresponded with enhanced efficiency of carbon uptake. More specifically, in mid-and long-term abandoned fields (5 and 15 years, respectively), the interactions among species in the soil community increased while carbon uptake became more efficient, even without major changes in species composition of the dominant plants, and without an increase in fungal biomass or shift in the bacterial-to-fungal ratio (Morriën et al. 2017). Such findings imply that during nature restoration the efficiency of nutrient cycling and carbon uptake relate more strongly to the belowground community network structure than to belowground biodiversity.

The soil self-organization that, according to the local factors of pedogenesis, differentiates the vertical and horizontal pattern of soil features is the main source of the diversity of network structure of the belowground biological community. The same self-organization is also probably the cause of the decoupling of above and belowground biological patterns and functionalities across the different spatial and temporal scales (Costantini and Mocali, 2022).

Thus, the nature and architecture of soil constituents dictate the life and the connections between organisms, but also the response to aboveground variations and management. In this sense, soil physical and chemical features can be distinguished according to the degree of “permeability” to organisms, between those features that allow a relatively rapid movement to most organisms, from those which are selective, and those which are very limiting or impeding. For this reason, the efforts made to preserve or enhance soil biodiversity must consider the favorable or limiting conditions given by soil self-organization.

Conclusions

Soil is an incredibly complex and diverse organization of pores and particles, which influences the organisms that live within it. It might be considered more than just a single environment, since it encompasses a wide range of environments that can contain distinct microbial communities. Different soil environments may be only micrometers to millimeters apart, but they can vary considerably in their abiotic characteristics, microbial abundances, rates of microbial activity, microbial community composition and interactions.

Any loss in microbial diversity as a consequence of global environmental changes such as land use, soil management and climate change would likely alter the capacity of microbes to sustain multiple above- and belowground ecosystem functions. Actually, soil biodiversity loss and simplification of soil community composition were shown to impair multiple ecosystem functions, including plant diversity, decomposition, nutrient retention, and nutrient cycling. Nevertheless, a shred of evidence on the direct relationships between microbial diversity and multifunctionality in terrestrial ecosystems at a global scale has been reported only recently (Delgado-Baquerizo et al. 2016).

The restoration of soil biodiversity and multifunctionality must be planned not only through the management of the aboveground biodiversity but also considering the boundary conditions given by soil self-organization and pedodiversity to biological relationships. Future research attention should be therefore given to the understanding of the interactions between biodiversity patterns, functionalities, and specific soil features of the whole profile.

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Chapter 16

Soil and water conservation promote soil biodiversity

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Introduction

Soil erosion is the process that soil, soil parent material and other ground components are broken, detached, transported, and deposited under the action of external forces. It can be classified into water erosion, wind erosion, gravity erosion, and freeze – thaw erosion according to the difference of external forces. Water erosion and wind erosion are the dominat types. Soil Erosion is the principal cause of land degradation and desertification. Erosion decreased soil organic matter content, deteriorated soil physical structure, aggravated soil salinization, affected the growth and succession of plant communities, thinned soil layer, and polluted soil, all of which threatened soil biodiversity. For the sustainable development of human society, soil and water conservation practices have been carried out all around the world. Soil and water conservation include preventive and rehabilitative measures taken against soil erosion. Preventive measure is to minimize the extent and intensity of human disturbance to land caused by farming, forestry, and animal husbandry, urbanization and infrastructure construction activities through process optimization and management, to keep the soil with sustainable use and thus maintaining soil biodiversity. Rehabilitative measure is to restore land productivity and promote soil biodiversity on the eroded lands (such as, slope land, mountain economic forest and fruit land, wind-eroded farmland, and grassland) that have degraded and cannot be self-recovered, by taking appropriate biological, engineering and agricultural tillage measures. Erosion control measures maintain and improve soil physical properties and soil organic matter and have significant positive effects on soil biodiversity. Two examples are given to show the role of soil and water conservation in the restoration and promotion of soil biodiversity. Although studies of soil biodiversity have yielded some results, it is necessary to further deepen the research on the impact mechanism of soil and water conservation measures on soil biodiversity.

Global soil erosion and damage

Global soil erosion

It is estimated that the global soil erosion area is about 16.43 million km^2, accounting for 10.95% of the total surface area (Eswaran et.al. 2019). The global water erosion area is 10.94 million km^2, the wind erosion area of 5.48 million km^2. The most serious area of water erosion is located within 50 °N ~ 40 °S, especially in the United States, Russia, Australia, China, India, the Mediterranean Europe and some countries in the North Africa and South America. The areas most affected by wind erosion are the Great Plains of the United States, the Sahara and Kalahari Desert of Africa, the Northwest China and Central Australia.

The spatial pattern of soil erosion in 2012 is illustrated in *Figure 1* (Borrelli et.al. 2017a). Areas classified as having very low, and low erosion rates (class 1 and class 2), represent about 71.9% and 12.7% of the total, respectively. Moderate (class 3) and high (class 4) soil erosion values account for about 4.2% and 5.1% of the study area, respectively. The remaining land surface (classes 5-7), about 7.5 million km^2 in total (6.1% of the land), exceeds the generic tolerable soil erosion threshold (T-value) (10 Mg ha^{-1}yr^{-1}) in 2012.

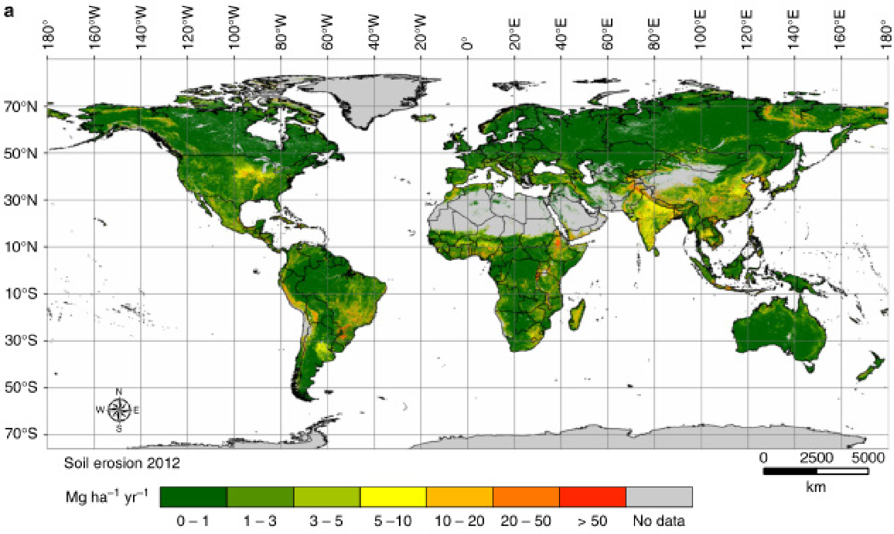

Figure 1. Soil erosion 2012.

Globally, Asia and Africa are the regions with the most severe soil degradation caused by erosion. According to the United Nations Environment Program, more than three quarters of Africa's land is at risk of soil degradation, and three quarters of its agricultural land has been degraded to varying extents. The Sahara Desert in Africa is expanding every year, with more than 15,000 square kilometers of land becoming desert each year in the southern part of the desert. In Nigeria, more than 3,500 square kilometers of land are turned into sand every year. More than 70% of Asia's land is degraded and 35% of its agricultural land is being affected by desertification, particularly in the countries such as Mongolia, Afghanistan, Pakistan and India, Asia is also the most densely populated region in the world affected by land degradation due to its large population. Soil degradation is also severe in the United States, where 25% of the topsoil has been removed through 200 years of large-scale farming.

Wind erosion is considered one of the important land desertification processes in areas exceeding 90% of the Egypt area in the western desert, eastern desert and particularly Sinai. These areas are characterized by a fragile ecosystem, scarcity of vegetation cover and severe drought. Studies indicate that wind erosion rate in Egypt is about 5.5 t/ha a year in oases areas in the western desert, and 71-100 t/ha a year in areas of rainfed agriculture on the northwest coast showing wind erosion risks in these areas fluctuating between moderate and severe classes. During rain falling periods in winter season, the water erosion in the north coast, Red Sea, Aqaba Gulf, and south Sinai coasts and some areas of eastern desert valleys, is considered one of the most serious desertification processes in these areas as studies are indicating that water erosion impact in Egypt on lands is low to severe.

According to the Survey of Soil and Water Conservation in the National Water Resources Survey (Ministry of Water Resources, People's Republic of China, 2013), in 2011, the national water and wind soil erosion area was 2,949,000 km^2, accounting for 31.1% of the survey area. Serious soil erosion is one of the major environmental problems and eco-civilization construction issues, which seriously restricts China's economic and social development. The annual economic loss due to soil erosion is 28.762 billion yuan, accounting for 1.45% of the GDP (Ma et al. 2009), and nearly 50% of the national poor counties are located in serious soil erosion areas (Li et al. 2007).

The total area of water erosion was 1.29 million km^2, accounting for 7.42% of the total area of the surveyerd territory. Based on magnitude of soil loss, erosion intensity was divided into five groups: light erosion, moderate erosion, severe erosion, most severe erosion, and extremely severe erosion. Soil loss rates in these groups was measured at 500-2,500 t/(km^2·a), 2,500-5,000 t/(km^2·a), 5,000-8,000 t/(km^2·a), 8,000-15,000 t/(km^2·a), and greater than 15,000 t/(km^2·a), respectively. Light soil erosion covered 667,600 km^2, moderate erosion 351,400 km^2, severe erosion 168,700 km^2, most severe erosion 76,300 km^2, and extremely severe erosion 29,200 km^2, accounting for 51.6%, 27.2%, 13.0%, 5.9% and 2.3% of the total soil erosion area, respectively. Light erosion covered the largest area, followed by moderate erosion, together representing 78.8% of the total erosion area, and the three degrees of severe erosion accounted for 21.2% of the total erosion area.

According to the report of State Environmental Protection Administration of China (SEPA, 1999), there are 83.7×104 km^2 of all kind of desertification lands, accounting for 8.7% of total territory. There are 38.57×104 km^2 of aeolian desertification lands, accounting for 44.3% of desertified land. The report also showed that there are 141×104 km^2 of lands susceptible to desertification, the area of land susceptible to aeolian desertification is 53.7×104 km^2. Desertified land and land susceptible to desertification amount to 224.7×104 km^2, and land of aeolian desertification and threatened by aeolian desertification amount to 90.8×104 km2. In addition to 219.1×104 km^2 of sand desert, Gobi and blown land account for 22.82% of the total area of continental territory.

The predicted potential soil losses from sheet water erosion in Bulgaria amount to an annual average of 902.5 million tons, including 344.4 million tons from Northern Bulgaria and 558.1 million tons from Southern Bulgaria. More than 50% of the soil erosion losses in Northern Bulgaria are formed from the territory of 3 districts: 31% (106.6 Mt/a) from Lovech district, 22.3% from Gabrovo (41.6 Mt/a) and Veliko Tarnovo (35.3 Mt/a). More than 50% of the soil losses from erosion in South Bulgaria are formed from the territory of 4 districts: 20.2% (112.6 Mt/a) from Sofia district, 10.7% (59.5 Mt/a) from Sliven district, 10.1% (56.4Mt/a) from the Burgas district and 9.6% (53.3 Mt/a) from the Kardzhali district. The average annual predicted potential soil losses from sheet water erosion from the territory of Stara Zagora, Plovdiv and Blagoevgrad districts are between 41.3 and 49.6 Mt/a; from Vidin, Montana, Vratsa, Shumen, Targovishte, Haskovo, Kyustendil and Pazardzhik are between 20.2 and 25.8 Mt/a, from Varna – about 15 Mt/a, Sofia-city and Pernik – respectively 12.6 and 18.6 Mt/a; and from the districts of Yambol, Pleven, Ruse, Razgrad, Silistra and Dobrich are between 4.9 and 9.2 Mt/a.

The estimated potential soil losses from wind erosion of the arable lands with a gradient of less than 3° in Bulgaria amount to 1163.5 thousand tons, including 759.7 thousand tons from northern Bulgaria and 403.8 thousand tons from southern Bulgaria. Approximately ¾ of the total soil losses from deflation are formed in the territory of 6 districts - Dobrich (476.5 kt/a), Burgas (139.7 kt/a), Varna (97.9 kt/a), Yambol (68.5 kt/a), Sofia-district (45.0 kt/a) and Sofia-city (40.3 kt/a). The average annual forecast for potential soil losses from wind erosion from Targovishte, Rousse, Sliven and Silistra districts are between 25 and 35 kt/a, from Veliko Turnovo, Razgrad, Plovdiv, Stara Zagora and Shumen - between 15 and 20 kt/a. Vidin, Vratsa, and Pleven - 10-11 kt/a, and from Kyustendil, Pernik, Kardzhali, Pazardzhik, Lovech and Montana - between 1 and 6.3 kt/a, and from Gabrovo, Blagoevgrad and Smolyan - under 1kt/a.

Erosion causes soil biodiversity loss

Erosion threats soil biodiversity at different aspects:

Erosion threatens soil biodiversity by decreasing soil organic matter content

As the degree of erosion intensifies, the organic carbon, total carbon and total phosphorus in the soil will be greatly reduced, resulting in soil impoverishment, which seriously threatens the survival and diversity of soil organisms and disrupts their important roles in the complex food webs and ecological functions in the ecosystem (Asuoha et al. 2019, Guerra et al. 2020, Qiu et al. 2021). Biodiversity in a natural ecosystem is directly related to the total amount of living and abiotic organic matter in the system (Asuoha et al. 2019, Filser et al. 2016, Liu et al. 2019). The effects of soil organic matter on soil organisms are manifold. Firstly, soil organic matter plays a key role in maintaining soil fertility, containing a variety of nutrients, especially nitrogen and phosphorus. Plants can directly absorb and utilize these nutrients under the microorganism decomposition and mineralization, which deeply affect the types, quantity and diversity of soil microorganisms and animals through living root systems and litters above and below ground (Sasse et al. 2017, Silva-Filho and Vivanco, 2017, Wu et al. 2018). Secondly, organic matter can actually improve several aspects of soil physical properties. The most important and direct role is to improve the soil structure and promote the formation of granular structures (Chen et al. 2010), thereby increasing soil porosity, improving soil aeration, and water permeability and storage (Naveed et al. 2014), and thus contributing to the activities of soil animals (Filser et al. 2016). The soil animals themselves accelerate the decomposition of organic matter (especially the decomposition of fresh organic matters) (Briones, 2014), which further improves soil permeability and heat preservation ability, and creates beneficial environmental conditions for the survival of soil microorganisms and plants (Filser et al. 2016, Or et al. 2007, Yang et al. 2019). However, several studies have found that soil destruction, separation, transportation and deposition in the process of soil erosion all have profound impacts on nutrient cycling (Lal, 2019, Quinton et al. 2010). Yan et al. (2005) reported the loss of soil organic carbon (SOC) by wind erosion in China was 75 Tg C/yr during 1990s. Yue et al. (2016) reported that water erosion displaced 180 ± 80 Tg C/yr between 1995 and 2015. Phosphorus could eventually escape the soil-vegetation biochemical cycle after being buried because it cannot be used by plants (Quinton et al. 2010). In addition, soil erosion is often accompanied by vegetation degradation, and severe vegetation degradation may have irreversible effects on soil structure and nutrient-related functions (Gao et al. 2011). A study found that the degradation of Amazon tropical rain forest vegetation significantly caused the decrease of soil carbon, phosphorus and cation exchange capacity (Celentano et al. 2017). Erosion-induced loss of available substrates and nutrients may restrict the survival of some nutrient-related taxa usually tend to decrease such as Bacteroidetes and Gemmatimonadetes (Qiu et al. 2021), which were reported to be positively correlated with soil nutrients (DeBruyn et al. 2011, Wolińska et al. 2017).

Erosion threatens soil biodiversity by deteriorating soil physical structure

Long-term soil erosion, especially water and wind erosion, had resulted in soil loss and profile configuration destruction, leading to the entire degradation of soil quality and quantity, including thinning soil layers, increasing surface soil bulk density and the proportion of coarser soil particles, and reducing water retention (Fattahi et al. 2020, Pimentel et al. 1995). It is well known that surface soil usually has a better physical structure than does underlying soil. Once the surface soil is lost, the lower layer will be exposed and thus the whole physical structure of soils is significantly reduced. For example, after the surface layer of red soil is eroded and stripped, the content of clay particles and iron-aluminum oxides in the lower layer increases, resulting in an increase in soil bulk density, a decrease in pores and infiltration performance, and thereby degrading structural indicators such as soil water, heat, and aggregate stability (Hao et al. 2019, Wu et al. 2017). In erode soils, the increase in soil thermal conductivity and decrease in soil heat capacity may result in greater daily and seasonal variation in soil temperature. Liang et al. (2018) have shown greater variation in soil temperature at eroded sites than at non-eroded sites. Thus, the loss of diversity and functionality of soil communities can indirectly be attributed to an increase in soil thermal variability, because most soil microbes are sensitive to local changes in temperature (Guo et al. 2018, van Elsas et al. 1998). Besides, erosion has destroyed the soil fundamental structure, which means the inherent habitats of soil organisms are also destroyed, causing a huge number of soil organisms to disperse, fluctuate and mix between habitats, which not only has an impact on the in situ soil biological communities, but also disturbs the original ecological stability of other habitats (Orgiazzi and Panagos, 2018). Supposing the existence of rare, probably still unknown, species of all soil organisms, soil erosion might be one of the main disturbance events in extinction processes (Veresoglou et al. 2015).

Erosion threatens soil biodiversity by aggravating soil salinization

Salinization affects soil physical properties, makes soil more likely to be harden, reduces porosity, and leads to soil permeability reduction. Salinization impacts soil chemical properties, increases ion concentration in soil solution, increases the pH value, increases the ratio of conductivity and exchangeable Na, reduces C and N mineralization. Soil salinity acts as an influential environmental stressor coupled with limitation of water availability and high intracellular concentrations of ions that are toxic to metabolic activities (Van Horn et al. 2014). It is found that some soluble salt ions in soil can directly affect the growth and reproduction of soil organisms to threaten the diversity of soil biological community. Soil animals are also highly sensitive to salinization (Owojori et al. 2009), Liu et al. (2006) found that with the increase of salinization, the community and diversity of Carabidae animals decreased. Salinization also affects the activity of soil microorganisms and the transformation of the organic matter, reduces soil nutrient utilization, organic matter content and soil fertility. Salinity is a major determinant of the microbial community, significantly decreasing the microbial diversity (Zhang et al. 2019), and the only taxa well adapted to high salt concentrations are able to prevail in high-salt environments, the strength of phylogenetic clustering is expected to increase when the environment is suitable for only a subset of microorganisms (Pontarp et al. 2013). Besides, high salinity in soil suppresses plant growth, decreases plant photosynthetic capacities (Paul, 2013), and poses a strong influence on the composition, distribution, and diversity of plant communities (Bui, 2013). It accelerates the process of soil biodiversity loss.

Erosion threatens soil biodiversity by affecting the growth and succession of plant communities

Vegetation, as primary producers, determine the quantity and quality of resource input in the ecosystem, and provide organic matter for the ecosystem, playing an important role in maintaining soil functions (Wardle, 2002). Root exudates and litter such as branches and leaves produced by plants and input into the soil have a direct impact on soil organisms. The cover of litter and its own branches and leaves can also indirectly affect soil organisms by changing soil physical and chemical factors (Zhao et al. 2012), and through the decomposition of microorganisms and soil animals to participate in the nutrient cycle of terrestrial ecosystems to effect belowground ecosystems (Cromack, 1981, Wardle et al. 2004). However, soil erosion has severely affected vegetation growth, development, and succession (Durán and Rodríguez, 2008, Milodowski et al. 2015). Erosion processes directly threaten natural vegetation regeneration and community composition by affecting seed redistribution, physically disturbing plants, or delaying plant growth seasons (Kou et al. 2016). In addition, long-term erosion can alter the physical template of soils, landforms and landscapes on which ecosystems depend for development (Quinton et al. 2010). Erosion causes soil impoverishment, slope increase, and terrain fragmentation, which has a “bottom-up” impact on vegetation and can dominate the structure and succession of vegetation communities on a local scale (Milodowski et al. 2015, Wang et al. 2007). For example, Guerrero-Campo and Montserrat-Martí (2000) found that erosion greatly reduced vegetation coverage in the northeastern Spain; Wang et al. (2007) observed a 26-year natural succession slope in the Donghe River Basin in Guangdong and found that the vegetation on the slope affected by erosion was short and the succession rate was very slow. Long-term soil erosion may affect the groundwater level and soil moisture status (Lal, 2001), and produce more lasting stress on plant habitats.

Generally, erosion strongly limits the coverage, diversity, and succession of communities through stress and disturbance to plants from seeds to seedlings to individual and community levels. This has a serious impact on the survival of soil microorganisms. Plant diversity can provide material sources and energy for soil animals (Pollierer et al. 2007, Scheunemann et al. 2015). Plants are the hosts of phytophagous nematodes, and the root environment of plants parasitized by phytophagous nematodes will affect the phytophagous nematodes, which may indirectly affect other functional groups of nematodes and the entire nematode community (Van der Putten and Van der Stoel, 1998). It is generally believed that the increase in plant species diversity can increase the net primary productivity of the plant community (Cross et al. 2006), thereby increasing the amount of resources input into the soil. Theoretically, the increase in plant species diversity will increase the diversity and abundance of soil animals. For instance, plant diversity increases soil nematode diversity by increasing the diversity of soil nematode food sources (De Deyn and Van der Putten 2005, Hooper et al. 2000). Gastine et al. (2003) found that the increase in plant root biomass would increase the density of predatory nematodes in the temperate grassland of Germany; Sohlenius et al. (2011) also found that different plant functional groups would have an impact on different nematode functional groups; Chen et al. (2016) show that the total number of nematodes increases with the increase the diversity of plant functional groups, and the abundance of nematodes also increases with the increase of aboveground biomass of plants on a large scale. In addition, soil microorganisms also obtain nutrients and living space from plant communities. Higher plant diversity increases rhizosphere carbon inputs into the microbial community, resulting in both increased microbial activity and carbon storage (Lange et al. 2015). Abundant microorganisms occupy their respective ecological niches, from the rhizosphere to the root surface to the inside of the plant roots (Hardoim et al. 2015, Lareen et al. 2016, Reinhold-Hurek et al. 2015). The decrease of plant coverage and diversity caused by soil erosion reduces the diversity and content of nutrients transported to the soil environment, which may inhibit the survival and communication between microorganisms and between microorganisms and plants (Asuoha et al. 2019, Durán and Rodríguez, 2008); moreover, the damaged root environment makes certain microbe-plant symbiosis, e.g., arbuscular mycorrhizal fungi (AMF) and root nodule symbioses, lost their living space. These have led to the loss of soil microbial diversity.

Erosion threatens soil biodiversity by thinning soil layer

Soil thinning is the most significant process of soil erosion. The main on-site impact is the reduction of soil quality caused by the loss of nutrient-rich upper layers of the soil and the reduced water-holding capacity of many eroded soils (Bathrellos et al. 2010). Soil thinning makes the soil layer thinner, and even destroys the original layer that harmoniously arranged soil structure, and the water, fertilizer, gas and thermal conditions and regulating functions are also reduced or disappeared. In addition, because the physical structure and chemical and biological properties of the lower layer that are successively exposed are worse than those of the upper layer, thinning and erosion form a positive feedback system, which further accelerates soil erosion (Wu et al. 2017). It may result in a decrease in arable land. The erosion of soil is one of several natural and human threats to sustained soil productivity, which may become irreversible if not mitigated (Chen et al. 2016, Lal and Moldenhauer, 1987). Soil erosion is aggravated by factors such farming system, soil management practices and rural poverty due to the pressure on soil, which threatens the biodiversity of soil organisms, and thereby threatening man's source of food, livelihood and destroys man's property and investments (Asuoha et al. 2019). Studies have shown that crop yields and ecosystem function are often critically correlated with soil biodiversity (Delgado-Baquerizo et al. 2020, Fan et al. 2020). Therefore, soil erosion will eventually threaten human food and livelihood sources and destroy human property and investment.

