Rock powder solubilization by *Aspergillus niger* as a source of potassium for agroecological systems

Maria Leonor Lopes-Assad\(^a\), Simoni Helena Avansini\(^a\); Greice Erler\(^a\); Márcia Maria Rosa\(^a\); José Ruy Porto de Carvalho\(^b\) and Sandra Regina Ceccato-Antonini\(^a\)

\(^a\)Centro de Ciências Agrárias, Universidade Federal de São Carlos, Araras, SP, Brazil, Email assad@cca.ufscar.br

\(^b\)Embrapa Informática Agropecuária, Campinas, SP, Brazil, Email jruy@cnptia.embrapa.br

Abstract
The objective of this work was to test the potassium (K) solubilization from minerals of an alkaline ultramafic rock powder by two strains (CCT4355 and CCT911) of *Aspergillus niger*. The experiment was carried out in triplicate in 250 mL erlenmeyer flasks over a period of 35 days. Soluble K, titratable acidity and pH were analysed and the solubilization rate (SR) was calculated relative to the total K in the rock powder (2.921 mmol/L). The data obtained showed that both strains of *A. niger* fungus have high potential of K solubilization and could be used as an alternative for small farmers to produce biofertilizers using rock powder.

Key Words
Soil remineralization, K biossolubilization, biofertilizers.

Introduction
The natural fertility of soils is directly connected to the availability and the release of chemical nutrient elements from the available rocks to the soil and it is influenced by pH, grain size, structure, porosity, permeability and organic matter content. Stonemeal practice, based on weathering and soil formation processes, is especially interesting for family farmers who want to conserve the land and wish to redesign the productive system following agroecological principles. Nevertheless, the long period demanded for the alteration of the minerals and the availability of important elements for plant nutrition makes this alternative unfeasible. Reports are found on satisfactory results for potassium (K) and phosphorus (P) solubilization by molds such as *Aspergillus*, *Penicillium* and *Fusarium*; and bacteria such as *Bacillus*, *Pseudomonas* and *Micrococcus* (Gaur 1990). The mould *Aspergillus niger* is known for its ability to solubilise P in rocks due to organic acid production, especially citric acid (Nahas et al. 1990; Vassileva et al. 1998). The use of microorganisms in association with rock powder could be an alternative to small farmers for obtain biofertilizer, using local materials. The objective of this work was to test the K solubilization from minerals of an alkaline ultramafic rock powder by two strains (CCT 4355 and CCT 911) of *Aspergillus niger*.

Materials and methods
The rock powder used was an alkaline ultramafic rock powder from Lages, Santa Catarina, Brazil, with 3.44 dag/kg K\(_2\)O (Barbosa-Filho et al. 2006; Moreira et al. 2006), presenting a grain size between 0.002-0.05 mm. Two *in vitro* experiments were carried out on a laboratory scale. The assays were carried out in triplicate in 250-mL erlenmeyer flasks with 50 mL of medium and 200 mg of rock powder, inoculated with *A. niger* spores (1.6 x 10\(^7\) spores/ml) at 30°C, 160 rpm for 35 days of incubation. The culture medium contained sodium citrate (2.85g/L), NH\(_4\)SO\(_4\) (1g/L), MgSO\(_4\)\(_7\)H\(_2\)O (0.5 g/L), CaCl\(_2\) (0.132 g/L), glucose (10g/L), distilled water (1 L) and pH adjusted to 7.0 with NaOH 0.05 N. Two strains of *Aspergillus niger* were used (CCT4355 and CCT911) and three treatments (T1 = *A. niger* + medium; T2 = rock powder + medium; T3 = rock powder + *A. niger* + medium) were designed. Soluble K was determined by flame emission photometry, pH using a digital pH-meter, and titratable acidity by titration of the samples to pH 7 with 0.05M NaOH solution. The solubilization rate (SR) was calculated relative to the total K concentration in the rock powder.

