**IMPROVEMENT OF PRODUCTIVE CAPACITY OF SOLONETZ BY APPLICATION OF MICROORGANISMS**

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**Abstract**


Solonetz are dark alkaline soils which are mainly bare or covered by grasses. These sodic soils are characterized by pore water regime, high sodium saturation that is harmful to plants, unfavorable chemical and physical condition which makes them unsuitable for agricultural uses. Productive capacity of solonetz soils are usually improved by different meliorative measures such as incorporation of gypsum, deep ploughing or planting of a sodium-resistant crop. Application of microbial inocula could be a possible solution for soil quality improvement. The microbiological properties of solonetz from Kumane region were investigated as well as the effectiveness of four isolates of *Azotobacter* sp. and two isolates of *Bacillus* sp. Bacteria were isolated from virgin solonetz soil which was not covered by plants and was not recultivated. Pure cultures of each isolate were obtained containing $10^{10}$ CFU/ml of *Azotobacter* sp. and $10^{12}$ CFU/ml of *Bacillus* sp., respectively. Soil samples were inoculated. The virgin solonetz soil without inoculation was the control treatment. Soil sampling was performed 30 days after inoculation. Microbiological analyses included the determination of number and enzymatic activity of microorganisms. All six investigated bacteria strains stimulated the microbiological activity of solonetz. The total number of bacteria, number of fungi, actinomycetes, free-living nitrogen fixing bacteria and aminoheterotrophs were increased in relation to the control. Dehydrogenase activity was also higher after bacterial inoculation. This research highlighted the positive effect of *Azotobacter* sp. and *Bacillus* sp. application. Further research needs to be carried out in order to increase the benefits of microbial inoculation in semi controlled and field conditions.

**Key words:** solonetz, bacteria, inoculation, microbiological activity

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**Introduction**

Solonetz are soils with a dense, strongly structured, clay illuviation horizon that has a high proportion of adsorbed sodium and/or magnesium ions. ABtnC- and AEBtnC-profiles with a black or brown surface soil over a nitrific horizon that starts at less than 100 cm from the soil surface. A calcic or gypsic horizon may be present below the nitrific horizon. Many solonetz have a field-pH around 8.5 indicative of the presence of free sodium carbonate. High levels of exchangeable sodium ions affect arable cropping, either directly (Na-toxicity) or indirectly, e.g. because of structure deterioration when soil material with a high proportion of adsorbed sodium and/or magnesium ions is wetted. Many solonetz in temperate regions have a humus-rich surface soil and can be used for arable farming or grazing. Solonetz is a soil type which occupies significant area in Vojvodina province (80.33 ha, approx. 3.75%) (Sekulić et al., 2005). These are alkaline soils mostly used as natural pasture.

The suitability of ‘virgin’ solonetz for agricultural uses is almost entirely dictated by the depth and properties of the surface soil (FAO report, 2006). A ‘deep’ (say > 25 cm) humus-rich surface soil is needed for successful arable crop production. Unfortunately, most solonetz have only a much shallower surface horizon, or have lost the surface horizon altogether. By applying different meliorative measures the productive capacity of solonetz can be improved and used as cultivated soil (Belić, 1999; Belić et al., 2003). Solonetz amelioration has two basic elements such as improvement of the porosity of the (sub) surface soil, and lowering of the ‘Exchangeable Sodium Percentage’ ESP. Most reclamation attempts start with incorporation of gypsum or, exception-
ally, calcium chloride in the soil (Kruzhilin and Kazakova, 2011). Traditional reclamation strategies start with the planting of a sodium-resistant crop, e.g. *Rhodes* grass, to gradually improve the permeability of the soil.

Application of microbial inocula could be a possible solution for soil quality improvement. Since halomorphic soils are present in Vojvodina, aside traditional amelioration, the application of microorganisms could be a potentially good measure for enhancing soil productive capacity. In this work, the effectiveness of several indigenous bacteria isolates (*Azotobacter* and *Bacillus*) was investigated.