Erosion threatens soil biodiversity by polluting soil

In some special cases, erosion on the mining area causes water and land pollution of the downstream. The impact of soil pollution on soil biodiversity is reflected in the following aspects: Soil pollution directly affects the growth and reproduction of soil organisms because of its toxicity and bioaccumulation characteristics, including the production of oxidative stress or the destruction of microbial proteins and DNA (Chen et al. 2020).The ecological condition of soil determines the diversity of microbial population, and the heavy metal pollution in soil will have a great impact on the physiological groups of soil microorganisms (van Elsas et al.1998). Most small invertebrates will change their biodiversity due to soil pollution. Heavy metal pollution will affect the life span of nematode individuals and damage the function of nematode community (Chen et al. 2009). These processes have greatly destroyed the environment on which the species originally depended, and also caused the loss of species diversity. Pollutants in the soil can affect the survival and reproduction of organisms at the back end of the food chain through bioaccumulation (Sarwar et al. 2017).

Efforts and achievements for soil and water conservation

Soil plays such a major part in supporting life on the earth. It is one of the world's most important natural resources. Together with air and water it is the basis for life on the earth. Soils have always been central to human civilization and life and are an integral part of physical and cultural environment. Water and soil are the most valuable assets for man. It has many important functions which are essential for life. Not only does it play the major part in allowing us to feed the world's population, but it also plays a major role in the recycling of air, water, nutrients, and maintaining a number of natural cycles, thereby ensuring that there will be a basis for life in generations to come. Without soil, the world's population neither would nor could survive.

Global soil and water conservation actions

For the sustainable development of human society, it is urgent to carry out the prevention and control of soil and water loss.

European Union countries pay more attention to soil conservation. Germany adopted the Federal Soil Protection Act in 1998, which clearly defined and divided the responsibilities and obligations of federal government departments and individuals to prevent soil pollution. A number of soil conservation measures have also been introduced at the EU level, including the compilation of a list of contaminated sites, the setting of target values and the identification of priority sites for remediation, the Sixth Environmental Action Plan of the European Union (2000-2010) has set up a special soil protection topic.

The United States has also taken active actions in soil and water conservation. On one hand, the United States government has established soil and water conservation agencies from the federal to the local level, the Natural Resources Conservation Service, soil and water conservation zones, and state soil and water conservation agencies. On the other hand, the United States has a number of soil and water conservation laws and acts, such as the Clean Water Act, agricultural law, water pollution control law, coastal zone management law, etc. These specialized soil and water conservation agencies and laws are the institutional and administrative basis for soil conservation in the United States (Huang, 2021). In 1982, after intensive soil and water conservation efforts in the United States, annual soil erosion was reduced to 3 billion tons.

During the long agricultural history in China, the impact of human activities on soil erosion has obvious duality. On the one hand, unreasonable utilization of land, especially the destruction of natural vegetation, aggravated soil erosion. On the other hand, soil erosion was effectively controlled through a series of soil and water conservation measures and ecological restoration and protection projects.

Generally speaking, during the past nearly 70 years, soil and water conservation can be divided into three distinct phases in China (Liu et al. 2018). The first phase is from 1950s to 1980s. In this phase, the priorities were to reduce river sedimentation and ensure river safety, and to meet the basic need of people's living in the eroded regions. Many river bone engineering projects and basic farmland construction, such as terraced fields, dam lands etc., were the main tasks of soil and water conservation. The second phase is from 1980s to the beginning of this century. In this phase with the developing Chinese economy and industry soil and water conservation entered a new period. Besides paying more attention to local economic development and grain production, and improving the land productivity, it is a very obvious characteristic to take small watershed as a unit for the comprehensive allocation of measures of soil and water conservation. The third is since the beginning of this century. With the rapid growth of Chinese economy and society, besides more and more investments to soil and water conservation, more new and scientific concepts and policies were proposed and were also put in practices. It has become the common understandings to achieve the harmony between human and nature, to win both economic and ecological benefits, and the mountains with green vegetation and clear water exactly are "Gold/ Silver" mountains. The Chinese Government proposed GGP (Grain for Green Project, meaning conversion of sloping farmlands to permanent vegetation cover) and other related polices, such as the natural forest protection project and the large scale afforestation project, in soil erosion areas. The GGP has achieved great progresses and benefits, such as in many regions degraded ecology system has got restoration, slope cropland has been covered by trees and grass, bare mountain and hills have been changed by green vegetation cover. As a result, the soil erosion has been controlled and reduced obviously in China.

According to the 2010 erosion survey, the conservation measures implemented on slope land in China covered an area of 943,908 km^2, of which the area covered by engineering measures represented 170,120 km^2 and that covered by biological measures made up 773,788 km^2. In terms of gully control, 58,446 check dams had been built and 928 km^2 of farmland formed. In terms of biological measures, trees had been planted on an area of 522,446 km^2, newly planted grass covered an area of 41,131 km^2, and ecological restoration efforts covered 210,211 km^2. Terrace construction, tree planting, grass planting and ecological restoration accounted for 18.0%, 55.3%, 4.4%, and 22.3% of the total measures used.

The soil erosion in Bulgaria has been acknowledged as an important problem at the end of 18th century when the initial attempts for it control have been made mainly by afforestation around settlements. The beginning of the organized erosion control in the forest fund was set in 1905 when the first Bureau of torrent stabilization and afforestation was established in the town of Kazanlack. Very good results have been achieved in the erosion control activities. Examples exist of very successfully performed complex activities for controlling of torrent watersheds, as well as unique decisions for stabilizing of torrent beds and landslides (Panov, 2000, Zuckov, 2005). By the middle of the 20th century a considerable amount of erosion prevention works has been done on a significant part of the forest fund by Bulgaria– about 130,000 m^3 barrages and 13500 m^2 clay wattles were built, and a total of about 170,000 ha of eroded lands have been afforested (Kostov et al. 1995, Kerenski and Marinov, 1995; Zakov and Marinov, 2003). The period 1952-1980 was characterized by the greatest afforestation (486,000 ha). In this period

326,000 m^3 barrages and thresholds and 337,000 m^2 clay wattles were built. Extensive erosion prevention work has been done in the watersheds of dams. Over 80 big and comprehensive projects of controlling the erosion in the watersheds of dams and many of the torrents have been prepared and realized. The data on the measurements of the sediments that have entered some of the bigger dams show that the siltation is slight in most of them, which is mainly the result of anti-erosion activities. Projects for correcting whit biological (afforestation) and technical facilities of the beds of the torrential rivers - Perperek, Stryama, Dzherman and others have been successfully implemented. A significant reduction in the afforestation rates and in the building of technical facilities was occurred after 1990.

Soil conservation promote biodiversity

Soil and water conservation preserve and promote soil biodiversity in following aspects:

Effects of minimizing disturbance to land

The first principle of soil and water conservation is to minimizing land disturbance by human activities. It will have significant positive effects on the biological soil crusts (BSCs). BSCs are common ground covers and are considered as an indicator of ecological health (Rodriguez-Caballero et al. 2018). BSCs are composed of soil particles entangled with colonies of cyanobacteria, lichens, mosses, algae, micro-fungus, heterotrophic bacteria, and their secreted/precipitated substances within the uppermost millimeters of the soil surface (Belnap and Lange, 2003). Biological crusts improve soil erosion resistance through the direct physical protection of biological mulch and biomass formation and the indirect change of soil properties (Gao et al. 2020). Belnap et al. (2008) found that biological crust coverage is an important biological factor affecting soil stability in wind erosion areas, and the development of biological crusts can explain 77% of the changes in soil stability. Chaudhary et al. (2009) also pointed out that the degree of explanation of the BSCs mulching on soil erosion resistance is three times that of other factors. Its enhancement of soil erosion resistance mainly has the following aspects. First, with the increase of the crust coverage, the soil anti-erosion performance is enhanced. The algae in the biological crust can fix the loose sand through the algae filaments during the growth process to improve the agglomeration of the soil. Its succession greatly increases the surface roughness, and the biological crust increases infiltration and reduces runoff during the rainfall process (Bowker and Belnap, 2008). Xiao et al. (2011) found that artificially propagated moss crusts significantly increased infiltration consequently decreasing overland flow and this increase in infiltration was positively correlated to the soil coverage extent and the start time of runoff process was delayed by moss crusts on soils of the Loess Plateau of China. Second, BSCs can promote the improvement of soil organic matter and soil cohesion. For instance, the biological organic matter content of moss crust and cyanobacteria crust is 4 times that of bare land, and biological crust contains more fine particles (<0.01 mm) and fewer coarse particles (0.05~0.25 mm) than bare land. The particle size of the soil becomes finer, the soil cohesive force is further increased, and it promotes the formation of water-stable aggregates >5 mm, which makes the soil water-stable aggregates changing from small to large, and the soil shear strength is obvious Strengthened, thereby improving the soil erosion resistance (Bu et al. 2015, Yang et al. 2015).

Biocrusts prevent soil erosion, enhance soil water holding capacity, boost soil nutrients, facilitate the establishment and growth of vascular plants, and promote soil biodiversity (Bowker et al. 2014). Biological crusts can significantly increase soil nutrient content through the fixation of photosynthetic organisms and nitrogen-fixing microorganisms on carbon and nitrogen, as well as the capture of atmospheric particles and rainfall nutrients (Belnap and Lange, 2003, Zhao et al. 2010), such as soil organic matter, total nitrogen and available nitrogen, total Phosphorus and available phosphorus, quick-acting potassium; and also significantly enhance the activity of soil enzymes related to carbon, nitrogen, and phosphorus (Ghiloufi et al. 2019, Zhang et al. 2015). Xiao and Veste (2017) found that the number of bacterial colonies in moss crusts was much higher than that of non-crusted soils, which were 2.29 times and 4.41 times that of fixed sandy land and mobile sandy land, and the fungal density was 2.76 times and 6.41 times that of fixed sandy land and mobile sandy land, respectively. The richness of bacteria and fungi was also significantly higher than that of sandy land, which played an important role in the function of the ecosystem by improving soil fertility. Therefore, creating/developing a biocrust in a few millimeters of the soil surface by inoculating of microorganisms on the soil surface for controlling erosion and rehabilitating degraded lands is a green and sustainable conservation practice which has received significant attention recently (Fattahi et al. 2020).

Take cyanobacteria an instance. Due to unique features, cyanobacteria are preferred for the rehabilitation of desert soils. Cyanobacteria grow in nearly all environments because of their low nutritional requirements (Belnap and Lange, 2003). By having evolved to sense and respond to changes in their environment, cyanobacteria have been adopted to high and low temperature, intense irradiation, violent salinity, and a wide range of pH (Ward et al. 2012). Additionally, cyanobacteria enhance the fertility of the soil by assimilating nitrogen and carbon from the air (Maqubela et al. 2009). Moreover, growing in a filamentous shape and secreting plenty of exopolysaccharide substances (EPS) enable cyanobacteria to entangle soil surface particles and stabilize soil matrix against erosion (Wu et al. 2013). Consequently, cyanobacteria facilitate the colonization of other species and enhance biodiversity in inhabitable ecosystems through promoting soil nutrient and stabilizing the soil surface (Patzelt et al. 2014).

Effects of conservation tillage

Human intervention, especially deep farming, will aggravate wind erosion and water erosion. In terms of agricultural management, the less-tillage and no-tillage measures to reduce physical disturbance, as an approach to sustainable farming (Derpsch et al. 2010), not only directly protect soil organisms from threats, but also coordinate soil water, air, and heat, and create better and more living space for soil organisms (de Graaff et al. 2019). De Ruiter et al. (1994) found higher numbers of microarthropods in conventional tillage than in reduced plots. Brennan et al. (2006) have reported that positive effects of reduced tillage on the diversity and abundance of soil fauna. The reduced tillage had positive effects on earthworm biomass levelling out in spring (Moos et al. 2016) and led to a general increase of individual numbers of Collembola with fostering the diversity of Collembola and of functional groups (Coulibaly et al. 2017). Legrand et al. (2018) found tillage had obvious effects on both bacteria and fungi, with species richness and evenness significantly higher in fields under minimum tillage practices than in fields under conventional tillage. The functional diversity of the bacterial communities was also significantly higher in fields under minimum tillage notably that involved in nitrogen cycling (denitrification, respiration). As for soil fungi, lessening physical soil disturbance may promote the growth and colonization of fungi, which has been predicted to particularly impact soil fungi as it causes fungal hyphae to continuously heal or regrow their mycelial network (Verbruggen and Kiers, 2010).

Adl et al. (2006) studied the impact of different no-tillage years on soil biodiversity and found that as the age of no-tillage management increased, the organic matter content, profile stratification and body type diversity increased in the soil. Some biological groups responded more quickly to no-till management, while most had increased diversity within a few years. During the first 8 years without cultivation, the abundance of organisms increased; but only two older fields (8 to 26 years) accumulated abundance and species richness, which were close to the abundance and richness of species in undisturbed sites. Fortunately, the temporal stability (−3%) of no-tillage does not differ significantly from those of conventional tillage indicating that a transition to no-tillage did not affect yield stability (Knapp and van der Heijden, 2018).

Effects of vegetation restoration

Vegetation restoration is the most important and extensive ecological measure of soil and water conservation. Vegetation restoration is the application of ecological principles to the restoration or reconstruction of damaged forests and other ecosystems through the conservation of existing vegetation, the closure of forests or the creation of plantations, shrubs and grasses, help to restore biodiversity and ecosystem functions (Bakker, 2013). The quality and quantity of photosynthetic products affect the content and stability of soil organic carbon (Poirier et al. 2018). Vegetation restoration can significantly increase soil organic carbon storage in degraded soils by increasing litter and root biomass (Deng et al. 2014). As an effective measure for vegetation restoration, conversion of cropland to forest can increase vegetation cover, reduce surface runoff, control soil erosion, reduce river deposition and nutrient loss, and improve ecosystem services fundamentally (Jia et al. 2014).

In the process of vegetation restoration, soil biodiversity has also been improved to a certain extent, and it has participated in the process of restoring or improving the soil's ability to resist erosion (Lehmann et al. 2017, Liu et al. 2020, Rillig et al. 2014). Firstly, plants can directly affect the species, quantity and diversity of soil microorganisms through living roots (Sasse et al. 2017), this is mainly due to rhizosphere secretions and symbiotic microorganisms such as rhizobia and arbuscular mycorrhizal fungi. For instance, the "net bag" formed by roots, cyanobacteria and fungal hyphae interspersed and entangled in the soil, which mainly affected the formation of large aggregates (Lehmann et al. 2017, Wang et al. 2018); root secretion of polysaccharides and amino acids and bacterial metabolites can bind the soil fine particles (Raven, 2018); Wu et al. (2014) also have found that the glomamycin secreted by arbuscular mycorrhizal fungi (AMF) has a "super glue" effect on soil particles to enhance the stability of large aggregates. Secondly, through the input of dead roots and litter, plants provide diverse food resources and decomposable substrates for soil microorganisms, and indirectly affect microbial community structure and diversity through more niche differentiation, for example, vegetation produces hard-to-decompose litter (such as higher carbon-nitrogen ratio and lignin) , which promotes the development of fungal-dominated microbial communities, however, unstable litter (low C/N ratio, lignin and polyphenols) stimulated the development of bacterial-dominated soil food web (Wu et al. 2018). Thirdly, plants affect the structure and community of soil microorganisms by changing soil moisture and temperature (Yao et al. 2019). Vegetation can reduce erosion to a certain extent, and a number of experiments have explored the erosion-reducing effects of above-ground and underground roots, such as vegetation coverage reduces splash erosion and rill erosion by reducing the kinetic energy of raindrops, the resistance of roots to concentrated flow erosion can be significantly increased by soil reinforcement (Wang et al. 2014). The effects of above-ground characteristics of plants on erosion dynamics are mainly reflected in direct interception of rainfall, reduction of raindrop kinetic energy and increase of slope roughness to reduce runoff velocity. The effects of plants on soil erodibility are mainly due to the binding effects of roots and the bonding effects of plants on soil. Vegetation restoration reduces a series of negative impacts of soil erosion on soil biodiversity and promotes the restoration of biodiversity. In summary, a large number of studies have reported the effect of vegetation on the stability of aggregates. It is worth noting that recent studies and reviews have emphasized that a better understanding of the effect of vegetation on soil structure should be shifted from a method based on species comparison to a method based on plant species.

Effects of engineering measures

Soil and water conservation engineering measures such as terraced field, check dam can effectively increase soil organic matter content, and promote soil biodiversity. Terraces change the terrain by artificial preparation, effectively intercepting the nutrient loss carried away by surface runoff, avoiding the nutrient loss carried away by soil erosion, and adding the organic matter supplemented by artificial fertilization, thus increasing the organic matter content. The natural litters enriched on the ground surface, through decay fermentation and various chemical reactions under the action of microorganisms, produce different metabolites, so as to continuously supplement the organic matter in the soil and make the organic matter content in the soil relatively high. It is conducive to building a suitable habitat for soil organisms.

Effect of forest fires on land restoration

To promote biodiversity, soil protection in burned areas must be framed within a broader watershed restoration context applying functional and landscape criteria. We understand by restoration the set of actions allow to reestablish the composition, structure and functionality of the ecosystem. It is therefore a complex multidisciplinary process with a strong initial emphasis on protection aspects, sustained with medium-long-term actions that will allow the area affected by a fire to recover its natural dynamics, self-regulated and integrated in the biophysical and climatic context of the region. In turn, a broader context would include the increasingly strong tendency to consider and enhance the ecosystems services provided by the forest in terms of maintaining biodiversity, landscape quality, regulation of the hydrological cycle, regulation of greenhouse gases and protecting the soil (Lindenmayer et al. 2000). Traditionally, soil protection in burned areas has followed methodological schemes based on the principles of forest hydrological correction. These restoration techniques include classical soil conservation approaches initially developed in the United States and extended with modifications and adaptations to the rest of the world, including Mediterranean environments (Bennet, 1965; Fournier, 1975; Magister, 1991; Tragsa, 2003). However, current soil conservation schemes are expanding to include new perspectives in a broad context of sustainable use of terrestrial ecosystems. This section of the article discusses the basic approaches for soil conservation in burned areas, including the options that the emerging field of bioengineering provides for soil restoration and stabilization, protection against different types of erosion, and maintenance of the landscape quality and in terms of biodiversity and aesthetic values (Rubio, 2009a). The Department of Soil Degradation and Conservation of the Centre of Desertification Research-CIDE (Valencia, Spain) actively contributed to the conceptual development of these technologies and has collaborated in their application in areas affected by forest fires in diverse Spanish regions.

The traditional approaches for soil conservation and forest restoration in burned areas are framed in schemes of forest hydrological correction which include a series of sub-objectives such as the maintenance of productive capacity of the soil, the reduction of the intensity and effects of the catastrophic floods, protection of reservoirs against sedimentation, improvement of the hydrological regime and the safeguard of water quality. Within the specific measures of soil conservation, there are the preventive or cultural practices and the defense measures. The former, through appropriate soil management techniques, seeks to improve soil properties (structure, organic matter content, infiltration), adequate for plant cover management, and hydrological management and control (runoff, permeability, drainage, erosion). Some of the preventive measures are grazing control, stubble treatments, organic fertilization, phytosanitary control, mechanized surface work, mulching and maintenance of plant covers.

Traditional defense measures can be very varied depending on the enormous complexity and diversity of Mediterranean and other forest environments. The complicated topography, the variety of soil types and the climatic conditions open a wide range of application of highly varied techniques that, however, can be broadly grouped into five types. These groups include level cultivation, strip cultivation, terraces, bench-terraces, and drainage channels. The main objective of these techniques is to decrease the volume of runoff and to control soil erosion. In the USA there is old and extensive experience in their use as techniques for the rehabilitation of burned basins (Heede, 1960; Baley and Copeland, 1961; Noble, 1965). These techniques are widely described in traditional soil conservation manuals and are complemented by techniques for restoring basins and river ecosystems and for correcting torrential channels (Tragsa, 2003). More recently, some of these actions have been the subject of controversies and the consideration of new approaches with less impact or with more adjusted intervention on the burned ecosystem is increasing. These new methods include principles of minimal intervention and approaches to help ecological succession, which is reinforced with very careful interventions in terms of succession. Once the critical initial post-fire stage has been overcome, silvicultural management is also oriented in the direction of ecological succession. It is pursued, with adequate silvicultural treatments (thinning, pruning, clearing), that the plant cover follows the modeling and structure of the natural masses and deviation trends such as augmented density of tree foot and competition phenomena are avoided.

Bioengineering techniques applied to soil conservation offer interesting possibilities in the restoration of burned areas. The increasing demand for the maintenance of soil biodiversity, the regulation of the carbon cycle, the maintenance of landscape quality, the regulation of the hydrological cycle and the prevention of soil degradation, require new approaches with broad criteria of multifunctionality and multidisciplinarity (Rubio, 2009b). It should be achieved without losing the essential aspects of the previous-traditional schemes, including the protection of the soil, the maintenance of its fertility and the soil-water interaction. Bioengineering is part of the broader concept of ecotechnology or ecological engineering that has been the subject of extensive conceptual discussions and numerous definitions. One of the most synthetic definition considers that ecotechnology is simply “the management of nature” (Odum, 1971). For Straškraba (1993) it consists of “the use of technological means for the management of ecosystems, based on a deep ecological knowledge, to minimize the cost of interventions and their impact on the natural environment”. A third definition considers it to be “the proactive design of sustainable ecosystems, which integrate human society into its natural environment, for the benefit of both” (Mitsch, 1996). In this context, bioengineering applied to soil conservation can be considered as the set of techniques that, on one hand, are oriented to the restoration and creation of terrestrial ecosystems and, on the other hand, to their management. and protection. Traditionally, the stabilizing role of vegetation on slopes has been generally accepted by empirical experience. Although reforestation with various plant species for erosion control has centuries of history, research on the effects of vegetation on soil stabilization in unstable areas only began in the 1960s. Its protective effects can be grouped into hydrological, mechanical and biological effects. Among the hydrological effects would be the interception of rain, which on one hand reduces the impact of rainfall and on the other hand increases infiltration and subsurface flows. The absorption of water by the roots, would also give rise to a positive effect by reducing the water pressure in the pore volume. As a negative hydrological effect, there would be an increase in evapotranspiration, with the subsequent result of decreasing soil moisture. Among the mechanical effects, there would be the reinforcement of the soil by the roots, which increases the resistance to shear stress. This mechanical reinforcement varies widely depending on the characteristics of the soil profile and even, in certain circumstances, includes favoring the formation of sliding planes. In this sense, the weight overload represented by the vegetation cover on a slope can increase the shear effects and the water pressure in the pores. This overload effect can be increased by the action of the wind. However, and as long as the angle of the slope is not excessive, the overload due to the vegetation increases its stability. The biological aspects, especially those related to soil biodiversity, have already been discussed in this article.

Bioengineering applied to soil restoration and conservation includes methodological aspects and principles from traditional and soil conservation techniques. It also incorporates various civil engineering techniques such as the use of gabions, geotextiles or meshes. They are mainly used in the initial phases of intervention but coordinated with the progressive incorporation and covering by biotic elements. Its novelty lies in the priority given to the use of plant materials and the drastic reduction in the use of hard elements. Ecological coherence and enhancing soil biodiversity is pursued in connection with the biophysical and climatic possibilities and limitations of the area. Basically, it involves the incorporation of live plant species for the performance of engineering functions under a design adapted to ecological succession.

The functions of vegetation in bioengineering can be grouped into four large groups (Norris et al. 2008): *a)* Soil protection techniques, which quickly protect it, due to the effect of covering its surface against surface erosion and degradation processes. These techniques improve water retention and increase the biological activity of the soil. *b)* Soil stabilization techniques, which reduce or eliminate the negative mechanical effects derived from the soil mass itself. They stabilize unstable slopes by penetrating roots, lowering air pressure in the pores, and increasing drainage. *c)* Combined reconstruction techniques, which stabilize unstable slopes or riverbanks through the combined action of living plants and inert materials (stones, concrete, gabions, wood, steel and geotextiles). *d)* Supplementary stabilization techniques, which use seedlings and plantations to ensure the transition between the initial phases of action and the completion of the intervention.