Results and discussion
Both strains (CCT911 and CCT4355) solubilised K from the rock powder, and the results obtained were significantly different at the 5% level (Table 1 and Figure 1). The behaviour of each strain varied, in such a way that the K concentration, pH, titratable acidity and the SR were significantly different comparing the three treatments (Table 1).
Table 1. Average comparison by Tukey’s test at 5% of significance for potassium concentration (K), pH, titratable acidity (TA) and solubilization rate of potassium (SR) in the culture medium with two strains of *A. niger* (CCT911 and CCT4355), during 35 days, at 30ºC, 160 rpm.

<table>
<thead>
<tr>
<th>Incubation time (days)</th>
<th>K (mmol/L)</th>
<th>pH</th>
<th>TA (mmol/L)</th>
<th>SR (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Strains</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>CCT 911</td>
<td>0.96 a</td>
<td>5.59 a</td>
<td>18.84 a</td>
<td>32.73 a</td>
</tr>
<tr>
<td>CCT 4355</td>
<td>0.88 b</td>
<td>6.06 b</td>
<td>9.05 b</td>
<td>30.17 b</td>
</tr>
<tr>
<td>Treatments</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>T1</td>
<td>0.53 a</td>
<td>4.09 a</td>
<td>34.30 a</td>
<td>18.27 a</td>
</tr>
<tr>
<td>T2</td>
<td>0.73 b</td>
<td>7.15 b</td>
<td>7.16 b</td>
<td>25.00 b</td>
</tr>
<tr>
<td>T3</td>
<td>1.49 c</td>
<td>6.23 c</td>
<td>0.38 c</td>
<td>51.08 c</td>
</tr>
</tbody>
</table>

T1 = *A. niger* + medium; T2 = rock powder + medium; T3 = rock powder + *A. niger* + medium. Averages followed by different non-capitalized letters in the lines differ significantly at 5%.

Figure 1. Potassium concentration (K) and solubilization rate of potassium (SR) in the culture medium with two strains of *A. niger* (CCT911 and CCT4355), during 35 days, at 30ºC, 160 rpm. (● = CCT911 + medium; ○ = CCT4355 + medium; ▲ = rock powder + CCT911 + medium; ▼ = rock powder + CCT4355 + medium).

Approximately 70% of the K released was solubilised after 35 days of incubation of CCT4355 in liquid medium. When the strain was CCT911, the solubilization rate was higher than CCT4355 up to 21 days. After this period, CCT 4355 was more efficient in K solubilization than CCT 911. Lopes-Assad et al. (2006) have verified as much as 53.7% of SR in assays with the strain CCT4355 using alkaline ultramafic rock after 21 days of incubation. There was a significant increase in the titratable acidity and decrease in the medium pH in the treatments where the *A. niger* was inoculated, indicating that this fungus produces acids, as had been found by Nahas et al. (1990) and Vassileva et al. (1998). Lian et al. (2007) have studied a strain of the thermophilic fungus *Aspergillus fumigatus* cultured with K-bearing minerals to determine if microbe–mineral interactions enhance the release of mineral K. They observed that the K solubilization rate showed a
positive dependence upon pH when fungi and minerals were mixed directly, and exhibited no correlations with solution acidity if cell–rock contact was restrained. Concerning the use of rock powder in agricultural systems, of particular importance is soil pH; P (and K) will be released more quickly in moderately acid soils than in neutral or alkaline soils. Supanjani et al. (2006) studied the direct applications of P rock and K rock in conjunction with P-solubilizing bacteria and K-solubilizing bacteria for cultivation of hot pepper Capsicum annuum and verified a variety of benefits (improved nutrient uptake, higher biomass harvest and fruit yield) and a promising, sustainable alternative to the use of classical fertilizers. Aspergillus niger offers a great potential for biotechnological applications, especially in the production of organic acids and solubilization of rocks for biofertilizers.

Conclusion
The data obtained showed that Aspergillus niger has high potential of K solubilization and could be an alternative for small farmers to produce biofertilizers using rock powder.

References