**Materials and Methods**

Soil samples were taken from Kumane region (experimental field Kumane-Faculty of Agriculture) with sterile soil sampler at the depth of 10, 20 and 30 cm. The samples were put in sterile polyethylene bags and transported to the Microbiology Laboratory. The number of microorganisms was determined by agar plate method, inoculating selective media with soil suspension of appropriate dilution (Trolldenier, 1996). The number of *Azotobacter* (AZB), number of aminoheterotrophs (AMH), total number of bacteria (TNB), number of actinomycetes (ACT) and number of fungi (FNG) were analyzed. The dehydrogenase activity (DHA) was determined spectrophotometrically by method of Thalmann (1968) modified by Lehnard (1956). Bacteria (*Azotobacter* and *Bacillus*) were isolated from virgin solonetz soil which was not covered by plants and was not recultivated. Pure cultures of each isolates were obtained on Fjodorov medium and MPB medium containing $10^{10}$ CFU of *Azotobacter* sp. and $10^{12}$ CFU of *Bacillus* sp., respectively. Soil samples were inoculated. The virgin solonetz soil without inoculation was the control treatment. Soil sampling was performed 30 days after inoculation. Microbiological analyses included the determination of number and enzymatic activity of microorganisms.

**Results**

Bulk and rhizosphere soil, as well surface and subsurface soil profiles, differ in their chemical, physical as well as biological properties (pH, salinity, redox potential, porosity, water retention, abundance and diversity of microbial species, etc) (Table 1).

Productive capacity of solonetz soils are usually improved by different meliorative measures such are incorporation of gypsum, deep plowing or planting of a sodium-resistant crop. Application of microbial inocula is a possible solution for soil quality improvement.

Microbiological activity of virgin solonetz is presented in Table 2. The top soil is characterized by high total number of bacteria. Moreover, aminoheterotrophs are dominant in soil. Fungi and actinomycetes as major organic matter decomposers are also present in substantial number. Azotobacter is a free living nitrogen fixing bacteria and are present in soil. Since bacteria from N cycle were dominant in soil, several *Azoto*

### Table 1
**Physical and chemical properties of solonetz**

<table>
<thead>
<tr>
<th>Horizon</th>
<th>Depth, cm</th>
<th>pH</th>
<th>CaCO$_3$, % H$_2$O</th>
<th>CaCO$_3$, % IMKCl</th>
<th>Humus, %</th>
<th>P$_2$O$_5$, mg/100g</th>
<th>K$_2$O,</th>
<th>Ecc 25°C, ms/cm</th>
<th>Salts, %</th>
<th>pH, paste</th>
</tr>
</thead>
<tbody>
<tr>
<td>Aoh/E,na</td>
<td>0-13</td>
<td>5.9</td>
<td>4.6</td>
<td>0</td>
<td>6.05</td>
<td>7.3</td>
<td>87.6</td>
<td>0.71</td>
<td>0.03</td>
<td>5.57</td>
</tr>
<tr>
<td>Bt, na</td>
<td>13-36</td>
<td>7.86</td>
<td>6.38</td>
<td>0.27</td>
<td>1.69</td>
<td>1.5</td>
<td>17.6</td>
<td>2.69</td>
<td>0.14</td>
<td>7.07</td>
</tr>
<tr>
<td></td>
<td>36-58</td>
<td>8.2</td>
<td>6.87</td>
<td>0.20</td>
<td>1.48</td>
<td>5.4</td>
<td>23.4</td>
<td>3.6</td>
<td>0.19</td>
<td>7.69</td>
</tr>
<tr>
<td></td>
<td>58-85</td>
<td>8.93</td>
<td>7.3</td>
<td>0.42</td>
<td>0.83</td>
<td>6.4</td>
<td>28</td>
<td>2.15</td>
<td>0.17</td>
<td>8.21</td>
</tr>
<tr>
<td></td>
<td>85-114</td>
<td>9.31</td>
<td>7.4</td>
<td>0.29</td>
<td>0.43</td>
<td>3.1</td>
<td>19</td>
<td>0.93</td>
<td>0.13</td>
<td>8.62</td>
</tr>
<tr>
<td>Bt,na C, na</td>
<td>114-154</td>
<td>9.38</td>
<td>7.49</td>
<td>20.01</td>
<td>0.99</td>
<td>1.3</td>
<td>15.4</td>
<td>1.05</td>
<td>0.14</td>
<td>8.67</td>
</tr>
<tr>
<td>C, na</td>
<td>154-200</td>
<td>9.4</td>
<td>7.52</td>
<td>16.26</td>
<td>0.16</td>
<td>2</td>
<td>12.2</td>
<td>0.11</td>
<td>8.62</td>
<td></td>
</tr>
</tbody>
</table>