In the aspects related to the conservation-restoration interventions of burned areas, besides the previous classification, specific techniques that are in continuous evolution are included.

These include the use of burnt tree trunks and branches barriers, fajinas, aerial seedlings, the use of organic blankets, biocylinders and blocks of degradable materials. The use of plant species in bioengineering techniques involves numerous factors to be considered. These include, among others, the choice of species and where to get them; the preparation of the land and machinery to be used; the specific actions of sowing and planting; its subsequent maintenance and the necessary complementary work. The choice of plant species is a particularly critical aspect because, to a large extent, the success of the performance will depend on it. The situation is more favorable in humid climates than in climates such as semi-arid or dry-sub-humid, in which the range of options and the chances of survival are smaller.

There is an extensive botanical bibliography on the bioclimatic species best adapted to each area. In general, the species selection criteria include: ecological factors (phytogeographic, climatic, lithological and edaphic and physiographic), economic factors (availability, origin, cost, transportation) and other factors such as the availability of labor or the knowledge about plant succession. Regarding the selection of the specific techniques of action in each case, the available information is gradually increasing. An initiative in this sense was developed within the framework of the EU project. "Eco-engineering and conservation of slopes for long term protection from erosion, landslides and storms-ECOSLOPES" (QLRT-2000-00.289) in which a review and evaluation of the available principles and techniques of bioengineering applied to soil restoration and conservation was carried out. In this context, Andreu et al. (2008) prepared a catalogue of techniques indicating in each of them their purpose, use, methodology, materials used and their advantages and disadvantages.

Bioengineering techniques applied to soil conservation and restoration-protection of burned areas provide a more environmentally adapted approach that is consistent with plant succession, with the enhancing of soil biodiversity and with the preservation of the ecological functions of the affected areas. This greater environmental coherence favors greater long-term efficiency on post-fire protection evolution. They also provide economic advantages for their lower cost, for improving the landscape and visual quality of the interventions and for the increasing the range of ecosystems services of the area. On the other hand, they pose a series of drawbacks and disadvantages such as the need for specialized labor, the lack of knowledge and availability of the appropriate plant species and also their availability at a given time. Despite these drawbacks, bioengineering techniques are increasingly perceived as an emerging and promising approach that is also showing rapid growth and acceptance.

Effects of salinized land improvement and polluted land restoration

Salinized land is one of the key points of wind erosion control. Many studies have found that planting proper halophytes can not only significantly improve saline-alkali soil but also increase the content of organic matter and total nitrogen in soil (Xia et al. 2019, Yang et al. 2021, Zhongping et al. 2012). For instance, Yang et al. (2021) found that Tamarix chinensis and salt-resistant grass exerted a definite absorption of the upper soil salt. And the accumulation of salt carried by upward migration soil moisture was also inhibited due to the surface layer of soil was covered by litter and low herbaceous plants. The fallen leaves from Tamarix chinensis and death grasses are decomposed, and then humus formed easily, and the interpenetrating effects of the Tamarix chinensis root and grass systems promote the formation of good physicochemical properties of soil, and the available nutrient contents increased finally (Nyakatawa et al. 2012). Notably, the considerable differences of the available nutrient contents between the various vegetation patterns should be noticed (Yang et al. 2021). Anyway, it provides a better environment for soil organisms to live in.

As we discussed above, soil contamination by heavy metals is one of the hazards caused by erosion. Over the years, various in-situ remediation technologies have been developed to alleviate or restore soil contaminated by heavy metals, such as surface capping, soil washing, solidification, chemical immobilization and biological measures (Liu et al. 2018). However, phytochemistry, referring to the use of green plants and associated microorganisms to minimize the toxic effects of potential contaminant in the environment, has been widely recognized as a cost-effective method of remediating metal-contaminated soil (Sarwar et al. 2017). Soil microorganisms associated with plants may influence heavy metal availability and uptake by plants in the rhizosphere. Chen et al. 2020 have defined that microorganisms that have the resistance or tolerance to heavy metals are classified as "resistant", like *Noviherbaspirillum*, and these microorganisms, defined as "actor", can mitigate the heavy metal pressure of the environment by passivation or enriching heavy metals in vivo to improve the environment (Marrero et al. 2015), such as *Hydrogenispora*, *Mobilitalea* and *Saraina*, which is conducive to the survival of other microorganisms (Chen et al. 2020). There is a significant correlation between actors and sensitive microorganisms, that is, actors may modify environmental pressure, thereby providing favorable conditions for the survival of sensitive microorganisms and even other soil organisms. Another example is plant growth promoting bacteria (PGPR). PGPR, defined as symbiotic bacteria and free living rhizobacteria (Liao et al. 2003), can assist plants to remediate soils contaminated by heavy metals (Seth, 2012). Farwell et al. (2007) reported that the Cu toxicity to *Brassica napus* was alleviated with inoculation of *Pseudomonas puteda*. Inoculating proper microbial inoculum may help plant species to remediate soils polluted by heavy metals effectively. In addition, fungi can associate plant roots to absorb and transport plant nutrients such as N, P, K, Zn, Ni and Cu availability through extensive hyphal network (Zaidi et al. 2006). These fungal associations can also modify the chemical composition of root exudates and soil pH and hence heavy metal bioavailability in the soil. For instance, Chen et al. (2003) demonstrated that arbuscular mycorrhizal fungi (AMF) could enhanced Zn uptake and accumulation of plants. These can provide a better environment for phytostabilization plants to establish.

Taken together, during such restoration processes, whether the microbial population inoculated externally, or the improvement of soil habitat and fertility brought by this method, can greatly promote the survival of belowground organisms, which undoubtedly increases the diversity and ecological stability of soil organisms.

Cases report

Two ecological restoration case studies in China are summarized.

Ecological restoration achievements in Shendong mining area of China

The natural environment of Shendong mining area

The Shendong mining area is located in the transition region between the Loess Plateau and the Mu Us Desert and on the border area between the Inner Mongolia and Shaanxi province. The annual average temperature is 7.3 ^{o}C. The average annual precipitation is 362 mm, concentrating in July and August, and in the form of a rainstorm. The annual evaporation is 2300 mm, which is 6-7 times of the precipitation. The annual average wind speed is 3.2 m/s, the maximum is 24m/s, there are 70 days annual that the wind speed is over 5m/s--the threshold wind velocity of sand-moving.

The original environment of the mining area is very fragile, the intensity of water erosion is as high as 9000-13000 t/(km^2·a), the mobile dunes are widespread and the wind erosion land covers 70% of the total area. The aeolian sandy and loess have low organic matter and is poor of N, P, and K. The main vegetation types are grass and deciduous broad-leaved shrubs; the vegetation coverage is 3-11%. The representative phytobiocoenose is *Artemisia ordosica* Krasch. group, forming a monodominant community. The main companion plants are annual herbs, such as *Setaria villosa* P. *Beauv*., *Tribulus terrestris*, *Chenopodium aristatum*, etc.

Ecological protection and erosion control

Since the coal exploitation in 1998, efforts have been taken to build ecological mining areas and green mines. The cumulative area that has been taken ecological restoration or erosion control measures has reached to 384 km^2. The vegetation coverage rate has increased from 3%-11% to 64% the above. The contradiction between ecological protection and large-scale resource exploitation in ecologically fragile areas has been well resolved, and the original fragile ecological environment in mining areas has rehabilitated to a positive succession stage.

Combining the natural ecological characteristics and influencing factors of Shendong mining area, corresponding prevention and control technologies are adopted. Prior to coal exploiting, the functions of the surface ecosystem had been continuously enhanced through measures such as wind-proof and sand-fixing, soil and water conservation, and 103 km^2 of mobile sandy land was controlled to improve the resistance to mining disturbances on the ecological environment, and to avoid ecological degradation caused by large-scale mining. During mining, the super large working face underground is adopted to reduce the fragmentation of the subsidence ground. The underground storage and purification of mine water, the replacement of underground coal gangue, reduced the amount of external drainage and gangue, and minimized the impact of mining on the surface ecological environment. After mining, the subsidence area was enclosed and protected, microbial measures were used to promote natural restoration, and to increase vegetation coverage. Over the past 30 years, a total of 130 km^2 of forest land and 3 km^2 of cultivated land have been planted or reclaimed in the mining area. 2.55 million large-fruited sea-buckthorn and 0.9 million fruit trees had been planted, ecological resources have brought economic income.

Spatially, the ecological rehabilitation of the mining area is divided into three function circles: the outer protection circle, the middle evergreen circle, and the core beautification circle.

Peripheral protection circle: vegetation restoration was implemented to control the mobile and semi-fixed dune and prevent railway road sand damage. An herb-based forest combination with grass and shrubs were structured. Accumulatively, 221 km^2 of mobile sandy land had been fixed, reduced 654,700 tons of sand transported by wind enter to the river every year.

Middle evergreen circle: in view of the exposed mountain areas with severe soil erosion around the mine, a mixed forest of coniferous and arbor-shrubs have been constructed, with an afforestation area of 19 km^2. It not only controls the soil erosion in the mountains, but also creates an evergreen landscape.

Core beautification circle: for the barren production and living environment in the early stage of mining, garden landscape and ecological town construction were implemented. The green land rate has reached more than 40%, and the per capita public green area has exceeded 10 m^2.

The effect of ecological rehabilitation

384 km^2 land in the Shendong mining area has been implemented ecological management and construction, where reversed the fragile ecological degradation and built an oasis in the desert area. The monitoring results show that the fragile ecological environment in the mining area has achieved to positive succession. The vegetation coverage rate in the mining area has increased from 3% to more than 64%. The plant species has successfully upgraded from an herbaceous community dominated by *Artemisia sphaerocephala* to a shrub community dominated by Sea buckthorn. There are more than 100 types of plants, and the vegetation configuration is more scientific. The erosion modulus was reduced from 3322.53 t/(km^2·a) in 2005 to 852.17 t/(km^2·a) in 2018. The area of desert land was reduced from 123.29 km^2 to 22.86 km^2. Compared to the background, the content of soil organic matter increased by 32.48% on average, total nitrogen content increased by 36.36%, available phosphorus content increased by 397.46%, and available potassium content increased by 44.57%.

Soil and water conservation in Tangbei small watershed, Jiangxi province

The Tangbei small watershed is located in Xingguo County, Jiangxi Province in southern China. Vegetation was destructed on account of agricultural production and cutting of fuelwood in the watershed before 1980, causing serious soil erosion, as well as the decline of land productivity. Comprehensive management was implemented from 1980 to 1992. After nearly 30 years of maintenance and natural vegetation restoration, soil erosion was controlled, farmers' production and living conditions were improved, and the ecosystem was restored. The management experience of Tangbei small watershed has been widely promoted in the red soil region of southern China with similar conditions.

The outline of Tangbei small watershed

The area of Tangbei small watershed is 16.38 km^2, of which, the mountainous area is 11.65 km^2. The survey in 1980 showed that the area of soil erosion was 11.53 km^2, accounting for 99% of the mountain area. Vegetation coverage over the hills was less than 15%, erosion modulus was up to 13500 t/(km^2.a). In 1980, before the erosion control was carried out, there were 3193 persons living in the Tangbei small watershed. The watershed had been plagued by drought and floods for years, with low land productivity, low agricultural production capacity, and poor life of the farmers. The per capita grain output was 196.3 kg, the net income per capita was 40.6 RMB¥ (about 6.09 US$).

The watershed belongs to subtropical monsoon humid climate zone, with average temperature of 18.9°C and rainfall of 1538.7 mm. The soil is mainly red soil developed from weathered granite, and the landform is low hills with partly river valley flat land, the altitude is between 160 m to 340 m, and the ground slope is between 8° and 25°. The vegetation in the watershed is mainly artificial coniferous forest, and the main tree species include *Pinus massoniana*,

Liquidambar formosana, *Schima superba*, *Cunninghamia lanceolata*, *Navel orange*, *Camellia oleifera*, *Castanopsis eyrei*, *Photinia rubra*, *Cinnamomum camphora* and so on.
The soil and water conservation measures

The comprehensive control measures had been implemented in the small watershed began in 1980, and by 1988, all the planning measures had been completed and technical indicators had reached to the planned requirements. Since then, some of the small-scale measures was taken.

The following five kinds of measures have been implemented in the process:

a. Energy adjustment measures: Promote the construction of wood-saving stoves, replace firewood stove with coal stoves, and construct biogas ponds in the whole watershed to reduce the cutting of vegetation;

b. Closing and reinforcement planting measures: It mainly relies on the natural restoration of the ecology system by closing, and appropriate reinforcement planting, on the light erosion land;

c. Storage and drainage water measures: Build small storage and drainage projects on the slope to increase the use of water resources;

d. Artificial forest and grass measures: In the areas of moderate or severe soil erosion, after digging horizontal ditches, implement fishing scale pits and other land preparation measures, plant soil conservation forest ---- combination of grasses, shrubs, and trees;

e. Economic development measures: In gentle slope areas with good soil quality, plant a large amount of economic forest and fruit trees, conserving soil and water and having higher income. At the same time, rural roads in the watershed were reconstructed and expanded. A new pattern is emerged to protect mountains, rivers, forests, farmlands, and roads in a coordinated way.

By the end of 1988, 650 hm^2 of soil and water conservation forest was planted, 325.5 hm^2 of soil and water conservation forest was reinforcement planted. 18 check dams and 7 ponds were built, 3 erosion gullies were treated, and 560 m of riverbanks were built. At the same time, 2 small reservoirs were renovated, 555 wood-saving stoves, 480 coal stoves, and 208 biogas ponds, 4.5 km of new roads were built, 8 km of roads were expanded, 3 stone arch bridges, and 58 culverts were built. In total, 975.5 hm2 of comprehensive treatment was completed, 84.5% of the soil erosion area was treated.

After 1988, post-management and follow-up treatment were continued. To the end of 1992, more of 13.3 hm^2 of economic forest was planted, more of 164.2 hm^2 was closed. The total treatment area reached 1153.4 hm^2 and the treatment rate reached 99%. By the end of 1992, a total of 1.87 million yuan, 611 thousand man-days were invested in the treatment of this small watershed, 1.42 million cube meters of earth and rock were completed. The vegetation coverage rate rose from 15% to 72.8%, and the soil erosion quantity decreased by 82%.

Effects of the erosion control

Through comprehensive treatment and follow-up management of soil and water conservation, the natural landscape and ecological environment of the whole watershed have been significantly improved, the ecological, social, and economic benefits are prominent, and the land in the watershed has basically realized sustainable use.

The degraded ecosystem has been restored, and the natural ecological has entered to a virtuous cycle stage. The forest structure in the watershed tends from a pure forest of *Pinus massoniana* to a good forest ecological structure that combines trees, shrubs and grasses, with mixed coniferous and broad-leaved forests. Plant and animal species are increasing, wild animals such as pheasants and rabbits, which had not been seen for many years, are inhabiting in the forests. Mosses, lichens, mushrooms, and other low plants are multiplying again. Springs that had been depleted for many years are gushing again.

The organic matter and fertility of the soil in the watershed have been continuously improved. Monitoring data from 1980 to 1919 shows that, the water storage efficiency in the watershed has increased by 37.6%, and the annual soil erosion reduced 42,300 m^3. The average thickness of the woodland litter layer is 0.5 to 1.5 cm in 2019, the organic matter of the topsoil has increased by an average of 8 times (from 0.09% to 0.83%), the nitrogen content has increased by 11 times, and the soil microbial biomass has increased by more than 10 times.

Conclusions

The importance of soil biodiversity and the ecosystem services provided to global ecosystems have long been widely recognized in the scientific community. Research on soil biological potential value and its status in the ecological system is a very difficult task, has the very strong feasibility study, to explore the mechanism of various factors on the pressure of biodiversity, can to a certain extent quantitatively comprehensive evaluation of the potential threat of soil biodiversity in the future, to avoid the deterioration of ecosystem and the negative effect on the human society. This paper reviews global soil erosion and the progress of soil and water conservation, the effects of soil erosion on soil biodiversity are summarized, and the stress mechanism of soil erosion on soil biodiversity and the mechanism of soil and water conservation promoting soil biodiversity are expounded, however, due to the wide range of soil organisms involved, there is no detailed and specific analysis of the threats faced by each soil biological group. Two examples are given to show the role of soil and water conservation in the restoration and promotion of soil biodiversity. To foreground the study of edaphic biodiversity from the broad interdisciplinary perspective, in order to promote the advancement of its area of knowledge. In the future, it is necessary to further deepen the research on the impact mechanism of environmental change on soil biodiversity, quantify the change trend of soil biodiversity threat, deepen the awareness of interdisciplinary research, so as to provide theoretical and technical support for soil biodiversity conservation.

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Interdisciplinary perspectives from soil physical point of view

Chapter 17

Physical properties of volcanic ash soils related to soil biodiversity: effects of pasture improvement managements

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Introduction

Soil structure is a physical property that can be used to assess the state of pasture systems and their biodiversity as a consequence of agricultural management. Volcanic ash soils are characterized by a high capacity to store and conduct water, air and nutrients through an interconnected pore system associated with their structure, which in turn allows them to host a great diversity of organisms. This dynamic behavior can be conserved over time through pasture improvement management strategies (PIMs). The objective of this chapter is to explore the following question: Can we expect pasture improvement strategies that conserve soil structure to improve pasture herbage mass accumulation and maintain soil biodiversity? To answer this question, we used the results from studies carried out from 2010 to 2016 on a volcanic ash soil in southern Chile, where we analyzed how pasture management affected soil structure and pore functions over time.

Challenging scenarios

The global warning scenario is challenging the current traditional management of livestock production due to increases in the probability of extreme drought events (AghaKouchak et al. 2015), heat waves (Seneviratne et al. 2012) and temperature variability (Zhang et al. 2018). When these factors interact with agroecosystems with low species diversity characterized by low tolerance and resilience to abiotic stresses (Patzek, 2008), which depend upon an intensive use of natural resources (Millennium Ecosystem Assessment, 2003), i.e., soil (Zúñiga et al. 2015) and water (Vörösmarty et al. 2005), the result is pastures that are unstable and unsustainable.

There are about 1.3 million ha of pastures in the south of Chile, 91% of these pastures are naturalized, i.e., pastures composed of introduced forage species that have persisted under the existing conditions of climate, soil, and management for long periods (Allen et al. 2011), with 48% of them receiving no management to compensate for soil nutrient deficiencies, leading to their current signs of degradation (Flores et al. 2017). Pasture systems provide multiple ecosystem services, e.g., beef, milk, nitrogen fixation, among others (Teague, 2018). In grazing systems, pasture provide an essential provisioning service by supplying forage for domestic herbivores (Keim et al. 2015). In this context, pasture improvement management (PIMs) is a research subject of great relevance for grazing systems (Zúñiga et al. 2015).

Volcanic ash soils (Andosols) in southern Chile are characterized by < 25.0% of soil organic carbon (SOC), a bulk density ≤ 0.9 Mg m^{-3} (Dörner et al. 2009), a highly reactive colloidal fraction dominated by short-range-order minerals (CIREN, 2003; Clunes and Pinochet, 2021), a high phosphorus retention (Valle et al. 2015) and a great swelling/shrinkage capacity (Dörner et al. 2012). Furthermore, the hydraulic properties of these soils are affected by the complex nature of their porous system due to the presence of a well-defined (macro and micro) aggregation (Dörner et al. 2015), which provides them with a remarkable drainage capacity, aeration and root growth (Dörner and Dec, 2007). Andosols have thus been classified as having good soil quality (Valle et al. 2018).

Soil structure is a fundamental soil physical property for assessing the condition of agroecosystems e.g., after changes in agricultural management or land use, among others. Its role in soil functions, such as water storage and filtration (Dörner et al. 2015), carbon and nutrient storage and cycling (Clunes et al. 2021), habitat and maintenance of soil biodiversity (Zechmeister-Boltenstern et al. 2015), becomes relevant when the sustainability of agroecosystems over time is threatened by the intensification of agricultural management. In this context, studying the effects of PIMs on soil biodiversity with a soil function approach highlights the fact that the functional diversity of biota drives specific soil processes more than community characteristics (Vogel et al. 2018).

We therefore explore how different PIMs modify soil structure dependent properties and their relationships with conditions that maintain or stimulate biodiversity development. Thus, the aim of this review is to explore the following question: Can we expect pasture improvement strategies that conserve soil structure to improve pasture herbage mass accumulation and maintain soil biodiversity? This question was answered based on results derived from a sequence of PIMs field studies conducted in Andosol soils in southern Chile.

Materials and methods

Soil and climatic conditions

The studies were conducted at the Universidad Austral de Chile's experimental field station (Estación Experimental Agropecuaria Austral (EEAA), 39°46′ S, 73°13′ W, 12 m a.s.l.) located in Valdivia, southern Chile. The average annual temperature is 12 °C with a mean yearly rainfall of 2,440 mm between 1901 and 2005 (González-Reyes and Muñoz, 2013). The soil corresponds to a soil derived from volcanic ashes, classified as Silandic Petroduric Andosol (Salazar et al. 2005). The topography of this soil series is normally complex, with dominant slopes from 3% to 8%. Bravo et al. (2020) described the soil horizon distribution based on a morphological description of a soil profile with a depth of 2 m.

Experiment establishment

Two experiments were carried out in the same soil, exposed to the same previous pasture management conditions. Both included an array of novel approaches for sustainable pasture production in relation to soil structure and plant species biodiversity. Before the studies, the site was subjected to rotational grazing for over 30 years with no fertilizer input nor grazing criterion applied. Therefore, at the beginning of the experiments the pasture was degraded to poor conditions with a dominance of low-quality plant species (yields < 4 Mg DM ha^{-1} $year^{-1}$, Zúñiga et al. 2015).

Experiment 1 (Exp-1) was performed between 2010-2013 (Zúñiga et al. 2015) with 4 treatments: NFNP1: non-fertilized naturalized pasture; FNP1: fertilized naturalized pasture; LTCP1: cultivated pasture established after seedbed preparation; DCP1: diverse cultivated pasture established after seedbed preparation.

Experiment 2 (Exp-2) was conducted between 2013-2016 with five treatments (Ordóñez et al. 2018): DP: pasture established with direct drilling (zero tillage); DDP: diverse pasture established with direct drilling (zero tillage); NFNP2: non-fertilized naturalized pasture; FNP2: fertilized naturalized pasture; LTCP2: cultivated pasture established after traditional tillage and seedbed preparation.

In both experiments, a non-selective herbicide (glyphosate) was applied over the resident pasture and the pastures sown (LTCP1, LTCP2, DCP, DP and DDP). Before sowing LTCP1 and DCP1 for Exp-1 and LTCP2 for Exp-2, the seedbed was prepared with traditional tillage, including ploughing, harrowing and soil compaction (with a roller compactor). This preparation destroyed the soil aggregates and structure within the first 20 cm of soil depth. On the other hand, direct drill or zero tillage maintained the soil aggregates and structure, sustaining the soil functionality for the establishment of plantlets.

In both experiments, there were two types of evaluated variables: pasture and soil parameters. The pasture included: herbage mass, growth rates and botanical composition. The evaluated soil parameters were: bulk density (BD), soil porosity, air capacity (AC), plant available water (PAW), air permeability (Ka) and pore continuity indexes (C_2), pre-compression stress, soil organic carbon (SOC) and nutrient status.

Detailed descriptions of the pasture establishment, as well as the methodology used to measure each variable can be reviewed in Dörner et al. (2013), Zúñiga et al. (2015), Flores et al. (2017), Ordóñez et al. (2018), Haas et al. (2018) and Descalzi et al. (2020).

Statistical analyses

The normality and homogeneity of variance were evaluated using the Shapiro-Wilk and Levene tests, respectively ($p \leq 0.05$) (Zúñiga et al. 2015, Ordóñez et al. 2018, Córdova, 2019). The boxplot indicates the data distribution over time, where the top of the box represents the 75th percentile, the bottom the 25th percentile and the line in the middle the 50th percentile. The whiskers represent the largest and smallest values within 1.5 times of the interquartile range. Outside values indicate outliers. All of the mentioned analyses were conducted using RStudio 1.3.1093 (R x64 4.0.3).

Results and discussion

Pasture diversity in relation to physical parameters

The herbage mass production for both experiments showed a similar trend (*Table 1*); all pastures with PIMs increased their annual herbage mass accumulation to a range of 7 – 10 Mg DM ha^{-1} year^{-1}, while NFNP2 reached 3 – 5 Mg DM ha^{-1} year^{-1}. In both experiments, the sown species remained dominate in their respective pastures (*Table 2*). After the first year of evaluation, NFNP1 was dominated by broad leaf species (BLS), such as *Rumex acetosella* L. and *Leontodon nudicaulis* L. (71% of the total herbage mass). A similar trend was shown in NFNP2, which had a dominance of 29% of BLS (i.e., L. *nudicaulis, Taraxacum officinale*) and 47% grasses such as *A. odoratum* and *A. capillaris*. For the following three years of evaluation, NFNP1 was mainly composed of 59.2% *A. capillaris*, 20.9% *H. lanatus*, 10.6% *A. odoratum* and 4.8% *T. officinale* (Flores et al. 2017). During three years of evaluation, a study exhibited that dominant species in a naturalized pasture were mainly composed of 59.2% *Agrostis capillaris* L., 20.9% *Holcus lanatus* L., 10.6% *Anthoxanthum odoratum* L., 0.4% *Bromus valdivianus Phil*., 0.3% *Schedonorus arundinaceus* (Schreb.) Dumort., 0.3% *Lolium perenne* L., 1.3% *Trifolium repens* L., 0.2% *Lotus uliginosus* Schkur., 4.8% *Taraxacum officinale* Weber, 1.9% *Leontodon nudicaulis* L. and 0.1% *Rumex acetosella* L. (Flores et al. 2017). After three years, inputs of nitrogen, phosphate, potassium and lime were beneficial for the long-term species persistence and productivity, showed an increment of high fertilized response grasses, such as *L. perenne*, *B. valdivianus* and *H. lanatus*. Besides, the percentage of legumes increased by about 70% (Flores et al. 2017).

	Treatments	**Year 1**	**Year 2**
		(DM kg ha^{-1} yr.$^{-1}$)	
Experiment 1	DCP	7,880 ± 236	10,833 ± 105
	LTCP1	9,453 ± 369	10,921 ± 47
	NFNP1	4,190 ± 79	5,854 ± 105
	FNP1	8,415 ± 108	8,443 ± 84
Experiment 2	DP	7,302 ± 663	8,938 ± 747
	DDP	6,729 ± 498	9,217 ± 328
	NFNP2	4,069 ± 234	3,401 ± 226
	FNP2	8,089 ± 512	9,384± 531
	CP2	7,499 ± 618	7,599 ± 686

Experiment 1 data from Flores et al. 2017; *experiment 2* data from Descalzi et al. 2019.

Exp. 1: DCP1: Diverse cultivated pasture established after seedbed preparation. LTCP1: cultivated pasture established after seedbed preparation; NFNP1: Non-fertilized naturalized pasture; FNP1: Fertilized naturalized pasture; Exp 2: DP: Pasture established with direct drilling (zero tillage); DDP: Diverse pasture established with direct drilling (zero tillage); NFNP2: Non-fertilized naturalized pasture; FNP2: Fertilized naturalized pasture; CP2: Cultivated pasture established after traditional tillage and seedbed preparation.

Table 1. Annual herbage mass production (based on dry matter) for the first two years for the treatments of each experiment.

This gives the chance to minimize the input and to rely on legume fixed nitrogen rather than fertilizer nitrogen. These results demonstrate that without soil fertility stress, the ecological succession of pasture species could occur (Gastó et al. 1993), reflected by the colonization of faster growing species. These types of species confer the long-term stability of the pasture, along with an increase in the annual herbage mass production (Descalzi et al. 2020). Besides, after three years the percentage of legumes increased by about 70% in FNP1 (Flores et al. 2017), allowing for a new soil nutritional balance by decreasing fertilizer inputs and relying on legume nitrogen fixation rather than nitrogen fertilizer use. In Exp-2, after the first year of the implementation of PIMs, FNP2 reached values of 51% of high-quality grasses, 12% legumes and 23% BLS (*Table 2*).

	Treatments	**Bv %**	**Lp %**	**Hl %**	**Dg %**	**Legumes %**	**OG %**	**BLS %**
Exp. 1	DCP	12	35	27	0	6	6	15
	LTCP1	17	45	20	0	11	0	7
	NFNP1	2	0	4	3	11	7	71
	FNP1	0	5	0	3	13	13	64
Exp. 2	DP	1	68	2	0	7	19	2
	DDP	26	28	8	9	7	12	8
	NFNP2	0	1	8	0	12	47	29
	FNP2	14	20	0	17	12	13	23
	LTCP2	2	57	2	0	9	17	12

Bv *Bromus valdivianus*; **Lp** *Lolium perenne*; **Hl** *Holcus lanatus*; **Dg** *Dactylis glomerata*; **Legumes** *Trifolium repens*, *Trifolium subterraneum and/or Lotus uliginosus*; **OG** *Other Grasses Agrostis capillaris, Anthoxanthum odoratum and/or Poa annua*; **BLS** *Broad leaf species Leontodon nudicaulis, Hypochaeris radicata, Rumex acetosella and/or Taraxacum officinale*. Data collected the following spring, after the implementation of PIMs.

Table 2. Botanical compositions after one year of PIMs.

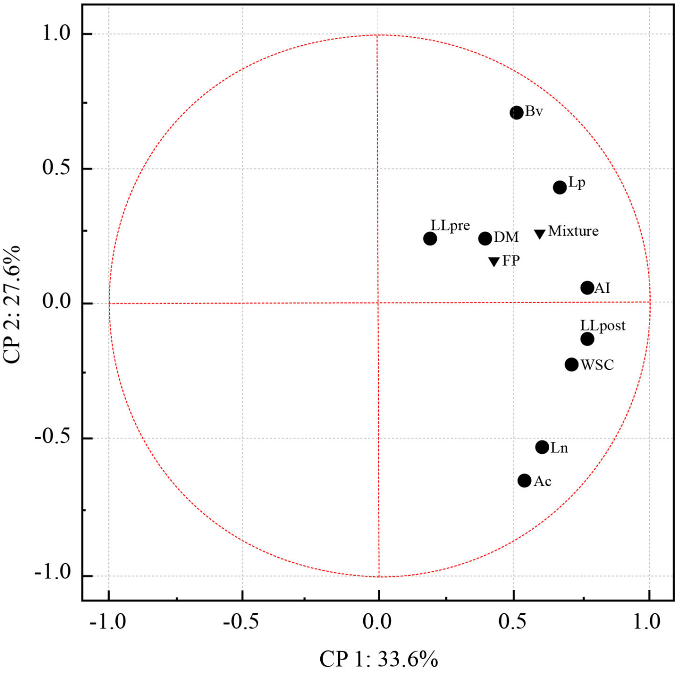

Figure 1. A plot of the first two PC loading vectors describing the relationships between pasture types (mixed and fertilized pastures FP), pasture species (Lp, L. perenne; Bv, B. valdivianus and Ln, L. nudicaulis), water-soluble carbohydrate concentrations (WSCs), pre-grazing lamina length (Llpre), post-grazing lamina length (LLpost) and apparent intake (AI). (▼) Score vectors; (●) Loading vectors.

Grazing management can have significant impacts on soil fertility status and pasture growth (Dubeux and Sollenberger, 2020). Proper grazing management improves nutrient distribution and pasture growth because it promotes a more uniform distribution of nutrients via excreta, which can potentially reduce fertilizer requirements and risks associated with the nutrient build-up in the soil when adequate stocking rates are used (Matthews et al. 2004). Rotational intensive sheep grazing often results in more uniform nutrient distribution than continuous grazing systems. The increase of high fertility responsive grasses (Flores et al. 2017) was positively correlated with increased pasture dry matter (DM) production (close to 14 Mg DM ha^{-1} year^{-1}), animal apparent intake and tillering of the perennial grass species (*Figure 1*).

The high presence of stress tolerant species in a pasture reflects the poor condition of the ecosystem at that time (López et al. 1995, Kemp and López, 2016). Normally, stress tolerators are slow growing species that provide little annual herbage mass and may invest energy in avoidance defoliation mechanisms that lower the probability of being grazed (Grime, 1988; Briske, 1991, 1996). Therefore, a sustainable improvement of pastoral systems with low productivity (< 5 Mg DM ha^{-1} year^{-1} *Table 1*; Descalzi et al. 2020) requires strategies that holistically integrate the progression of the pasture's condition (Gastó et al. 1993), increasing annual herbage mass production, but also considering the state of the soil and the future defoliation management of the site. The potential production of a pasture depends on the level of soil fertility and other inputs, such as chemical fertilizers, lime, macro and micronutrients (Keim et al. 2015). However, chemical fertilizers have the potential to induce eutrophication in (surface) waters (Silveira and Kohmann, 2020) due to (surface) runoff and nutrient leaching (Sharma et al. 2021). Due to these concerns, pasture fertilization strategies are generally aimed at balancing agronomic requirements, based on soil laboratory testing results (Silveira and Kohmann, 2020).

In any grazing system, the relations between soil, plants and animals are dynamic; if external forces modify one of these components, the other components adjust themselves accordingly over time (Greenwood and McKenzie, 2001). Pasture defoliation and plant senescence, in addition to animal excreta, are considered inputs of soil organic matter (SOM), which acts as organic binding between soil particles forming soil water-stable macro- and micro-aggregates (Tisdall and Oades, 1982). Sustainable pasture production depends upon a clear understanding of both soil physical processes at different scales as well as the impacts of human-induced management practices (Horn et al. 2019).

Ordóñez et al. (2018) affirmed that FNP2 had the highest biomass production during a severe drought event in the summer of 2015, when the soil reached permanent wilting point values at 10, 20 and even 60 cm soil depths. The growth rate for FNP2 during the summer of 2015 was 20.1 kg DM ha^{-1} day^{-1}, a highly positive value especially when compared with that of LTCP2 during the same period, which showed a growth rate of 2.5 kg DM ha^{-1} day^{-1} (Ordóñez et al. 2018). These differences may be explained by FNP2's soil water extraction effectiveness (Blum, 2009), an assumption supported by the lower soil water content (SWC) at the 60 cm soil depth, which can be related to the functional roots at this depth (Ordóñez et al. 2018). It is also important to consider that FNP2 had a higher number of species, including high-quality grasses and broad leaf species (weeds) (Descalzi et al. 2020).

Effects of PIMs over time on the physical properties of the Andosol

Soil structure is a dynamic physical property, which changes continuously due to external (i.e., trampling, soil compaction) or internal stresses related to wetting-drying cycles (Bronick and Lal, 2005). This provokes a reorganization of soil pore space, i.e., an improvement in the continuity and connection of the pore system (Zúñiga et al. 2015). Maintaining the soil structure improves the transport of water and nutrients (Ordóñez et al. 2018).

The mechanical resistance of aggregates in tilled soils proved to be less than that of no-tilled soils, although the bulk densities were identical (*Figure 2A*). Soil tillage favored medium pore size, improving the plant available water, while wide coarser pores and the air permeability was higher in no-tilled soils (*Figure 2B* and *C*). In the medium-term (2010-2012), lime applications maintained the soil pH in a range of 5.6 to 5.7, but the exchangeable calcium increased from 2.2 $cmol_{+}\ kg^{-1}$ to 4.7 $cmol_{+}\ kg^{-1}$ and mineral-N increased from 33 $mg\ kg^{-1}$ to 60 $mg\ kg^{-1}$ under pastures without soil tillage (*Figure 2D*). The wetting angles for tilled and no-tilled soils were 75° and 87°, respectively (*Figure 2E*).

This suggests that SOM under non-disturbed pastures tends to be more hydrophobic than pastures established through soil tillage and could explain the high presence of fungi and reduced wettability in no-tilled soils (*Figure 2F*).

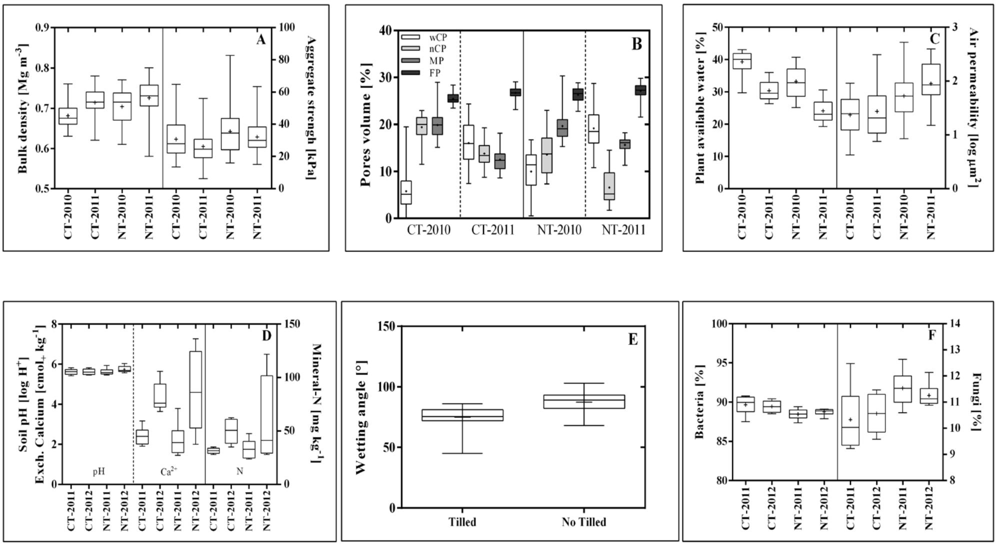

Figure 2. Physical, chemical, and biological properties under tilled and no-tilled soils in the medium-term. A: Bulk density and aggregate strength. B: Pore size distribution. C: Plant available water and air permeability- pre-drying at -60hPa. D: Soil pH, exchangeable calcium, and mineral-N. E: Wetting angle and F: Proportion of bacteria and fungi. Box plots show quartiles, median and mean (+).

When the soil is disturbed by tillage practices, the proportion of stable aggregates and the C and N associated with the aggregates have been found to decrease compared to no-tillage systems (Weidhuner et al. 2021). In Exp-1 and Exp-2 (*Figure 3*), an increase in air permeability

(Ka) during the second period of evaluation was assessed, indicating an improvement of pore functionality (Zúñiga et al. 2015), which facilitates the transport of air and water (Dörner and Horn, 2006). For Exp-1, an increase in soil physical quality from 2010 to 2011 was reported (e.g., in Ka), although this was not found when the treatments were compared in the same year (Zúñiga et al. 2015). The FNP1 treatment showed the highest Ka-value and a median increase, reaching 0.885 log μm^2. Similar results were assessed in Exp-2, where FNP2 also showed the highest value and a median increase, reaching 1.585 log μm^2. For the pastures established with traditional tillage (DCP, LPCP 1 and LPCP2) in both experiments, an important recovery of Ka was found, with an increase of up to 0.15, 0.65 and 1.51 log μm^2, respectively. The treatments established under zero tillage in Exp-2 showed constant Ka values during the first sampling period (2013), indicating the conservation of the soil structure and the continuity of macro porosity (Ordóñez et al. 2018).

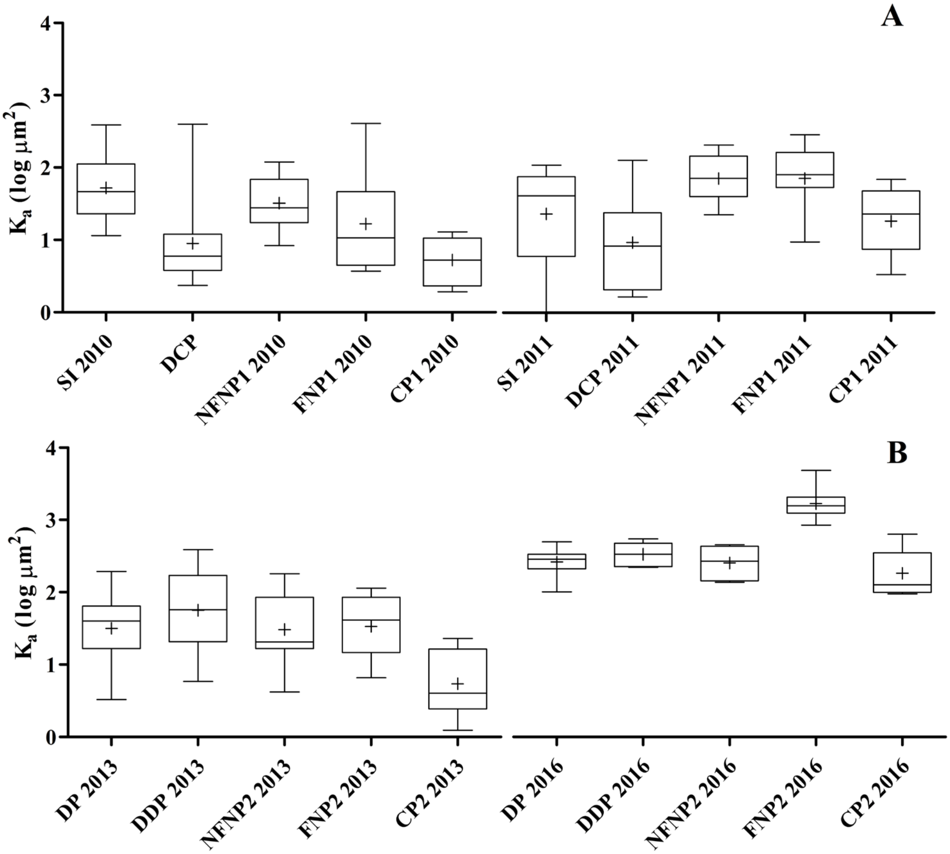

Figure 3. Temporal dynamic and treatment effects of the air permeability (Ka) for experiment 1 (A) and 2 (B). Symbols are described in section 2.2. Twelve samples per treatment for experiment 1 in 2010 and 2011, and experiment 2 in 2013, were collected. Seven samples per treatment for experiment 2 in 2016 were collected. Box plots show quartiles, median and mean (+). Lowercase letters indicate statistical differences.

Overall comments and future perspectives

The impacts of PIMs on the functionality of a pasture system depend on the initial physical condition of the soil and can be derived from the indicators presented in this analysis.

Traditional tillage has proven to destroy soil aggregates (Zúñiga et al. 2015), promote a loss of organic carbon (Haas et al. 2016, Shen et al. 2018) and reduce soil resilience (Dörner et al. 2020). The recovery depends on the wetting/drying cycles, cementation of the soil particles and the structure stabilization performed by the macro-, meso- and microfauna within the soil (Bronick and Lal, 2005; Ivelic-Sáez et al. 2015). Furthermore, land use, soil type, clay content and type, and the content and quality of SOC change structure-related physical soil functions, which can also be related to soil biodiversity in agroecosystems. The establishment of critical values of such physical parameters is essential for the sustainable management of volcanic soils due to their extreme soil physical properties (Dörner et al. 2013, Valle et al. 2018).

The important impacts of species traits on ecosystems have been the subject of many recent studies. These studies have addressed how species functional traits could modify the soil nutrient status (Fornara and Tilman, 2009), soil structure (Moreno-Espíndola et al. 2007) or soil water dynamics (Ordóñez et al. 2018), possibly generating a redistribution of water (Skinner, 2008).

In Exp-2 the differences between treatments were evaluated through the soil water uptake (derived from SWC measurements). The differences in SWC were intensified under severe drought circumstances, where the pasture growth rates were determined by the capacity of species to avoid or tolerate soil water restrictions (Ordóñez et al. 2018). The latter is related to differences in species functional traits within the pasture (Hooper and Vitousek, 1997), such as deep-rooted traits reflected by the SWC at different depths. It must also be considered that the soil functionality and pore continuity could facilitate roots vertical growth and soil water redistribution from deep within the soil to the surface due to differences in the soil matric potential (Descalzi et al. 2020). Therefore, species functional traits and soil structure may be important variables related to pasture growth during severe water restriction periods.

Changes in SOC content in relation to PIMs in Andosols

NFNP2 and FNP2 were found to be the treatments with the highest SOCs, with values at 10, 20 and 60 cm soil depth (Córdova, 2019) of 7.6%, 6.7%, 1.8%, and 7.5%, 7.0% and 1.7%, respectively. Furthermore, FNP2 showed higher values of air capacity ($p \leq 0.05$). Haas et al. (2018) studied the soil physical quality at soil depths of 45-55 cm (D1) and 55-65 cm (D2) in treatments NFNP2, FNP2 and CP2. An increase in air permeability from D1 to D2 was evident in all three treatments. However, even though no statistical differences were found, this increase was greater in FNP2 compared to the other PIMs applied. Furthermore, the authors indicate that D1 > D2 in FNP2, most likely due to its higher root exploration at that depth.

Changes in the SOC turnover in agroecosystems are explained by the aggregate rupture due to soil tillage, which provokes the mineralization of labile SOC (Six et al. 2000). In the long-term, the SOC is protected in occluded microaggregates (microaggregates-within-macroaggregates), which can be used as a total SOC indicator and are related to soil biota functions (Six and Paustian, 2014). Piazza et al. (2020) reported that N fertilization increases the mass of SOC in occluded microaggregates, in addition to the activities of soil enzymes under no tillage systems. In turn, no-tilled soils have lower bacterial diversity and community composition compared to soils under ridge tillage; however, after wetting-drying cycles, the soil bacteria community recovered faster under no tillage systems (Zhang et al. 2021). Therefore, the higher values of SOC for FNP2 and the higher soil water extraction reached by the roots (derived from the soil water content at the 60 cm soil depth in Ordóñez et al. (2018) in this treatment indicate how deep-rooted species (i.e., *B. valdivianus*, *D. glomerata* and BLS) can increase organic matter allocation and may improve physical and biological properties.

The importance of soil biodiversity in PIMs

Pastures provide multiple ecosystem services, such as the regulation and storage of water and nutrients in the soil (Havstad et al. 2007). They are essential for maintaining high levels of biodiversity as well as carbon sequestration (Conant et al. 2001). Furthermore, the literature has highlighted how an increase in plant diversity within the pasture (determined by the botanical composition) is an important driver for microbial biomass (Thakur et al. 2015, Flores et al. 2017, Descalzi et al. 2020) and soil biota resilience against disturbance (Bennet et al. 2019). Unsustainable pasture management (e.g., overgrazing, excessive fertilization and external input dependency) do not provide these services, driving the loss of soil functions and soil degradation (Rumpel et al. 2015). It is thus important to evaluate the effects of different pasture management strategies on soil properties, soil biodiversity and its functions through monitoring soil biological indicators. Thus, establishing a warning system of potential losses of soil functionality could be key to reduce the threat of soil degradation driven by climatic and anthropogenic effects (Schloter et al. 2018). Additional biological indicators are critical to improve soil quality assessments (Barrios, 2007). Nevertheless, soil biological indicators are still underrepresented in soil quality assessments (Bünemann et al. 2018), most likely due to the absence of well-defined standard methodologies to assess them (Schloter et al. 2018). The definition of the most appropriate bioindicators to represent and assess soil biodiversity and the attributes that best describe its variation is a developing debate.

The measured biological indicators should include soil organisms present at different soil food web levels (from soil microbial indicators to soil fauna indicators) and should be selected with regards to their: *i)* relevance for soil functions, *ii)* sensitivity response to management and *iii)* easy interpretation in terms of ecological significance (Paz-Ferreiro and Fu, 2016). Nevertheless, considering the advantages and disadvantages of technical molecular approaches (Lagos et al. 2015), methods focusing on DNA and RNA are tools that could complement existing biological and biochemical soil quality indicators. We considered that key genes, such as those related to biochemical cycling, may be used as indicators based on genotypic and phenotypic community diversity (Lagos et al. 2015, Morrison et al. 2017).