### Table 2
**Microbiological activity of virgin solonetz**

<table>
<thead>
<tr>
<th></th>
<th>TNB ($10^5$g$^{-1}$)</th>
<th>FNG ($10^3$g$^{-1}$)</th>
<th>ACT ($10^5$g$^{-1}$)</th>
<th>AMH ($10^3$g$^{-1}$)</th>
<th>AZB ($10^8$g$^{-1}$)</th>
<th>DHA (µg TPF/10g)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0-10 cm</td>
<td>74.2</td>
<td>31.24</td>
<td>14.19</td>
<td>69.03</td>
<td>98.94</td>
<td>661.9</td>
</tr>
<tr>
<td>10-20 cm</td>
<td>89.81</td>
<td>33.69</td>
<td>9.73</td>
<td>29.94</td>
<td>97.35</td>
<td>569.5</td>
</tr>
<tr>
<td>20-30 cm</td>
<td>68.95</td>
<td>36.31</td>
<td>15.15</td>
<td>22.04</td>
<td>79.76</td>
<td>502.8</td>
</tr>
<tr>
<td>average</td>
<td>77.65</td>
<td>33.75</td>
<td>13.02</td>
<td>40.34</td>
<td>92.02</td>
<td>578.1</td>
</tr>
</tbody>
</table>
bacter and Bacillus isolates were isolated, cultured in pure culture and reintroduced in soil in high number (10^10 CFU/ml of Azotobacter sp. and 10^12 CFU/ml of Bacillus sp., respectively).

Introduction of indigenous bacteria isolates from virgin solonetz led to the increase of all investigated number of microorganisms as well as dehydrogenase activity (Table 3).

**Discussion**

Increased salinity/sodicity (EC/Na) in the rhizosphere affects root exudation and biochemical transformations by microorganisms (Ondrasek and Rengel, 2012). High clay and increased Na content on soil at Bt horizon leads to colloid peptization and alkaline reaction of soil (Belić, 1999). These sodic soils are thus unfavorable or less favorable for microbial growth and activity. Similar observation were reported by Li et al. (2006), who noticed significant negative correlations between soil EC and microbial biomass C, suggesting that salinity had an adverse effect on microbial biomass and activity.

Brankov et al. (2006) investigated the microbiological activity of several soil types in Banat. According to the results of that research, soils with higher humus and nitrogen content as chernozem, solonetz and humogley were characterized by increased number of bacteria, azotobacter and dehydrogenase activity. Research showed that the most important limiting factor for plant production in solonetz is physiologically active depth of 22 cm (Škorić et al., 1985). Jarak et al. (2012) pointed out that number of microorganisms varied and depended on soil type and profile depth. They investigated the microbial abundance in five soils such were sandy soil, solonetz, black soil, chernozem and fluvisol from different sites. The number and diversity of nitrogen fixing cyanobacteria were considerably higher in sodic and hydromorphic soils. The highest number of bacteria was counted in surface layer (0-5 cm). These soils were not used for agriculture production so the majority of easily decomposing soil organic matter was placed in the top soil resulting in increased bacteria number. Katai et al. (2011) found lesser quantity of microscopic fungi in salt effected soil while larger number of aerobic cellulose decomposing and nitrifying bacteria was found in the cultivated soils. They also stated that the enzymes activities were generally higher in the natural soils than in the cultivated ones. The authors measured higher number of microorganisms and more intensive microbial activity in the soil from higher exposure indicating the importance of the micro relief in the occurrence and activity of living organisms of soil.

Generally, the abundance of fungi in comparison to bacteria is lower even after bacterial inoculation of soil. This is in accordance with the results of Sardinha et al. (2003). The authors found that the fungal biomarker ergosterol and the basal respiration rate continuously decreased from the non-saline sites to the saline-sodic sites. Fungi have been reported to be more sensitive to salinisation than bacteria in acidic soils, but also in alkaline soils (Badran, 1994; Pankhurst et al., 2001).

**Conclusions**

The total number of bacteria, number of fungi, actinomycetes, free-living nitrogen fixing bacteria and aminoheterotrophs were increased in relation to the control. Dehydrogenase activity was also higher after bacterial inoculation. This research highlighted the positive effect of Azotobacter sp. and Bacillus sp. application. Further research needs to be carried out in order to increase the benefits of microbial inoculation in semi controlled and field conditions.

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