It has been estimated that 1 g of soil contains close to 1 billion bacteria cells consisting of tens of thousands of taxa, up to 200 m fungal hyphae, and a wide range of mites, nematodes, earthworms, and arthropods (Wagg et al. 2014). In this complex environment the diversity of coexisting saprophytes, epiphytes, endophytes, pathogens, and beneficial microorganisms (Avis et al. 2008) are soil quality indicators (Chaparro et al. 2012). They play an important role provided that nutrient cycling (C, N, P, etc.), plant growth (Bastida et al. 2009), C sequestration and biotransformation of organic pollutants (Thiele-Bruhn et al. 2012) are essential soil processes. The soil biota is spatially and temporally dynamic (Blagodatskaya and Kuzyakov, 2013). Microorganisms are usually quite inactive in bulk soil and are more active in soil zones such as the rhizosphere (Schloter et al. 2018). The latter is defined as a soil zone under the influence of roots, with a high microbial diversity, richness, and activity. The characterization of the structure, diversity and functions of soil microorganisms is pivotal, when their role in soil nutrient cycling and plant growth is considered.

Soil fauna and its indices are considered an especially useful biological indicator because they provide a great amount of diverse information revealing soil quality and its changes due to the management (Yan et al. 2012). In fact, edaphic fauna does not only influence biological processes, but also soil physical and chemical activities, reflecting the interactions that these ecosystems experience (Domene, 2016; Haas and Horn, 2018).

Earthworms are a good indicator to assess the effect of PIMs (such as tillage) on soil biodiversity due to their positive relationship with physical and chemical properties (Bünemann et al. 2018). They represent one of the principal groups of soil macro-invertebrates. In temperate ecosystems, earthworms are generally more abundant and diverse in pastures where their direct and indirect influences on soil structure and SOC are critical. Depending on the intensities of management practices, the diversity and activity of earthworms, used as faunal indices, can be negatively affected (Singh et al. 2021). Soil nematode fauna biodiversity maintenance in different agroecosystems is also essential because nematodes have various functional links in the soil food web, contribute to the nutrient cycle, regulate microbial communities, redistribute other soil organisms in the soil matrix, and suppress pests (Sánchez-Moreno and Ferris, 2007). In addition, due to their high taxonomic diversity, the analysis of the nematode community can be used as an indicator of soil biodiversity. Therefore, understanding the functions of the soil food web in relation to the presence, abundance and diversity of its biological soil component is a basic requirement for the management, maintenance and improvement of grassland production and its sustainability.

Conclusions

Pastoral systems can promote soil structure conservation (i.e., no-till), generate higher values of soil organic carbon contents and higher soil water extraction from the soil profiles. The latter evidence how deep-rooted species (e.g., *B. valdivianus*, *D. glomerata*, *L. nudicaulis*, *Taraxacum officinale*) can increase the soil organic carbon allocation, and in turn, may improve the soil physical and biological properties.

Our results indicate that soil structure conservation may promote soil biodiversity. However, we must clearly define the soil biological indicators and their evaluation procedures to help connect the soil biodiversity-pasture conditions and production processes involved. Exploring the qualitative properties of soil organic carbon at a microaggregate scale could be good starting point.

Acknowledgements

We thank the FONDECYT Grant 1100957 and 1130795.

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Chapter 18

Soil health and biodiversity: Interactions with physical processes and functions

Rainer Horn, Stefan Schrader, Anneka Mordhorst, Heiner Fleige, and Richard Schroeder

Introduction

Risks and threats on soils as multifunctional reactors

Soils, as a three-phase system, are the most vulnerable constituents on earth, because they are not renewable. At the same time, they provide us not only food, feed, and fiber but also clean ground water and sufficient drinking water. But soils are also essential as a living medium for microbes up to macrofauna. Soil properties, like soil strength, total porosity, pore continuity, pore-size distribution, and air capacity, and soil functions, like plant-available-water storage and nutrient storage due to cation-exchange processes, all impact plant growth, root development, and the depth distribution of roots. Furthermore, these properties and functions affect total biomass production, activity of soil biota, and the type and amount of soil carbon (SOC) and vice versa. Thus, soils are multifunctional reactors.

The functions are, however, threatened by soil acidification, erosion by wind and water, or soil deformation due to soil compaction and shearing, which do not only result in changes in the proportions of air, water, and solid volumes, but also in their spatial arrangements and pore continuity. Furthermore, such modifications cause changes in soil structure, pore connectivity, and particle or pore surface accessibility. They depend on water saturation and physicochemical reactions. The role of the biodiversity pool in all these multifunctional interactions is, however, often neglected, although bioturbation, exudates, and physicochemical reactions are interlinked with the functional community structure of soil organisms and their individual abundances. The organic input of vegetation and the dynamics of root growth affect these interactions. Haas et al. (2018) described the complex reactions and interactions at the root surface within the rhizosphere and also documented the consequences on physical, chemical, and physico-chemical reactions. However, these interactions are even more complex. The dramatic loss of biodiversity in time and space is known (IPBES, 2019), including the fact that one million of the estimated 8 million species of animals, plants, and fungi on earth are threatened with extinction rates at least 10 times higher than in the last 10 million years. The corresponding adaptation of land-use and soil-management practices for sustainable agriculture and soil protection is still insufficient to overcome the loss. A recent report (Leopoldina, 2020) states that, for Germany and Central Europe, many species, plants, and soil inhabitants are increasingly being reduced as a result of the intensification of land use. Such intensification includes many interlinked processes, which cause not only direct changes in the processes but also alter many more soil properties and functions.

If we only analyze the interactions between soil organisms and vegetation in combination with soil temperature, water constituents, transport chemistry, and mechanical processes, it becomes obvious that soils, as a three-phase and living system, show utmost complex reactivity schemes and fragile soil functionality. These highly dynamic interactions and the consequences of any kind of impacts on soil properties and functions can be derived from the connections between the various compounds. If, for example, a stress is applied to the soil, it has a direct impact on mechanical properties and the pore system, but it also affects the water suction, water content, and water flow due to changes in hydraulic properties. Such impacts also alter the thermal and chemical properties, and they either directly affect the soil organisms and vegetation or indirectly affect them through changes in the soil temperature and heat flow. Finally, the transport chemistry is modified either due to increased chemical reactivity (Reaction velocity-Temperature-Regulation: RGT rule, van't Hoff's rule) or indirectly through interlinked chemical mass flow or diffusivity. There are many further interlinkages, which show how complex soil properties and functions are (*Figure 1*).

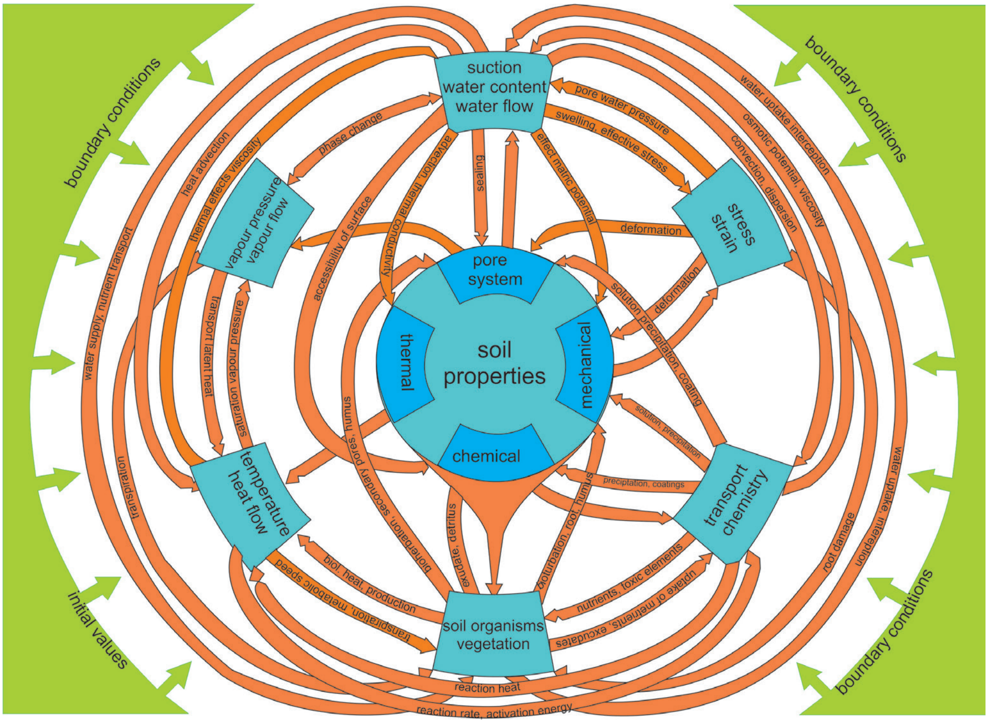

Figure 1. Interactions between soil constituents and multidimensional changes in soil properties and soil functions (Gräsle et al. 1995).

Soil resilience, therefore, only defines a quasi-dynamic equilibrium. Impacts beyond the internal soil rigidity, caused by thermal, mechanical, hydraulic, or chemical processes, alter the equilibrium conditions until a new steady state is reached. The consequences of such impacts due to either natural soil processes, like acidification and clay migration, or anthropogenic processes, like degradation due to sealing, erosion, or soil deformation, all affect this quasi-dynamic and, therefore, sensitive equilibrium. The equilibrium is finally affected by the worldwide changes of the climate.

Soil biodiversity perspectives from soil physics

Soil compaction and shearing deform the environment of soil biota

Each mechanical stress applied to soils can be attenuated without any change in soil functions, if the internal soil strength, defined as the precompression stress, is not exceeded down to a certain depth (Horn, 2021) (*Figure 2a*). In case of its exceedance, however, these properties will be changed until a new equilibrium is reached (*Figure 2b*).

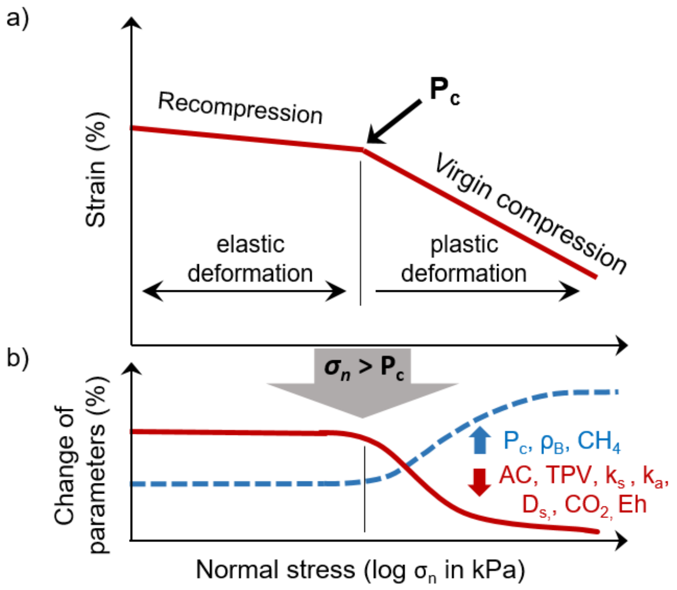

Figure 2. Stress-strain relation and the consequences for elastic (recompression line) or plastic (virgin compression line) soil behavior **a)** as well as for relative changes of soil properties. **b)** The precompression stress (P_c) defines the internal soil strength and the resilience limit. During the recompression load range, the properties and functions remain mostly unchanged while in the virgin compression load range the application of increasing stresses induce changes in the following: P_c, ρB = bulk density, CH_4 = methane emission, AC = air capacity, TPV = total pore volume, k_s = saturated hydraulic conductivity, k_a = air conductivity, D_s = gas diffusion, CO_2 = carbon dioxide emission, Eh = redox potential.

If divergent particle movements are caused by cyclic loading events, both air and water as 2 components of the 3-phase system must be also considered because stress induced drainage of air coincides with matric potential changes from negative to positive pore water pressure conditions. The loading events also coincide with weakening and altered swelling and are followed by shrinkage processes and with the formation of new and less rigid structure conditions or increased pore tortuosity with consequences for many soil processes (Zhai and Horn, 2018, Huang et al. 2021a). These interactions are furthermore enhanced if stress application includes shearing, because stress-induced strain does not only affect soil properties and functions through the aggregate breakage or particle rearrangement, but it also includes an interlinked change in pore water pressure. These dynamic processes result in an additional worsening of the pore tortuosity and reduced hydraulic flux and even retarded gas fluxes and a prolonged water saturation with further impacts on physicochemical processes, like a declining redox reaction followed by altered mobility of Fe, or Mn (Horn, 2021; Huang et al. 2021b). The alteration of the activity of the microbial community, and even the change of the soil from a sink to a source for greenhouse gases like methane (CH_4), can be detected as a consequence of soil deformation. The alteration is related to the (micro-)structural soil stability and the deformation behavior of soils (Haas et al. 2016).

Divergent and shear processes are, therefore, the main driving forces for changes in soil biodiversity. They impact the community structure of soil organisms and their activity. Keller et al. (2019) documented the long-term, tillage-dependent yield loss, as well as the impacts of heavy machinery on root growth. They also observed increased densification of the plowpan layer and its enlargement with depth. As a consequence, Horn et al. (2019) documented a more enhanced horizontal anisotropy of the saturated hydraulic conductivity in arable subsoils of a model region (Schleswig Holstein, Northern Germany). Corresponding alterations in water and gas fluxes between the top- and subsoil will certainly affect the biological activity and abundance of soil microorganisms with consequences for the whole soil biodiversity pool.

Bioturbation – burrowing soil animals as drivers of soil development

Soils differ in terms of their biodiversity. From a global perspective, climate is a determining factor (Phillips et al. 2020), but also other factors, such as parent material, relief, water, vegetation, and human activities are important. For example, it is well known that soil properties such as pH and soil carbon control earthworm occurrence and diversity. Most earthworm species prefer soils with neutral to slightly acidic pH values. Soil organisms are significantly involved in different soil processes, e. g. decomposition and transformation of organic matter and litter, new formation of humic substances, or structure formation and bioturbation.

Bioturbation has already been described by Darwin (1881). Intense bioturbation is only observed in soils with favorable water, air, and nutrient conditions. Burrowing soil animals mix the litter layer with the upper mineral soil and produce a humic (=h) topsoil (A-horizon) with a characteristic crumbly structure. Bioturbation also promotes infiltration, even in compacted layers (e. g. Ruiz et al. 2015) and prevents other soil processes (e.g., decalcification), because soil animal activities can move translocated substances back to the surface again or transport subsoil material and deposit it on or in the topsoil (Blume et al. 2016). They can even change the morphology of the soil significantly (e. g. termite hills, Kristensen et al. 2015). In some black humus-rich soils like Chernozems (IUSS Working Group WRB 2014), bioturbation due to earthworms (worm-bioturbated=Vermic) and steppe mammals (hamsters, etc.) is so dominant that an A-horizon of more than 1 m can develop. Some of the topsoil material can reach very deep in animal burrows that have been filled with organic material (crotovina). Those black humus-rich soils are typically distributed in continental long-grass steppe regions, because the animals burrow deeper to avoid dryness in summer due to heat and in winter due to frost.

Table 1 illustrates the soil fauna activity (bioturbation) of the macrofauna in different landscapes. Bioturbation depends, obviously, on soil and climatic conditions. Earthworms, termites (which are particularly numerous in Africa and America), and ants (which can nearly be found all over the world) are known as ecosystem engineers, which change the physical structure of soils due to bioturbation, and even small mammals like moles and voles dig through the soil. In fact, the highest bioturbation is reached by lugworms on flat tidal coasts (Tidalic Gleysols, IUSS Working Group WRB, 2014), where the upper 15-25 cm of the topsoil is constantly mixed during low tide.

Animals	Ecotope	Animals
Earthworms	Meadows (UK)[a]	18-45
Earthworms	Gardens (Centr. Europe)	10-25
Earthworms	Meadows (Centr. Europe)	5-120
Earthworms	Forests (USA)	18
Earthworms	Tropics	36-270
Ants	Forests, steppes (Europe)	31
Ants	Steppes	31
Ants	Ants moist area (USA)	20
Ground squirrels, susliks	Steppes (Russia)	18
Ground squirrels, susliks	Semi-deserts	1.5
Prairie dogs	Prairies	70
Moles	Forests (Europe)	7-120
Termites	Savanna	12-60
Termites	Deserts	0.02-5
Lugworms	Mud flats (Europe)	600-3000

[a] = after Darwin (1881)

Table 1. Bioturbation (in Mg ha^{-1} y^{-1}) in different soils around the world (Blume et al. 2016)

In this context, interactions within the soil biodiversity pool and with its abiotic environment provide ecosystem services that shape or stabilize the soil system and that are used by humans in agricultural management. Amongst others, we can define the following ecosystem services:

- regulation of the biogeochemical cycles

- storage and supply of nutrients for plant growth

- formation, strengthening, maintenance, and renewal of the soil structure

- breakdown of waste or pollutants

- control of the hydrological cycle

- regulation of atmospheric trace gases

- regulation of soil-borne plant diseases

- function as a genetic reservoir for further usage options (e.g., in biotechnology)

Thus, the structural and functional diversity of soil organisms is an essential prerequisite for the functioning of terrestrial ecosystems as a whole and of agro-ecosystems (for further details see also Fonte et al. 2010).

Earthworms as ecosystem engineers

Within the soil biota pool, earthworms are multiple actors (according to Turbé et al. 2010) as **(1)** biological regulators of microbial communities **(2)** chemical engineers as decomposers of organic residues, and **(3)** ecosystem engineers modifying their environment physically by shaping the soil structure. Their role as ecosystem engineers covers the formation of aggregates (clay-humus-complexes) and macropores (burrow systems). *Figure 3* demonstrates burrow formation and functioning, which are in conflict with mechanical stresses and soil conditions. In arable land (here with focus on physical conditions), the earthworms' activity for burrow formation is affected by the given local soil conditions and the intensity of mechanical measures. The resulting burrow system with its structure and stability is affected by both factors, which control the functions of the burrow system as a habitat for the earthworms, other soil biota, and roots. The burrows also control other soil processes, such as infiltration, transport of soluble compounds, aeration, reduction of runoff, and erosion. These burrow-controlled processes, in turn, shape the soil conditions.

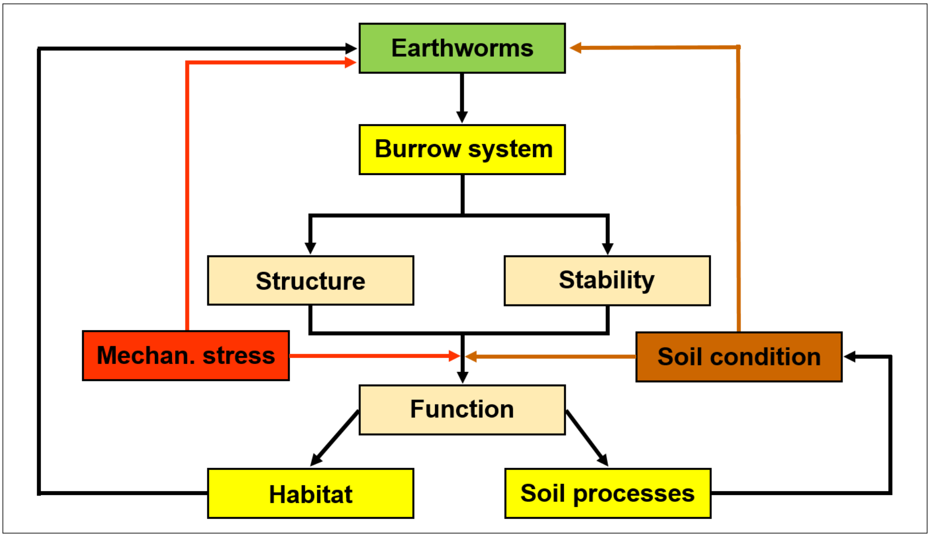

Figure 3. Soil as living space for earthworms under land use, which are in conflict with mechanical stresses and soil conditions.

Soil biota and alterations due to soil deformation

The activity of soil organisms concentrates in "hotspots" of functional domains of the soil system, such as the detritusphere, rhizosphere, drilosphere, and porosphere (Beare et al. 1995). Although these hotspots only cover less than 10% of the soil volume, they are characterized by much higher process rates compared to the average soil conditions (Beare et al. 1995; Kuzyakov and Blagodatskaya, 2015). Thus, the availability and accessibility of organic substrates inside such hotspots like intact aggregates or in the whole soil profile, determine the microbial activity, because micro- and macro-organisms, as potential decomposers, require adequate environmental conditions, such as oxygen, water, and energy supply (Doran and Linn, 1984; Six et al. 1998, King et al. 2019, Young and Ritz, 2000, Nawaz et al. 2019). If external mechanical stresses threaten these hotspots, changes in the soil-organism communities, and their composition therein, will occur because shearing interrupts the soil-pore continuity and connectivity resulting in isolated pores. In this situation, the oxygen level declines rapidly and modifies microbial communities, which results in a shift from aerobic to mainly anoxic processes with consequences for the cycling of nutrients (Stepniewski et al. 2002).

Consequently, physical soil parameters, like pore size distribution and matric potential, become crucial in understanding carbon-turnover processes and biological activity (variations in CO_2 efflux), and the variation in the belowground microbial and faunal community structure, at given locations within the soil matrix (Killham et al. 1993, Mordhorst et al. 2014, Althoff et al. 2009). Kochiieru et al. (2018) emphasized the influence of macroporosity, surface area, and macropore range on CO_2-efflux and its strong relationship to soil type and land use as a documentation of microbial activity and composition. Jasinska et al. (2006), Wiesmeier et al. (2012), and Carlesso et al. (2019) stated a strong link between the microbial activity and soil strength, carbon storage, and mechanical impacts. The relationships also underline the link between the mechanical hydraulic-pneumatic, thermal, and physicochemical processes and functions (for more details see also Richards et al. 1997, Horn, 2021).

The impact of compaction on changes in microbial community structure and activity is highly variable across different soil textures, compaction levels, and water contents (Santrucková et al.1993, Ruser et al. 2006, Frey et al. 2009, Pengthamkeerati et al. 2011, Carlesso et al. 2019). However, compaction-induced changes in the pore system leads to less favorable conditions for microorganisms, which is mainly attributed to restriction of gas or water fluxes and lower aeration status due to reduced porosity (Whalley et al. 1995, Otten et al. 2000, Beylich et al. 2010), refinement of pore sizes and disconnection of transport pathways by soil compaction (Doran and Linn, 1984, Pengthamkeerati et al. 2011).

In laboratory experiments, Mordhorst et al. (2014) showed that structural deterioration by mechanical loading exceeding internal soil strength led to a strong reduction in basal respiration (CO_2-efflux). They also gave a positive outlook, because the inhibition in soil respiration was not persistent, if the soil was exposed thereafter to natural structure formation processes, induced by wetting and drying, which re-improve the micro-environmental soil conditions of the habitat pore space. Furthermore, they stated that, if through structure deterioration by shear and compaction the accessibility of particle surfaces and coinciding biological impacts are increased, we can expect a coinciding reduction in organic soil carbon due to a complete release of CO_2 to the atmosphere (Wiesmeier et al. 2012) (*Figure 4*). This latter effect is, however, negative for global-change aspects and for the requested increase in carbon as contribution to the 4/mille initiative (Chenu et al. 2019). Thus, the rigidity, quantified as precompression stress, separates the beneficial storage and increased accessibility from the non-rigid conditions. This is followed by a complete alteration of the physicochemical properties and processes that affect the internal soil processes and the atmospheric composition.

In the medium term, microbial activity is likely to increase again after an enhanced energy supply (e.g., occluded carbon inside of aggregates) followed by structural rearrangements caused by mechanical and hydraulic stresses. This implies a high susceptibility of physical protection mechanisms for carbon and a high biological activity due to the mechanical disruption of the soil structure.

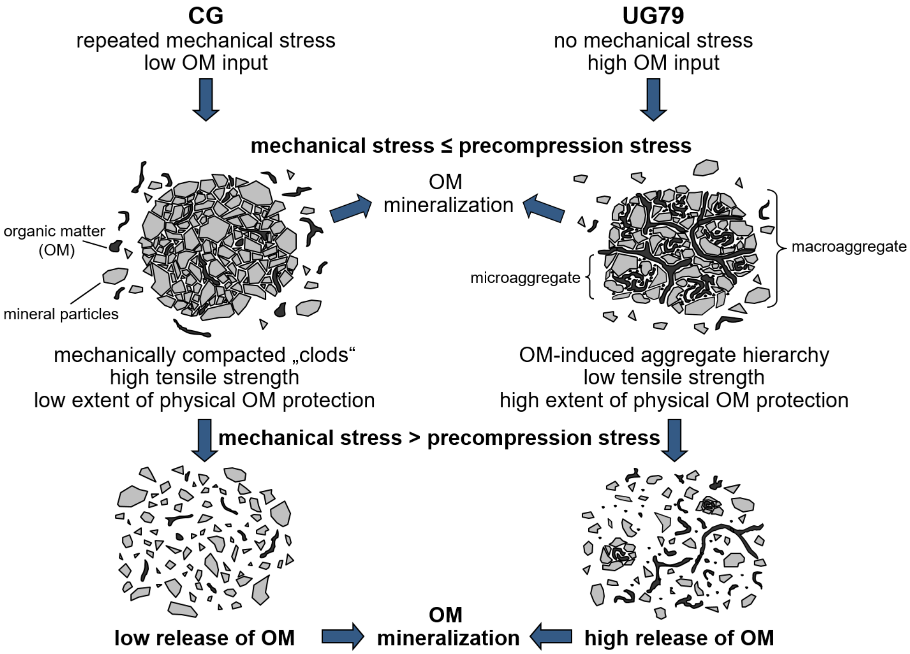

Figure 4. Effects of soil strength on pore rigidity and carbon as well as climate change (taken from Wiesmeier et al. 2012). CG = continuously grazed site, UG79 = ungrazed site since 1979

Besides the often-described indirect impacts, mechanical stress by soil compaction affects soil biota also directly. Heisler and Kaiser (1995) reported a decline in collembolan densities and changes in their community structure due to an increasing wheeling frequency on arable land. In relation to a modified pore size distribution, the size-class of soil fauna communities seems to play an important role. Numbers of earthworms and enchytraeids decrease generally, irrespective of the species diversity and the functional groups they characterize (Röhrig et al. 1998, Langmaack, 1999). Similarly, collembolan abundances decrease with increasing compaction; however, a few species remain unaffected or even increase in numbers (Dittmer and Schrader, 2000). Soil compaction does not change the numbers of mites, in general, but modifies their community structure with both a decrease and an increase of the abundance of different species (Schrader and Bayer, 2000). Finally, nematodes, as members of the soil microfauna, do not change in numbers, but they respond with a functional change in their community structure (Bouwman and Arts, 2000). Results from a field experiment in a grassland with increasing compaction by wheeling with 4.5 Mg, 8.5 Mg, and 14.5 Mg loads, compared to the uncompacted control, revealed a significant shift towards a nematode population with increased numbers of herbivores and decreased numbers of bacterivores and omnivores or predators (Bouwman and Arts, 2000). This functional change has important implications for the soil food web and the incidence of plant diseases by phytopathogenic nematodes (herbivores) feeding on living roots.

Three years after soil compaction by six times wheeling with 5 Mg, earthworm and enchytraeid populations recovered along with a recovery of soil physical parameters, like bulk density, air capacity, and air permeability in the topsoil (Langmaack, 1999, Langmaack et al.

1999a). Recovery of earthworm populations results in expanding burrow systems as a newly formed macropore network and increasing amounts of casts as newly formed aggregates. Such reformed aggregates are, however, less stable compared to compacted aggregates of the bulk soil. They have about 10% higher porosity, which promotes water and nutrient storage processes (Larink et al. 2001). Furthermore, these porous aggregates offer space as a habitat for soil microorganisms. Earthworms reorganize soil structure; however, they do not loosen a compacted soil profile (Sauzet et al. 2021).

Interaction between soil strength and pressures exerted by earthworms

Precompression stress defines the internal soil strength as the combined result of natural soil structure formation due to swelling and shrinkage, its intensity and frequency, chemical processes, and biological activities.

Thus, when earthworms modify their soil environment by ingesting and egesting soil, as well as by pushing soil as consequence of their peristaltic locomotion, they need to apply forces, which affect the surrounding soil in axial and radial directions forming the so-called drilosphere analog to the rhizosphere. Soil displacement declines with distance from the earthworm individual (Barnett et al. 2009), which results in a spatial gradient with decreasing bulk density towards the soil matrix (Schrader et al. 2007). Generally, radial pressures are higher than axial pressures (*Table 2*), which is the same relation as for root growth. Growth pressures of roots, for example of pea seedlings (Pisum sativum), are ca. four times higher compared to earthworm pressures (Misra et al. 1986). However, growth pressure declines rapidly with age of roots when hydraulic growth is replaced by cell division. Endogeic earthworm species exert the highest radial pressures, whereas anecic species exert the highest axial pressures (*Table 2*).

Functional group	Species	Axial (kPa)	Radial (kPa)
Anecic	*Aporrectodea longa*	115.77	177.00
	Lumbricus terrestris	90.60	136.26
Endogeic	*Allolobophora chlorotica*	65.43	78.66
	Aporrectodea caliginosa	68.95	294.57
	Aporrectodea rosea	70.11	182.43
	Octolasion cyaneum	63.49	183.91
Epigeic	*Dendrobaena octaedra*	43.10	81.82
	Lumbricus rubellus	50.43	126.02

Table 2. Maximum axial and radial pressures (kPa) of common earthworm species during burrowing activity with respect to their ecological classification of anecics (deep burrowers), endogeics (topsoil burrowers), and epigeics (shallow burrowers); data from Keudel and Schrader (1999).

Thus, the more the number of earthworms is reduced due to tillage or intense mechanical stress with compaction and shear effects, the less intense is the new formation of earthworm channels as highly connected macropores. Bioturbation and soil mass movements are also reduced. The vertical channels especially improve the mechanical soil strength, because these pores are already equilibrated with the major vertical stresses and maintain their functionalities (Zhai and Horn 2018, 2019), and serves as food for microorganisms.

Roughly estimated, the earthworm species *L. terrestris* mechanically affects a soil volume of 126 m^3 $ha^{-1}y^{-1}$ in arable soil profiles of Luvisols, based on field data of earthworm populations and computed tomographic recordings of burrow systems in soil columns (Schrader et al. 2007). An annual production of burrow length of 82.3 km ha^{-1} has been calculated for *L. terrestris* populations in arable land under conventional management without compaction and only 3.3 km ha^{-1} after compaction by six times wheeling with a load of 5 Mg (Langmaack et al. 1999b).

Mechanical stress by wheeling affects soil bioturbation of earthworm communities differently, depending on the tillage system. In a three-year field study on a Luvisol, soil bioturbation was always higher under conservation tillage (CS) than conventional tillage (CT) (Schrader and Larink, 2003). In the first year, in uncompacted plots, cast production was highest (37.8 Mg cast dry weight $ha^{-1}y^{-1}$) under CS, which was nearly twice as much as under CT (18.6 Mg $ha^{-1}y^{-1}$). As a result of six times wheeling with 5 Mg, cast production under CS decreased to 25.0 Mg $ha^{-1}y^{-1}$ and under CT to 5.7 Mg $ha^{-1}y^{-1}$ (Schrader and Larink, 2003). Two years later, under CT, soil bioturbation in the compacted treatment was still ca. 50% below the uncompacted treatment, whereas under CS soil bioturbation was on the same level in both treatments (Schrader and Larink, 2003). Thus, under consideration of the actual internal soil strength with respect to mechanical impacts and optimal climatic, soil moisture, and plant growth rate conditions, a long-term recovery of the soil biodiversity pool and its functionality can be expected. In different forest types of cold-temperate regions, an average soil bioturbation by earthworm communities of 25 Mg dry weight $ha^{-1}y^{-1}$ has been calculated (Taylor et al. 2019). But the effects of mechanical stress on community structure and frequency of microorganisms and the long-term recovery of soil functions are unknown (Riggert et al. 2019).

Interdisciplinary perspectives from Agronomy

Land management impacts on carbon storage for soil biota and consequences for soil properties and functions

It is well known that most soil properties, such as soil structure and hydraulic properties, as well as the diversity and activity of soil organisms, are influenced by the quantity and quality of soil organic matter (SOM, which contains about 58 % SOC). It is also a key attribute of environmental quality and agronomic sustainability (Carter, 2002) and serves as food for microorganisms. It is essential for chemical, physical, and mechanical processes in soils at all scales. SOM leads to a more stable, habitable pore space and increases soil aggregation, and, hence, the ability of soils to withstand shearing forces, resulting in a more stable, habitable pore system.

These statements are derived from data collected over the last 40 years. Increasing the content of SOM enhances the mechanical strength, defined as cohesion (kPa), for the different soil texture classes according to Ad-Hoc-AG Boden (2005). It is also known that these relationships furthermore differ based on soil structure (*Figure 5*) (Schroeder et al. 2021).

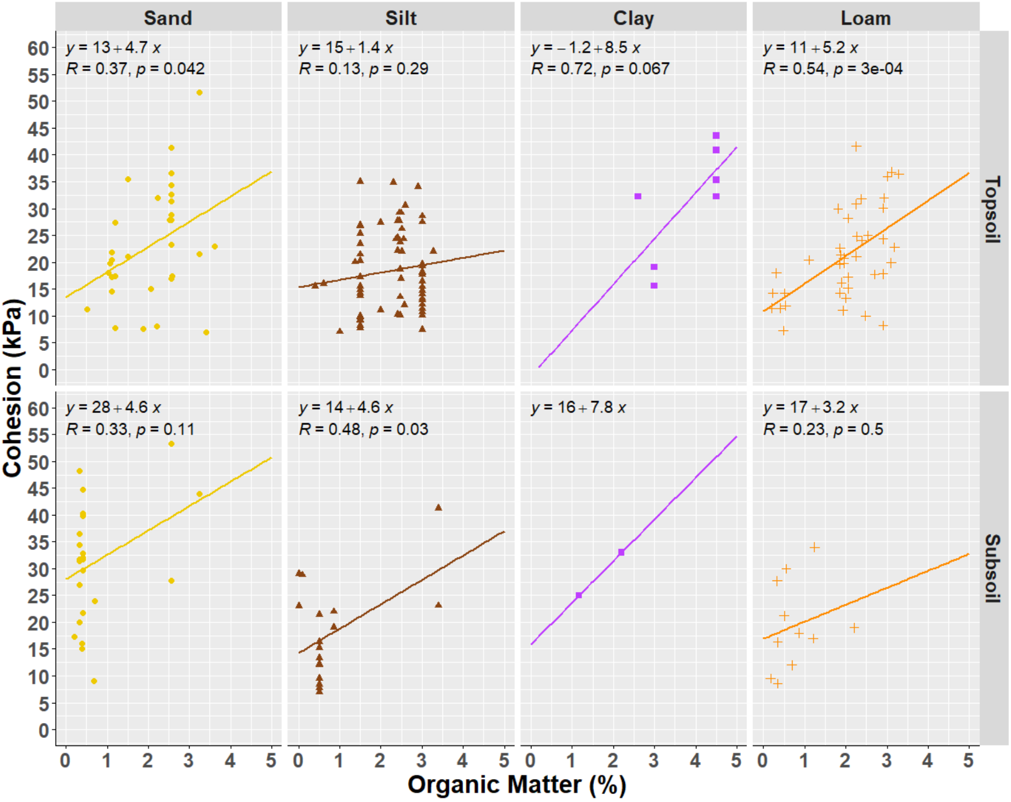

Figure 5. Changes in soil cohesion (kPa) with soil organic matter (%) divided into topsoil (n=139), subsoil (n=52), and the main texture classes of arable sites with CaCO3 < 1%, at a pre-drainage of -6 kPa, n = 191.

Besides the well-known organo-mineral bonding effects, the impacts of microorganisms enhance soil strength, which also explains the reduced rigidity of soils with a smaller contents of SOM.

Therefore, structural elements, like soil aggregates, plant roots, or preferential flow paths, can be expected to increase the biodiversity in soils, because the physical transition zone, in terms of the oxygen partial pressure across the coated walls of earthworm burrows (Haas et al. 2018), is known to be highly biodiverse. Additionally, instead of the expected increase in SOC according to the 4/mille predictions (Chenu et al. 2019), investigations on soil carbon stocks (down to 90 cm depth) documented the impact of land use (arable land, grassland, and forest) for 925 soil profiles in 4 different geological regions in Northern Germany (Mordhorst et al. 2018). Highest carbon stocks in the 0-30 cm depth were found in soils under grassland use, and the lowest were found under arable use (*Figure 6*).

Similar studies also stated that the high spatial variability in SOC content in top- and subsoils relates to the combination of soil type, climate, and topography, as well as the actual and historical land use (Wiesmeier et al. 2012, Mayer et al. 2019).

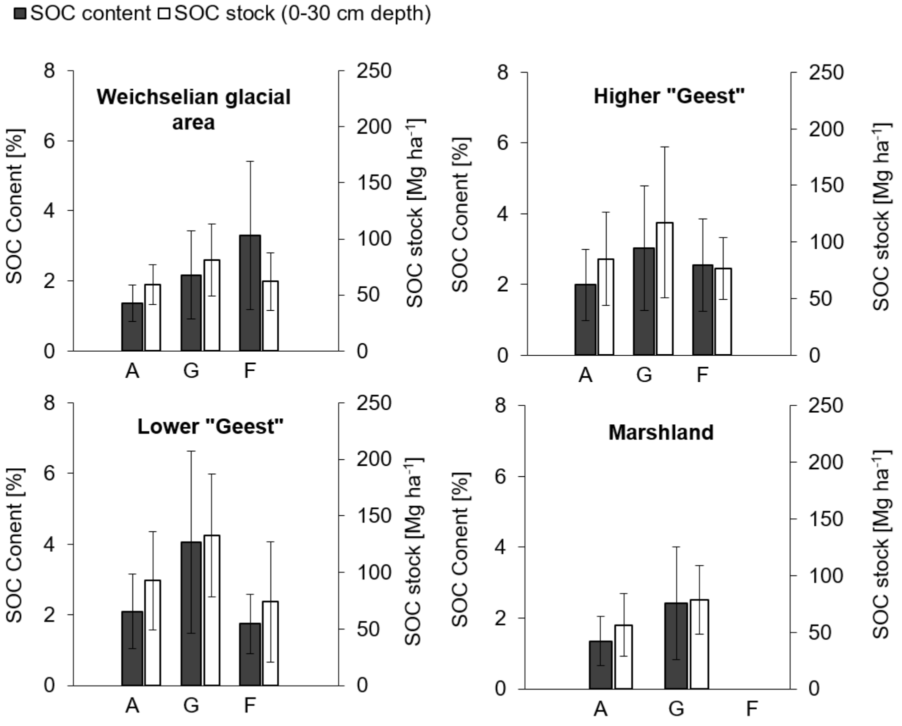

Figure 6. Effect of land use management (A = Arable land, G = Grassland, F = Forest) on mean soil organic carbon (SOC) content of mineral A-horizons and SOC stock within 0–30 cm depths for the region-specific soil types in Schleswig-Holstein (Northern Germany). Organic soils are excluded. Published in Mordhorst et al. (2018).

How far these processes can actually explain the 40-year trend in declining SOC content in arable soils in Germany (*Figure 7*), in combination with the increasing mechanical impacts (Keller et al. 2019), is often brushed aside. But the increasing irreversible soil degradation worldwide should strengthen our activities to prevent such impacts. The link between the decline in organic carbon with time and the increasing soil degradation due to soil deformation affects biological diversity and will end in a reduced functionality of soils, which are non-renewable. Feedback mechanisms react at all scales, because a reduced SOC content in soils coincides with a reduced internal soil strength, increased soil surface sealing, and increased surface runoff and water erosion, which, in combination, reduce nutrient and water storage (Wiesmeier et al. 2020).

Decline in storage of plant-available water and nutrients affects biological activity and community structure. Overall, soil properties and functions, including the total amount, the composition, and distribution of the organic substrates, and biological activity are directly linked and are strongly dependent on anthropogenic impacts. Thus, in conclusion, the need to fulfill the Sustainable Development Goals (SDGs) gains high importance.

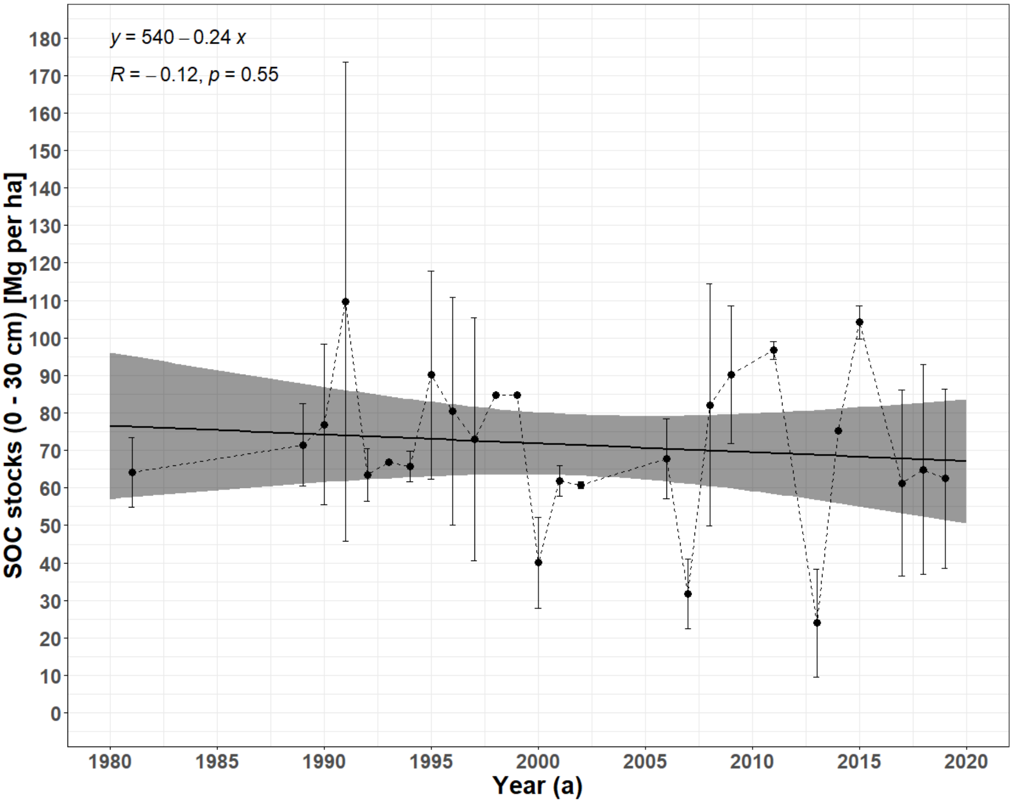

Figure 7. Average soil organic carbon stocks (SOC in Mg ha^{-1}) in arable topsoils at an assumed thickness of 30 cm between 1981–2019, n = 245.

Impacts of soil biota for economic approaches – some remarks

Because the ecosystem services of soil organisms in agro-ecosystems help to secure yields, they are also of economic value. The activities of earthworms, in their multiple functional roles (Turbé et al. 2010) as ecosystem engineers, chemical engineers, and biological regulators, can be assigned to indirect economic values (Decaëns et al. 2006). It is difficult to quantify the value in concrete terms. Nevertheless, a recent study has succeeded in assigning an economic value to earthworm services by calculating the standard gross margin (SGM) as a measure of the relative contribution of crop production to overall farm revenue (Plaas et al. 2019). While Plaas et al. (2019) used fungicide applications and their potential for reduction due to feeding of earthworms on phytopathogenic fungi, Jonsson et al. (2019) based their concept of an economic analysis on fertilizer applications. Schon and Dominati (2020) defined a set of proxies to develop a monetary valuation of earthworm services in different pastoral systems. These ecological-economic approaches open up the possibility of evaluating the performance of soil biota depending on soil conditions and management measures, taking them into account in the context of recommendations for sustainable management measures. Contrary to some examples of chemical impact, an ecological-economic assessment of mechanical impact is still lacking. Valuation of soil ecosystem services delivered by the soil biodiversity pool provides perspectives for assessing the sustainability of agricultural management measures with respect to, for instance, reaching the Sustainable Development Goals (SDGs) of the UN and the Green Deal targets of the EU; for the soil perspective of SDGs see Keesstra et al. (2016) and of the Green Deal see Montanarella and Panagos (2021).

Conclusions

Soil properties and functions are affected by natural pedogenic and anthropogenic impacts, which result in a site and management specific soil resilience due to the various sensitivity of soil horizons.

Exceeding the internal soil strength changes physical, chemical, and biological functions, including structure and activities of the biodiversity pool.

Earthworms can withstand external stresses applied, and, as well, they themselves can actively reform coarse pores and improve the pore continuity and mechanical properties over depth; they also can affect the accessibility of particle and pore wall surfaces for hydraulic, thermal, and pneumatic processes, as well as chemical or physico-chemical reactions.

Impacts of land management on carbon storage must be considered as a key issue, because carbon directly impacts soil biota with consequences on soil properties and functions.

Evaluation of soil ecosystem services delivered by the soil biodiversity pool provides perspectives for assessing the sustainability of agricultural management measures with respect to, for instance, reaching the Sustainable Development Goals (SDGs) of the UN and the Green Deal targets of the EU.

Acknowledgements

Many of the data is obtained during various research projects sponsored by the German Research Foundation (DFG), BONARES, Ministry of Research and Technology (BMFT), and the German Environmental Agency (DBU). The authors are thankful for the long-term continuous financial support

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Interdisciplinary perspectives from soil chemistry and education point of view

Chapter 19

Green chemistry and education as tools to protect the soil and its biodiversity

Laura Bertha Reyes-Sánchez

Introduction

Chemistry is present in our life all times and everywhere. Everything that surrounds us and makes our modern life easier includes chemical products. However, most citizens do not associate the products they use daily -soaps, medicines, electronic devices, cosmetics, etc.- to their chemical composition, and when they do, it is through a negative perception in which chemistry is related to environmental pollution and harmful products to the environment.

For example, if we talk about food production, it is common to hear recommendations to grow them using manure and compost instead of chemical fertilizers. However, manure and compost are also chemical compounds, and the difference is that fertilizers are inorganic chemical compounds while compost and manure are organic chemical compounds. Notwithstanding, both are chemical fertilizers. Organic and inorganic compounds are chemical matter. However, each substance and compound, has a unique and distinctive chemical structure that determines its chemical, physical and biological properties. That chemical structure also defines its possible applications and uses for our daily life, as well as its effects. That is why the response of crops to inorganic and organic chemical fertilizers is different, and experimental evidence indicates that the nutritional quality of organically grown foods may be better than that based on inorganic fertilizers (Diacono & Montemurro, 2010).

Impact of chemical compounds on the environment

Every compound and product have a chemical composition produced through some chemical transformation. Most chemical products that are part of our daily life are manufactured through industrial chemical processes that transform the various raw materials into products of quotidian use. However, industrial chemical processes do not always consider their repercussions on the environment and for life (De La Hoz, 2009). These industrial activities' impact on the environment can be punctual or extensive and of different orders, types, and degrees of affectation to the natural resources of the planet and all living beings. But not all the chemical products that are dumped and spread to the environment do not always come from the chemical industry, but reaction processes based on fertilizers and biocides application in agriculture, or cleaning, pharmaceutical, cosmetic, electrical industries, etc., all affect the environment due to long-term decomposition and corresponding changes. *Ibid.*

It is well known that the soil is one of the most critical life-supporting components of the biosphere. Soil provides many ecosystem services, such as a habitat for biodiversity, water and nutrients, food, feed, fiber and energy (Horn, 2021). However, notwithstanding its value for life, chemical substances can spread through the air, water, or soil during their manufacturing process, when using them or during their elimination, and their impact on the environment depends on what substance it is, what is the amount and concentration, as well as from the place where that substance is poured and accumulates. That is how the transformation of agriculture into a high technology system aimed at constantly improving yield has increased the number and concentration of agrochemicals dispersed in the environment in recent decades with strong impact on soil and water resources. (Tilman, 2001).

According to Rodríguez-Eugenio (2019), "Soil pollution refers to the presence in the soil of a chemical or substance out of place and/or present at a higher than the normal concentration that has adverse effects on any non-targeted organism". A broader definition of the concept is that of Albert (1994). He defines contamination as the insertion of biological, chemical, or physical agents to a medium to which it does not belong, causing an unfavorable modification in its natural composition when this modification manifests itself through adverse effects in the three natural environments: air, water, and soil.

Among the chemicals most used in agriculture are the various biocides and fertilizers, which can pollute the soil. Although the chemical action of biocides is aimed at the control of certain species, in reality, they also affect human beings either by inhalation or through the consumption of other animals and plants that were exposed to high concentrations of them (Sánchez-Bayo, 2011). According to information generated by the scientific community, there is no doubt that pesticides are a central factor responsible for the decrease in terrestrial biodiversity (Sánchez-Bayo and Wyckhuys, 2019). Likewise, fertilizers are a source of accumulation of salts in the soil, variation of the pH, deterioration of its structure and consequent alteration of both its physical and physicochemical properties, eutrophication of water, acidification and contamination of drinking water due to excess nitrogen, and phosphorus, thus affecting soil microbial communities that are critical for their resilience (FAO, 2020).

This deterioration or enhanced alteration of the physical, chemical, and biological properties of the soil due to its contamination by agrochemicals prevents it in different degrees from developing its elementary functions of biomass production, decomposition, production and transformation of matter (Meena et al. 2020), and transport of nutrients and substances, as well as its mechanical properties affecting their storage and filtering of water capacities, but also the loss of soil biodiversity (Horn and Blum, 2020). The loss of these functions directly affects the sustainability of ecosystem services which are indispensable for human life because soil organisms are essential for most of those services provided by soils: from the provision of healthy food and the improvement of agricultural production to water filtration, carbon sequestration, the degradation of target contaminants, and the supply of medicine (FAO, 2020). Therefore, soil biodiversity determines the main processes that make life possible on the Earth, thanks to the multiple connections between above and belowground reactions *Ibid*.

The degradation and the consecutive loss of the soil resource coincides with changes or finally the loss of all terrestrial flora, and with it that of the fauna that is part of it and to which it feeds. It also means a terrible loss of planetary biodiversity and therefore a serious destruction of the food chain at all levels (Reyes-Sánchez, 2018).

The change started with the spring

It was not until 1962 that the publication of "A Silent Spring", a work published by Ph. D. Raquel Carson, led the scientific community for the first time to seriously consider the negative results of using agricultural pesticides on the food chain since the documented information about the harmful effects given by this scientist objectively stated that the widespread use of pesticides caused more damage than the pests sought to be controlled and eradicated (Carson, 2001). The impact of Dr. Carson's publication led to a second scientific study on the environmental state of the planet, leading to the publication in 1972 of "Limits to Growth: Report for the Club of Rome Project" (Meadows, 1977).

At the same time the strong questioning towards science and technology as a way of solving all environmental problems (Feyerabend, 1981) also generated an intense and controversial discussion about the need for an education that would make citizens aware of the care required by the environment in which we all live. However, it is not until the meeting of the "International Coordinating Council of the Program on Man and the Biosphere" is held, convened by FAO, WHO and UNESCO, that for the first time "Environmental education" (UNESCO, 1971) is defined as that education that must:

> *"… provide knowledge in natural and social sciences necessary for the rational use and conservation of the biosphere resources, and for the improvement of the global relationship between man and the environment, as well as predict the consequences of today's actions on the world of tomorrow and increase human ability to effectively order the natural resources of the biosphere".*

Even then it lasted until 1987 when the World Commission on the Environment and Development of the United Nations defined the concept of Sustainable Development as part of the Brundtland Report (UN, 1987):

> *"The development that meets the needs of the present generation without compromising the ability of future generations to meet their own needs",* establishing in 1972, during the United Nations Conference on the Human Environment (UN, 1972), June 5 as the World Day of the Environment.

In this framework, the integration of ecosystems at global level and planetary environmental interdependence were fully demonstrated based on the research by Molina and Rowland (1974). They proofed that the use of chlorofluorocarbon compounds (CFCs), are causatively responsible for the formation of the hole of the ozone layer in the atmosphere. Farman et al. (1985) stated that as a consequence of its release to the environment, it already existed on the Antarctic, causing for the first time, voices of alarm regarding its effects and capacity to generate a global climate change.

After another nearly two decades, UNESCO in 2002 established the integral relationship that exists between education and the advancement of science for the achievement of development (Reyes-Sánchez, 2018). Recognizing that there was no real advance in the perception of people for those topics worldwide but there was an urgent global educational effort to educate, UN in 2002, declared 2005-2014 The decade of education for sustainable development: *There is no possibility of achieving planetary sustainability if there is no educational change* (UN, 2002).

Science and its practice, as well as its teaching had to change

Chemistry is directly related to the Rio Declaration (UN, 1992) on environment and development, which in principle *proclaims that human beings are at the center of concerns related to sustainable development. Human beings have the right to a healthy and productive life in harmony with nature.*

Meanwhile, SDG 9: to build resilient infrastructure, promote inclusive and sustainable industrialization, and foster innovation; it proposes “By 2030, upgrade infrastructure and retrofit industries to make them sustainable, with increased resource-use efficiency and greater adoption of clean and environmentally sound technologies and industrial processes”. Drawing our attention to the fact that the contamination and pollution of water, air, and soil because of the *“inadequate management of chemical, industrial, mining, and agricultural activities contribute, as well as the absence of waste management practices”* are the principal causes of the severe impact on the whole biological flora and fauna: terrestrial and aquatic, affecting planetary biodiversity and therefore the balance of terrestrial and aquatic ecosystems (UN, 2015).

Before “A Silent Spring”, all kind of residues from chemical processes were released directly into the air, water, and soil. The limited biotic capacity of planet Earth was perceived as unlimited to absorb the transformations of the biosphere caused by man (Gómez and Reyes-Sánchez, 2004). But after “A Silent Spring”, the world knew that the problem with chemicals released into the environment is not only their toxicity but also their ability to persist in organisms by accumulation in their fatty tissues (bioaccumulation) until reaching levels intolerable to most living beings. The public controversy generated by Dr. Carson’s book made evident interrelations between all living organisms, initiating a cascade of intellectual and humanist reactions worldwide; triggering the emergence of environmental awareness that led to the United Nations Conference on the Human Environment, held in Stockholm in 1972. Over the years and for half a century, a set of environmental actions was announced and supported at institutional, governmental and O.N.G’s levels in favor of respect for life on the planet.

Consequently, in 1990 the United States Environmental Protection Agency (EPA) began actions to prevent the generation of pollutants through various means such as engineering controls, inventory control, process optimization, and the development of a new concept for the generation of processes and chemical products giving origin to the Green Chemistry.

The Pollution Prevention Law can be considered the first precursor of green chemistry, while in Europe, it is the REACH regulation: Registration, Evaluation, Authorization, and Restriction of chemical substances.

An intelligent person solves a problem. A wise person avoids it

Albert Einstein

Day by day, awareness increases about the need to produce and process food and raw materials that natural resources offer us, generate our inputs, and obtain the satisfiers that the market demands through environmentally friendly processes: allowing us to enjoy them without risk of life (Reyes-Sánchez, 2006b).

One of the possible paths towards achieving these environmentally friendly processes and products is the practice of one chemistry that allows us to seek routes and carry out chemical processes, that reduce the negative impact that has been carried out so far on the environment: this is a fundamental premise of sustainable development, and it is in this sense that green chemistry is a natural way from which to start to contribute to planetary viability. (Reyes-Sánchez, 2012).

Therefore, Green chemistry is directly related to sustainability and the protection of natural resources: air, water, and fertile soil as indispensable resources to preserve biodiversity and hence the environmental equilibrium. The approaches of green chemistry are regulated by a PROTOCOL consisting of 12 principles (*Table 1*).

1.	**Prevention:** it is better to prevent the generation of waste than to treat it or eliminate it after it has been generated.
2.	**Atomic economy:** synthesis methods must be designed in such a way, that they are incorporated to the maximum, in the final product, all the substrates used during the process.
3.	**Less dangerous chemical synthesis:** synthesis methods must be designed to use and generate substances that have low or no toxicity, both for humans and the environment.
4.	**Safe Chemicals Design:** Chemicals shall be designed in a way that maintains their efficacy and low toxicity.
5.	**Use of safe or auxiliary solvents:** avoid the use of auxiliary substances such as solvents, separation reagents, etc., and if they are used, they should be as harmless as possible.
6.	**Design of energy efficiency:** energy requirements in a chemical process are classified by their economic and environmental impact. Therefore, it is suggested to carry out the synthesis methods at ambient temperature and pressure.
7.	**Use of renewable raw materials:** the raw material should preferably be renewable rather than exhaustible, provided it is technically and economically feasible.
8.	**Reduce derivatives:** avoid the use of blocking, protection-deprotection groups, or the temporary modification of the physicochemical processes, because their use requires additional reagents and generates waste.
9.	**Catalysis:** consider the use of catalysts, as selective as possible, preferably of natural origin
10.	**Design biodegradable substances:** products must be designed in such a way, that at the end of their useful life, they do not persist in the environment.
11.	**Real-time analysis to prevent contamination:** will be developed the necessary analytical methodologies at the time of the process, which will allow real-time monitoring and control of the process before the formation of dangerous substances.
12.	**Safe chemicals to prevent accidents:** the substances and the form of a substance used in a chemical process should be chosen to reduce the risk of chemical accidents, including emanations, explosions, and fires.

Table 1. Green Chemistry principles (Adapted from Morales et al. 2011).

Green Chemistry emerges as a work philosophy whose primary objective is to prevent pollution rather than remedy it (Anastas and Warner, 1998). It proposes the realization of a collective effort to reduce to a minimum, or if possible, to eliminate contamination from its origin by carrying out a set of actions such as: *1)* using "clean" processes, *2)* avoiding waste, or *3)* indiscriminate use of non-renewable raw materials, *4)* avoid the use of toxic or polluting materials in the production of chemical products, *5)* design chemical products, and processes that do not harm species' health, and *6)* recycle the waste generated to minimize impacts on the environment protecting biodiversity and human beings, without sacrificing scientific and technological advance.

Precaution is the first (1st) of its principles and proposes: to incorporate as much as possible all the reagents so as *not to generate waste* (2nd), design *substances of low or no toxicity* (3rd), create *processes and substances that are safe* (4th), avoid the use of solvents, *and if necessary to use the safest* (5th), *efficient use of energy* by conducting reactions at environmental conditions and *avoiding the use of fossil fuels* (6th), *use of renewable raw materials* instead of nonrenewable (7th), reduce subproducts to *avoid waste* (8th), use *natural selective catalysts* to activate the reactions instead of contaminating acids, bases or solvents (9th), *design biodegradable substances* (10th), *follow up and control in real time* the processes *to prevent the formation of hazardous substances* (11th) and *design safe chemicals* to prevent accidents (12th) (Reyes-Sánchez, 2012 and 2018).

Through those 12 principles, Green Chemistry seeks to prevent pollution before it occurs by designing products and processes with less impact on the environment and humans. Indeed, this being the point where it converges with the precautionary principle, which is also the pillar of sustainability.

And what does green chemistry have to do with soil sustainability and preservation of its biodiversity?

A) The soil resource and its catalytic capacity in green chemical reactions:

All chemical reaction requires an activation energy expenditure to be carried out, and when it is very large, it is possible to use substances that reduce energy expenditure and, at the same time, the reaction time. The compound capable of reducing the energy expenditure of a reaction, accelerating it, is called a catalyst, as it does not react with the reactants and will not be transformed into one of the products. The catalysts can be homogeneous, heterogeneous, electrocatalysts and biological catalysts (enzymes). However, most chemical catalysts are toxic, so they require treatment for their elimination in order to avoid contamination, which entails waste of energy and generation of wastewater.

In the traditional practice of chemistry, conventional catalysts used in many organic reactions to produce agrochemicals, pharmaceuticals, cosmetic products, etc., tend to be acid-corrosive, highly toxic, difficult to handle, and therefore, of difficult final disposition (Anastas, 1998).

Green chemistry requires the design and use of new catalysts of natural origin and catalytic systems that simultaneously achieve the dual objectives of environmental protection and economic benefit, as its most important fundamental pillars (Anastas, 2001 and 2007).

Catalysis offers numerous benefits to the scientific practice of Green Chemistry, and therefore for the sustainability of the soil resource and the protection of its biodiversity. Among those benefits are the reduction of energy requirements, the use of materials in catalytic quantities stand out instead of stoichiometric amounts, the increase in selectivity within a reaction, decrease in the use of separation substances, and a reduction of the use of less toxic materials (Morales et al. 2011).

> *For this reason, the use of supported catalysts as well as the use of natural catalysts, are an environmentally desirable alternative in these moments of environmental crisis, with severe deterioration of natural resources, water, soil and its biodiversity.*

The use of natural catalysts in green chemistry has promoted over the last two decades, the generation of basic research to develop processes that reduce the negative impact has been exerted on the environment and human health (Reyes-Sánchez, 2006a). It has also favored that the scientist reflects and changes his way of thinking and acting toward the environment by practicing a socially responsible science: introducing the necessary modifications to minimize and avoid the generation of waste using natural catalysts such as clays present in the soil (Morales et al. 2011 and Reyes-Sánchez, 2012b).

B) The role of the soil's clay as a green catalyst

In soil science, the term clay has three different meanings. As a granulometric fraction, as a textural class, and as a mineral. As a granulometric fraction, it refers to particles whose diameter is less than 2 µm, regardless of their mineralogical composition. As a textural class, it refers to a type of soil texture in which there is a predominance of particles of that size. As clay minerals, we speak of phyllosilicate compounds and non-lamellar silicates such as allophane (Porta et al. 2013). Phyllosilicate compounds are built of two different three-dimensional chemical structures distributed in layers.

Layers of tetrahedra (T layer) joined by one of their vertices and in the center of which is the element Si, forming four covalent bonds with four oxygen atoms whose orbitals are directed towards the four vertices, resulting in a crystalline structure (*Figure 1*).

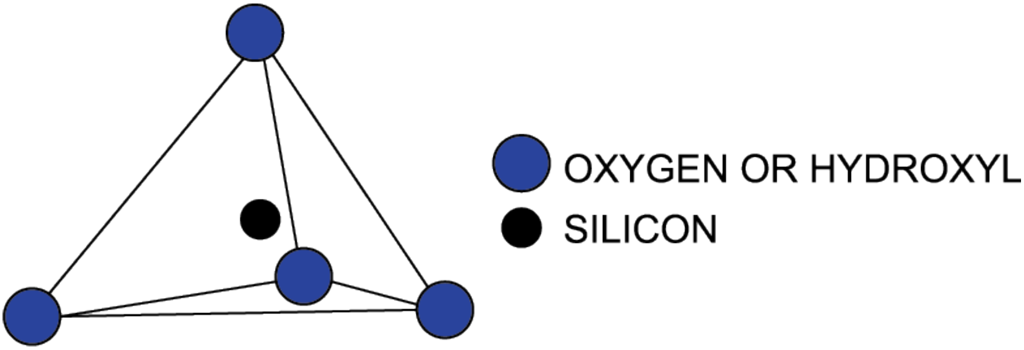

Figure 1. Silicon tetrahedra

Octahedron layers (O layer) in the center of which is the aluminum element, forming covalent bonds with oxygen or hydroxyl (Fig. 2).

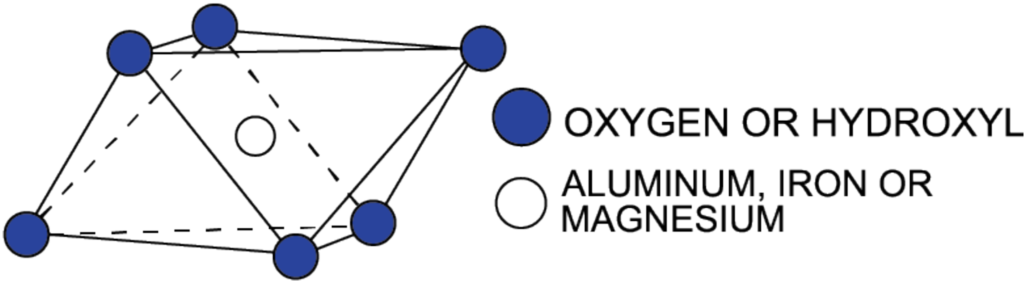

Figure 2. Aluminum Octahedron

Phyllosilicates are classified according to the three-dimensional stereochemical structure of their clays defining the soil matrix:

Clays 1:1 or T-O layers are builds by a layer of tetrahedra joined by the vertices to a layer of octahedra.

Clays 2:1 or T-O-T layers are conformed by two tetrahedral layers joined by one octahedra layer in the middle of both.

Thus, soils as three-phase systems constitute a fundamental matrix with limited resilience and sustainability conditions to offer these soils physical, chemical, and biological properties and functions (Horn and Blum, 2020) to achieve the sustainable development goals (UN, 2015). The difference in electrostatic forces existing between the elements that make up the 2: 1 clay is essential both for the formation of that fundamental soil matrix, as well as for its stability and cohesion to resist both the mechanical effects of the rain and wind, as well as the derived from agricultural practices (Horn, 2021), and are also responsible for their chemical and catalytic behavior.

Among the main characteristics of clays 2:1 are the following: *a)* they are chemical compounds that form small crystalline structures (2 µm ø), *b)* they can exchange ions, *c)* they are capable of varying their internal chemical composition, *d)* they can host molecules within their interlaminar space, *e)* the surface of the sheets shows considerable chemical activity and *f)* the physical changes of the laminar structure, for example, its expansion is reversible. This ability of clays to expand and contract in response to the water content in the soil is a determining factor in the formation of the granular structure of agricultural soils (Porta, 2013; Blum, 2018), the conformation of pore spaces, and their percentage, defining accordingly soil permeability of air and water, the penetration of the roots of the plants, and the conditions of existence of the soil´s micro and macrobiota (Horn, 2021).

One way to protect the soil resource, its fertility, biodiversity, and its various functions and ecosystem services is the use of its 2:1 clay as catalysts for carrying out green chemical processes in the absence of solvent or using water or CO_2 as such, using alternative energy sources to the use of fossil fuels (microwave, infrared, ultrasound, mechanochemistry), with shorter reaction times and a significant reduction in by-products to be disposed of or released to the environment (or none if possible).

An extraordinary property of these structures is their possibility of being able to replace the silicon atom (Si^{+4}), located in the center of the tetrahedra, with another with a lower charge, such as aluminum (Al^{+3}): generating a deficiency of positive charge or an excess of negative charge in the tetrahedron, which needs to attract other cations to compensate, for example, Na^{+}, K^{+}, Ca^{+2}, etc.

At the same time, the substitution of aluminum and silicon by a cation with a lower electrical charge, such as Mg^{+2}, causes a deficiency in electrical charge that is responsible for the most important surface properties of clays: their exchange capacity, their absorption capacity of polar molecules such as water, and of organic compounds that, when introduced into the interlaminar space, cause the swelling of the laminar structure of clays 2:1.

> *That is also the mechanism that allows silicates to have unique acidity and ion exchange surface properties, so important that the catalytic properties of clays are due to them.*

That is why the 2:1 clay of the montmorillonite type is a heterogeneous catalyst of natural origin and reusable, of low cost, and widely used in the Green Chemical industry for the synthesis of organic products of great human benefit and commercial value.

Its catalytic capacity comes from the chemical properties of the before mentioned clay minerals: surface electrical charge formation capacity, ion exchange capacity, hydration capacity, its high contact surface, and adsorption, all of which provide a great capacity as support and a high number of active sites with acidic or basic Lewis or Brönsted-Lowry characteristics that allow it to activate the reactions.

Examples about how green chemistry can help to improve environmental conditions by reducing the negative impact on natural resources, water, and air, as well as on the soil and its biodiversity, without giving up obtaining the chemical products that constitute our current way of life, is the production of biofuels (Guerrero et al. 2011), aspirin synthesis (Morales et al. 2011), and different organic compounds reported by Miranda, R. R. et al. (2003) using a Mexican bentonite clay with a catalytic character that is commercially known as Tonsil Actisil FF (TAFF). (*Figure 3*).

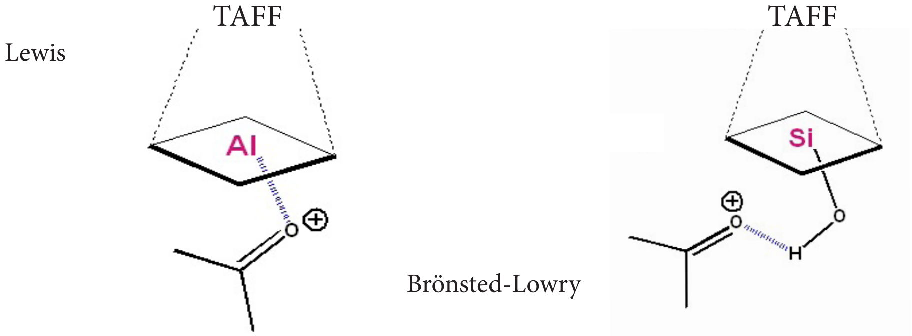

Figure 3. Enhancement of the electrophylic character of a carbonylic substrate by means of TAFF: L as a Lewis and BL as acidic Brönsted-Löwry catalyst. (Adapted from Miranda et al. 2011).

C) Green chemistry for remediation of contaminated soils

An alternative for the remediation of soils contaminated with heavy metals with Cr, Pb, Ni, Hg, Cd is using iron-based nanoparticles and zero-valent iron (Cundy et al. 2008, Kamran et al. 2019). That is why, knowing that heavy metals persist indefinitely in the soil matrix, altering the soil-flora-fauna balance, and ultimately affecting human and animal health through bioaccumulation processes, the use of iron-based nanoparticles, and zero-valent iron (ZVI), represents an alternative to remove or stabilize of heavy metals (Colombo et al. 2017, Urooj et al. 2019).

These processes can be carried out by nanoparticles with adsorption capacity of heavy metals presents on the surface or inside the soil matrix, or by nanoparticles that trigger oxidation, degradation, transformation, and complete mineralization reactions of these pollutants (Cundy et al. 2008) through reactions of precipitation-dilution, acid-base, redox, ion-exchange, or photocatalysis. Iron nanomaterials can be obtained naturally from minerals found in rocks and soil, as well as through synthetic methods (Lagroix, et al. 2016).

Examples of using NPs for remediation of contaminated soils

Stabilizing heavy metals

An example of the application of green chemistry to the preservation of the soil and its biodiversity is the use of nanostructured systems based on bio-based iron nanomaterials (BioFeNMs), to obtain nanostructures for the remediation of contaminated soils. Those processes are carried out at a temperature and pressure close to environmental conditions, and starting from molecules obtained from plant extracts like carbohydrates, flavonoids, polyphenols, alkaloids, etc. (Karn et al. 2009, Wang et al. 2015).

Thus, the use of vegetal biomass offers a great diversity of species and tissues to be used and molecules to be obtained from them, but polyphenols are one of the most efficient biomolecules to carry out reduction and stabilization of FeNMs. They are easily obtained from agro-industrial waste such as sugarcane bagasse and tea residues. (Kamran et al. 2020, Colombo et al. 2017). Its effectiveness is due to the nanometric scale of FeNMs as it not only allows them to be mobile within the soil matrix but also to interact closely with the pollutants present in it. Their transformation products can be assimilated into the soil environment by some microorganisms and plants since iron is an essential micronutrient. Contaminants that can be degraded by oxidation-reduction by monovalent iron or superficially adsorbed by oxides such as magnetite: Fe_3O_4 and hematite: Fe_2O_3 that can be extracted by applying a magnetic field (Kamran et al. 2020, Kadhum, et al. 2020).

Iron is an element present at the nanoscale in the soil matrix in the form of oxides and hydroxides nanomaterials covered by humified substances and other organic molecules such as carbohydrates, metabolites, etc. (Lagroix, et al. 2016, Colombo et al. 2017). That is why the obtaining of nanomaterials based on Fe (II) and Fe (III) species, and zero-valent Fe (FeNMs), stabilized by aqueous extracts of biomolecules in low concentrations is considered not to alter the biogeochemical cycle of Fe or soil health. Notwithstanding the heavy metals toxicity is very high, those can only affect living beings if they are bioavailable. Therefore, one of the most significant applications of iron nanoparticles FeNMs and zero-valent Fe (ZVI) is that they can change the oxide-reduction state of heavy metals, affecting their bioavailability for the benefit of the entire food chain and facilitating their removal from the soil, which is in the interest of the trophic chain.

During the decontamination process, reactions between the heavy metal and the zero-Valent iron (ZVI) depend on the standard redox potential (E^o) of the heavy metal to be removed. Cr, for example, is a toxic and carcinogenic element that, due to various industrial activities, can be found both in water and in soil solutions in its oxidized form as Cr (VI), which can be removed as Cr (III) by reducing agents such as Fe_2SO_4, SO_2 or $NaHSO_3$.

An environmentally friendly alternative is its reduction by Fe (II) nanoparticles:

$$Cr_{(VI)} + 3Fe_{(II)} \longrightarrow Cr_{(III)} + Fe_{(III)}$$

Stabilizing organochlorine compounds

The removal of pollutants by fuels spill or leak is another example of soil contamination with serious consequences on its physical, chemical, and biological properties, as well as for human health. Their removal is difficult and expensive, so one possibility is the reduction of polychlorinated hydrocarbons by (ZVI) nanoparticles.

In the general dechlorination reaction of halogenated hydrocarbons, the zero-valent iron acts as a reducing agent, releasing chloride ions.

$$C_x H_y Cl_z + 2H^+ + 2Fe^0 \longrightarrow C_x H_{y+z} + 2Fe^{2+} + 2Cl^-$$

However, the prime source of soil and water contamination by organochlorine compounds comes from the biocides used in agriculture -affecting plants, animals, and soil biodiversity-, not by fuels spill or leaks.

Organochlorine compounds used to control pests and diseases not only act on a single species or organism but affect all other non-target organisms, provoking deleterious or fatal consequences when they are exposed to those chemicals. They are high-spectrum compounds with high chemical stability, highly fat-soluble, and water-insoluble. That is why they accumulate in the fatty tissues of living beings, producing long-term damage to health that also affects humans through its bioaccumulation.

Organochlorine compounds are neurotoxic that act directly on the nervous system of arthropods, notwithstanding also affect other vertebrate animals (Driver, 1991 and Gyldenkærne, 2000), and indirectly, humans ingest them as residues in agricultural or animal products: milk and meat mainly (Den Hond and Shoeters, 2006).

Thus, the dechlorination reaction of halogenated hydrocarbons by the zero-valent iron nanoparticles (ZVI) is a green chemistry alternative to protect the soil biodiversity and health of living beings.

What do these examples mean for soil and environment preservation?

These are examples of how green chemistry uses the iron nanoparticles present in soils to synthesize iron bio-based iron nanomaterials (BioFeNMs) as an alternative to remove or stabilize heavy metals pollutants from it to protect all the trophic chain.

In these processes, green chemistry acts according to the prevention principle (1) to remove or stabilize heavy metals which are highly toxic to plants, animals, and humans. Using vegetal biomass as renewable raw material (principles 7 and 10) to synthesize BioFeNMs as safe, efficacy, and low toxicity chemical (Principle 4), carrying out processes at a temperature and pressure close to environmental conditions (Principle 6).

According to principles 5 and 12: synthesis of FeNMs is based on Fe (II) and Fe (III) species, and zero-valent Fe (ZVI) stabilized by aqueous extracts of biomolecules in low concentrations to not alter the biogeochemical cycle of Fe or the soil health as a guarantee of soil biodiversity.

In the second example, FeNMs are used to remove chlorine from halogenated hydrocarbons, but the principles of green chemistry applied are the same. However, in both processes, FeNMs works as selective natural catalysts (Principle 9) as a function of the standard redox potential (E^o) of the element to be removed or stabilized, looking less impact on the environment and human health.

Green chemistry and sustainability

However, not every green chemical process is necessarily sustainable. Green chemistry is an advance in this necessary transition, and in the hands of scientists and teachers of all sciences, it is the challenge of approaching the ideal of socially responsible chemistry that contributes to the achievement of sustainable development by protecting soil resource and their biodiversity. Reaching this implies that the scientific community assumes in their practice both the philosophy of green chemistry and the philosophy and basic principles of sustainability (shown in *Table 2*) to accordant to Lubchenco (1998), “reorient its machinery” to make the XXI century the century of the environment.

1.- The recognition *of the limits of growth that nature imposes on us.*	
2.- The precautionary principle.	6.- The principle of interdependence.
3.- The uncertainty principle.	7.- The principle of interdisciplinarity.
4.- The principle of incompleteness.	8.- The articulation principle.
5.- The principle of integration.	9.- The principle of social equity.

Table 2. Basic principles of sustainability. (According to Reyes-Sánchez, 2012b).

Green Chemistry and sustainability are therefore philosophies that share the precautionary principle, but at the same time differ in that *precaution is not the only purpose of sustainability, nor is this the objective of all its principles* (Reyes-Sánchez. 2012b). Both philosophies seek to prevent pollution from its origin instead of spending time, money, and effort to decontaminate and "recover" the air, water, or soil that were previously degraded. However, they differ in that precaution is not the only purpose of sustainability, nor is this the objective of all its principles.

In this regard, green chemistry protects the soil resource and its biodiversity by ***a)*** using natural catalysts to reduce the energy expenditure involved in carrying reactions, ***b)*** minimizing both the use of acids, bases, and solvents as well as the generation and discharge of wastes ***c)*** avoiding produce or use toxic, hazardous, or harmful substances to reduce their negative impacts on soil and water ***d)*** using renewable raw materials in reactions and processes ***e)*** synthesizing and using biodegradable substances and ***f)*** carrying out processes at a temperature and pressure close to environmental conditions; all this to guarantee their physical, chemical, and biological equilibrium.

The soil is a complex, open, and dynamic natural system that presents a wide variety of interactions and interdependencies of a physical, chemical, and biological nature (Horn and Blum 2020) which support fundamental aspects of life on planet earth, such as the balance in biogeochemical cycles associated with adequate agricultural production, and sustainability of the soil resource to preserve the balance of all its properties, functions, and capacities. Its sustainability depends on our ability to preserve soil and its biodiversity as a guarantee of its fertility to sustain the production of food, natural products, and ecosystem services that are indispensable for human life. But to preserve soil and its biodiversity also depends on our ability to produce energy, food, and chemicals without compromising environmental balance and human health, so our ability to develop a science practice that allows us to achieve this is not only a challenge but a responsibility.

That is why the interdisciplinary development of sciences must address sustainability practicing of environmentally friendly, economically profitable, and socially responsible processes as an alternative that brings us closer to achieving SDG 15.

Concluding remarks

Because of its transcendental importance to preserve life on Earth, Soil Science, cannot, and should not ignore other disciplines but interact with them.

Green chemistry is a work philosophy totally in line with the approaches of the achievement of the SDGs, and therefore an ally to join the team to work interdisciplinary in the same direction, and add it equally to the work of the IUSS International Decade of Soils and the FAO Global Soil Partnership.

Therefore, to identify common points, to build interdisciplinary partnerships with other sciences and to work as a team and in the same direction should be one of the tasks to dedicate time and effort by all in the IUSS along the "International Decade of the Soils", to prevent contamination from its origin instead of spending later time and effort to decontaminate the air, water or soil that had previously degraded; focusing on common points, to visualize new perspectives of collaboration and learn to listen to the others, in order to tackle environmental challenges from multiple angles.

From the ideal of sustainability, it is essential not only to seek less polluting and more efficient raw materials and processes but to practice and teach sciences to build an education that advocates for the search of alternatives *socially viable, ethically acceptable, economically profitable, energetically desirable, and environmentally friendly* (Reyes-Sánchez, 2012b) to achieve the 17 SDG's.

In this regard, educating children and young people in the interdisciplinary knowledge of the science and environmental values to preserve natural resources is a moral responsibility that must be manifested in the daily commitment of scientists to the future of humanity.

For the IUSS scientists, educating in soil sciences in an interdisciplinary and consciousness-raising way should not only be a commitment but an obligation that must be present in their scientific practice to provide their students not only knowledge but also teaching with their the example, new forms of coexistence between human beings and to relate all to nature.

Acknowledgements

I sincerely thank UNAM, AAPAUNAM, and the green chemistry working group of FES-Cuautitlán for their support.

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Chapter 20

Music and theater for soil science education in schools

Patrick Lavelle and Elena Velásquez

Introduction

For at least 40 years, maybe more, ecologists have described the negative impact of human activities on the Earth environment, in all its components. Soil scientists have made their contribution to this communication. Each time more precise and refined, the models predicted degradations which soon after were verified and even sometimes appeared underestimated (MEA, 2005; IPCC, 2014; FAO, 2019), in almost general indifference.

Humanity looks elsewhere, guided by the socio-economic models that represent its global project. A project more than 3 centuries old, that of progress imagined in the movement born in the "siècle des lumières".

This modernity project that has considerably increased the well-being of most people on Earth until recently, began in the 18th century. Developed by Descartes, Kant and many other great philosophers, this project separated the spiritual and material domains, leading humans to posit individual well-being as an objective of life (Goswani et al. 1995). Since that time, science and technology, supported by a capitalist economic system, have allowed enormous progress. Slowly on the scale of successive generations, this model has improved the lives of humans, overcoming the most brutal forms of slavery, great epidemics, famines and even wars.

Today, this model has reached its limits, illustrated by the current great environmental crisis. Undefined economic growth and the consumption that supports it are no longer compatible with the level of resources available on this planet.

Science has played a fundamental role in this progress with technologies and great advances in biology. The only science behind, Ecology is struggling to play its societal role. Based on values long ignored by the dynamic of progress, it barely emerges at the most critical moment in this history.

Soil is a basic part of this process for the wide variety of ecosystem services it produces. It is now clearly established that sustainable food production, carbon storage and the functions of infiltration, storage, transfer and purification of water can only be achieved in a sustainable manner in healthy soils (MEA, 2005).

The progress of successive Conferences of Parties, which is still insufficient, will only be accelerated with the support of conscious and proactive societies. The priority is therefore to create in young people a well-informed environmental conscience and to give them the elements to develop the new human project that no one at the moment is able to draw with any precision.

Here we present some ideas for giving soil a place in the educational process and explore ways to achieve it.

Children love the soil and earthworms

Don’t touch the soil!! It’s dirty! Don’t play with the ants! They will sting you! Take off your boots, you will put mud everywhere! The perception of the soil generally begins at home with these remonstrances from parents who, very early on, convey the idea that soil is a dirty element inhabited by dangerous organisms for our health. The earthworm is sticky, the spider dangerous, microbes infectious. Tractors equipped with impressive devices transform a messy grassland into a nice carpet. Harvested hay is soon wrapped in thick green plastic bags. This field transformed into a short green carpet and the vegetation wrapped in plastic reflect our vision of order and cleanliness that everyone strives to implement, from the small suburban garden to the vast agricultural spaces where trees and hedges have been gradually banned.

The child will learn the names of African mammals, will play with dinosaurs and even unicorns, but no one will show him the Lithobiidae centipedes, Isopoda or Geophilidae millipedes which are not lacking in attraction if we take the trouble to observe them.

At school, he will not be taught soil. An analysis of primary and secondary education programs in France, for example, shows that even if the strengthening of the teaching of environmental themes has been proposed since 2020, only 20% of school teachers declare that they cover all areas of current Science and Technology (Académie des Sciences, 2020)

In contrast, 85% of students say they like scientific experimentation and thus discover the world around them, acquire an integral vision of the scientific approach and use it to understand the environmental issues present.

Supports for education of children to soil science are now proposed by a large number of associations and institutes. See for example the soil safari programme of the Soil Science Society of America (2015) and of the British Soil Science Society (2021) for primary schools, education supports of the French Association pour l´Etude des Sols, (AFES, 2021) and many others that we cannot list here.

We present here two different experiences we have had in South America, one with children from a remote rural place in the Transamazonia highway region of northern Brazil, the other one in the urban area of Cali, a big city of Colombia. The first one uses simplified scientific protocols and drawings to illustrate the abundance and role of soil macro invertebrates. However, scientific message alone may not be sufficient. In the second experience, we used a bit of theater and music to reinforce the messages, with original songs dedicated to soil and environmental issues. While it certainly reaches our reason, art is necessary to reach our heart and turn us creative and enthusiastic to create a new world for Humans, urgently. In the second experience, we used a bit of theater and music to reinforce the messages, with original songs dedicated to soil and environmental issues.

A seminar at Pacaja, km 338 of the trans-Amazonian road, Brazil

The AMAZ project proposed a complete diagnosis of the landscapes, social, economic, biodiversity and ecosystem services of soils in deforested Amazonian sites and their dynamics between 10 and 80 years after deforestation (Lavelle et al. 2016, Grimaldi et al. 2014). Seminars were offered to children to make a rapid diagnosis of the condition of the soil and its macrofauna. Parents often joined the children, taking advantage of the scientific animation in this very isolated place, linked to the Trans-Amazonian by a precarious 70 km track.

Figure 1: One day with the school children of Pacaja and their parents, km 338 of the Transamazonian highway: 1. Sampling of two blocks of soil 25x25x20 cm in a pasture and in a forest; 2. Sorting of fauna and visual separation of macroaggregates produced by roots, earthworms and physical processes, and organic debris; 3 and 4: Identification, drawing of the invertebrates encountered; 5. Comparison of sampled stands, macro-aggregation and organic resources between forest and pasture; 6. Experimentation of the effect of soil aggregation and the presence of organic matter on the infiltration and storage of water in the soil. Here with false plasticine aggregates, and a treatment with small pieces of toilet paper simulating organic matter. In other presentations, we use the extracted earthworm macroaggregates vs. non aggregated soil as treatments.

Science, theater and music in the urban area of Cali, Colombia

Environmental education seminars are offered to colleges in Cali with video or live supports of the “Cantándole al Planeta” series (Hijos de Gaia, 2020) . An activity on soils is proposed, after the presentation of one of the videos 3 that is dedicated to soils and Agriculture. In these videos presented by teacher Ana, planet Earth tells her history and current problems, the exotic French scientist Patrick provides the scientific background, and the music band “Hijos de Gaia” illustrates the topics with thematic songs.

Figure 2: soil activity in a college of the urban area of Cali, Colombia. Looking for earthworms and other soil macroinvertebrates and separating soil macroaggregates produced by earthworm activity in a tropical grassland soil.

Learning more about the earthworms

The soil is alive, there are many and various invertebrates. The children sort the soil and observe and count the invertebrates found. A simple experiment illustrates the role of soil aggregation for water infiltration and retention.

How many earthworms are there in the soil of a football field?

Students extract earthworms from 10x10x10 cm blocks taken with a metallic frame from a nearby site (*Figure 2*). They are kept alive in a jar with a little water. Once counted and congratulated the group which has found the largest number, a rule of three makes it possible to extrapolate the averages obtained to the surface of a football field. On this field which measures 6830m^2 by law, we would find between 1 and 2 million individuals, sometimes up to 3. Astonishing result, isn’t it?

Worms build the soil and enhance the water cycle

These earthworms, in particular, by rejecting the digested soil, leave droppings in the soil, which will be called castings. Once deposited and slightly dried, they become solid and rounded structures that can be easily separated from the rest of the soil. A Brazilian child from the Amazonian site of Pacaja, did say that these are the bricks with which the worm builds its house in the ground. Worms create habitats!!

How much have they digested and left in the soil? We separate these structures and weigh them. They can represent half the weight of the soil of this block.

After having observed them, noted their solidity, their regular shapes and their dark color which indicates a fairly high content of organic matter and natural fertilizing elements, a simple experiment will allow to better appreciate their value for the physical processes of the soil.

We have two plastic bottles. The upper part cut, and returned above the lower part of the bottle constitutes a funnel. A cone of screen mesh is placed at the bottom of the funnel and one is filled with worm castings separated from the soil, the other with non-macro aggregated soil. We pour the same quantity of water into each funnel and note that, on the side of the castings, the water infiltrates quickly and comes out clean; in the other treatment, it infiltrates much slower and comes out turbid. We can then discuss the significance of this result for soil and water management.

And we end with a song…

These simple experiments can be accompanied by a song that talks about the soil, and the chorus of which will be song by the participants. The song Amazonia: a happy soil, features an Amazonian soil that tells its story, before and after deforestation, the effect of the arrival of an invasive species of earthworm after the fire has destroyed a part of fauna, the social problematic linked to land tenure and the impact of intensive agriculture, ending with the proposal of agroecological solutions (Hijos de Gaia, 2011).

Amazonia: a happy Soil

I was a happy soil of the Amazon
Colorful, fertile, swarming with life
Shaded trees and under dead leaves
Drank up the rain and fed on debris

All forms of life like a symphony
Playing happily night and day
Sheltering villages, tunnels and cathedrals
The harmony of life in all of its tiers

Chorus: Amazonia nurse your life, Amazonia nurse your soil (bis)

Earthworms and termites, ants and roots
Lively burrowed chambers and galleries

Listless bacteria their beauty asleep
Peacefully bathed in many small ponds

Hundreds of sweet hairy roots Secretely entwined networks of trees
Green deep sea in the planet´s blue
Storing its treasures and healing its wounds

Chorus: Amazonia nurse your life, Amazonia nurse your soil

One deadly day shivered the Earth
Slashed and scorched many a tree
A carpet of ashes and many charred stumps
Broke my heart and tore me with sadness

I´m compacted, dried, burned and drawn
Fauna fled their nests on fire
Springtails dead centipedes all gone
Sleeping beauties suddenly became old

Chorus: Amazonia nurse your life, Amazonia nurse your soil

Indigenous people displaced or killed
Nature crushed and climate changed
Earthworms invaded from faraway lands
Graceless and clumsy, working alone

They compacted me, my color I lost
Gray and stinky, my flowers have died
Rain slips away, my strength is gone
Lost my avenues, towers and galleries

Chorus: Amazonia nurse your life, Amazonia nurse your soil

Tractors arrived with their iron plows
Crushed my skin and tore out my guts
Force fed chemicals pesticides
Sick with excess I vomit to rivers and air

Soybeans I feed to pigs in China
Palm oil I grow for their western cars
Foreign banks all interested of course
My destroyed landscapes don´t run them away

Chorus: Amazonia nurse your life, Amazonia nurse your soil
Re-plant my trees and nourish me
Don´t feed me poisons but manure and leaves

Goodbye to your tractors, leave me alone
Allow me to recreate the magic of life

Cocoa and maize, peas and fruits
Wise peasants who understand me
Climate I´ll fix and rivers I´ll clean
No more emissions of toxic gases

Chorus: Amazonia nurse your life, Amazonia nurse your soil

Life back to playing its symphony
Playing happily day and night

This song has important scientific content that can be discussed with the students, and detailed scientific elements.

With a bit of theater, ...

… children´s interest may be even greater. The Planet Earth represented in the video series "Cantándole al Planeta" by a middle-aged lady, good-natured and falsely naive, tells her story (Hijos de Gaia, 2020). She was able to create life, 3.5 billion years ago, after generous comets made of ice and rock covered it with water. She tells of the creation of millions of species, increasingly more perfect, in her workshop. She speaks of the great successes that trees and earthworms have been. The very particular way earthworm reproduces, which alternate sexes and homosexual mating which allows reproduction. This is achieved thanks to the preservation of the sperm in spermathecae, small pouches from which it is extracted when the time comes to fertilize the ova of the animal that has become female.

There is a lot more to be said about the earthworm, apart from these peculiar sexual characteristics: its ability to transform its droppings into solid macroaggregates that fertilize the soil and build its structure. These castings contain hormones produced by the microflora stimulated during intestinal transit, which stimulate plant growth and can even modify expression of their gene, promoting the plant's defense against parasites (Blouin et al. 2013).

The song *Lombricita perdida* (Hijos de Gaia, 2012) tells the story of the earthworm *Pontoscolex corethrurus*, an invasive parthenogenetic species whose life seems boring for a parthenogenetic animal (which "ni amores tenía" … "no had romances"), a life entirely devoted to the production of thousands of castings, stable macroaggregates of the pasture soils where it thrives. In a Mexican ranchera drama, the worm will end up drowned in the bottom of a tequila bottle ...

Planet Earth talks about her invention of the *Homo* self-proclaimed *sapiens* species which poses the worst problems to it (Hijos de Gaia, 2020). She had designed it with great care and expectation. All the latest biological improvements had been used, a high-performance brain, the forelegs free to allow the development of hands. She thought that this animal would help her to better manage her climate and thus avoid another massive extinction which had saddened her so much, five times, in the past. But this animal betrayed her. When a child, he distinguished himself by his propensity to eliminate all the large animals, mammoths, woolly rhinoceros, cave bears and many others which then populated the earth.

Now a teenager, it's worse. He discovered the climatic garbage of the Earth, coal and oil formed in carboniferous from the trunks and roots of trees, slowly transformed and covered with sediment. 200 million years which had made it possible to remove a very large quantity of CO_2 from the atmosphere cooling considerably a temperature which was excessive at that time.

Humans don't treat each other well either. Farmers who produce food often have difficult lives due to a game invented by others, called capitalism which often mistreats them in the name of invented myths such as the law of the market, economic growth or money (Harari, 2014).

However, there are solutions and people who implement them, as in the Star farm of Edilson, a farmer from the Caquetá Colombian department who has transformed a degraded land into an agroecological model collaborating with a scientific project project (Hijos de Gaia, 2012).

The Star Farm

Farm La Esperanza
Was not at all nicel
With many bare grasslands,
And eroded soils
Lean and dull cows,
Did burn in the sun
There were no trees left
To give them any shade bis

Bored flocks of birds
They all had gone
They had no racks to pose
Creeks were all polluted
They had no forest edges
The cattle came in,
And made them dirty

Chorus:

Restore me Edilson, Restore me
Begged the farm
The cows and all the birds
Restore me Edilson, Restore me
I want them to call me the Star farm

He invited scientists
Professors, engineers
All his companions
To help him do the task

A new pasture was sown
Of Kudzu with Brachiaria
On sides of all the creeks
Trees planted on the banks

Rows of legume shrubs
For cattle browsing
Pretty purple yucca
Yopo, cedar and banana

La Esperanza turned to be
The Star farm
Agroecological model
For Amazonia

As a conclusion

Science produces sometimes fascinating stories ... but we most often do not know how to tell them. The observation of an Onychophora invertebrate, an extraordinary intermediary between arthropods and annelids, the social life of termites, quorum sensing phenomena in bacteria, the extraordinary spectacle of thousands of nests of the ant *Camponotus punctulatus* in the abandoned rice fields of northern Argentina, the decisive impact of some companion plants in agricultural systems or the communication of trees through their mycorrhizal networks are all phenomena that should interest people, as much as the line and performance of a newly produced car or any brand new product offered to our compulsive consumption.

Let us tell these stories to artists and ask them to help us translating them into songs, poems and theater dedicated to the beauty and sustainable use of soils (Altieri and Gomez, 2015) and their products (Campo y Sabor, 2013).

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IUSS thanks all the authors for their collaboration and work that allowed the publication of the first book of the IUSS Open Access Book Series within the framework of the International Decade of Soils 2015-2024.

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International Union of Soil Sciences®

ISBN: **979-8-9862451-0-2**

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Decade of Soils
2015-2024