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EFFECTS OF AZOTOBACTER CHROOCOCCUM INOCULATION WITH DIFFERENT ORGANIC WASTES ON YIELD OF SPRING WHEAT (TRITICUM AESTIVUM L.)

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This study is conducted under controlled conditions in the greenhouses, for the purpose of determination of effects of indigenous Azotobacter chroococcum RK 49 inoculation on the yield of spring wheat (Triticum aestivum L.). For this purpose different types of organic wastes are added to the soil at the rate of 5%, and thus the organic matter content of the soil is increased. In some pots only the effects of organic wastes on the yield of wheat plant are determined, in some pots effects of addition of some organic wastes together with indigenous Azotobacter chroococcum RK 49 inoculation and only the effects of A.chroococcum inoculation on the yield of the wheat plant. At the end of study, it is found that both the addition of organic wastes and A.chroococcum inoculation has increased the yield of the wheat plant. At the end of the experiment, 237.8 kg/da grain productivity is gathered from the controlled application, 277kg/da grain yield is gathered from A.chroococcum inoculation application, which is increased 16.5% more compared to controlled application. It is also determined that, an increase in the yield has also occurred in only in the addition of organic waste applications, and highest productivity increase has occurred in the addition of 5% wheat straw. In addition, the highest yield is determined on the application of A.chroococcum inoculation made together with organic wastes addition, and in the all experiment application, the application that made highest increase in the yield of grains is determines as wheat straw + A.chroococcum inoculation application, which has increased the yield by 25.4%.

Key words: Azotobacter chroococcum, wheat, organic waste, inoculation

1.INTRODUCTION
Depletion of non-renewable sources of energy, escalating cost of fertilizers and environment quality of aspects necessitated the review of various approaches focusing on the use of available renewable sources of plant nutrition for sustainable agricultural production. As a result renewed research efforts are being made systematically to evaluate the feasibility and efficiency of bio-degradable wastes of produced at farms, agroindustries and cities, in re-furnishing soil productivity and improving the efficiency of chemical fertilizers (Chauhan and Thakur, 2012).

One way to increase crop yield is using the beneficial microorganisms. Plant growth promoting rhizobacteria (PGPR) are some of bacteria that can grow in the root environment and be effective on plant growth (Vessy, 2003; Yolcu et al. 2012). Mechanisms that can promote plant growth include production of phytohormones, biological nitrogen fixation and increased solubility of insoluble elements in soil (Rovera et al., 2008; Rosas et al., 2005). Studies showed that the inclusion of wheat plant with PGPR increased the growth characteristics of wheat; bacteria studied were included Azospirillum (Bashand and Levanoyny, 1990), Azotobacter (Rai and Gaure 1988), Basillus (Freitas, 2000), Pseudomonas (Zaidi and Khan, 2005), Clostridium (Gasoni et al., 2001), and Herbaspirillum (Baldani et al., 2000).

Azotobacter is a free living N2 fixing bacterium. It can successfully grow in the rhizospheric zone of wheat, maize, rice, sorghum, sugarcane, cotton, potato, brinjal, cabbage and many others and fix 10-20 kg N ha-1 cropping season-1 (Jadhav et al., 1987). Besides N2 fixation, Azotobacter synthesizes and secretes considerable amounts of biologically active substances like B vitamins, nicotinic acid, pantothenic acid, biokin, heteroauxins, gibrellins etc which enhance root growth of plants (Rao, 1986). Another important characteristics of Azotobacter association with crop improvement in excretion of ammonia in the rhizosphere in the presence of root exudates, which helps in modification of nutrient uptake by the plants (Narula and Gupta, 1986; Kader et al., 2002). All these factors combined together produce positive effects on crop yield. Under the above circumstances, this study was undertaken to test the effects of indigenous Azotobacter chroococcum RK 49 inoculant with different organic wastes on yield of spring wheat (Triticum aestivum L.) under controlled condition.
2. MATERIAL AND METHODS

2.1. Organic waste

Wheat straw, rice straw and soybean waste were collected during the grain harvest season in Samsun, Turkey. Tobacco production waste was taken from the tobacco production industry. All organic wastes were dried and sieved into less than 0.50 mm. The properties of the organic wastes were expressed on a dry weight basis and were analyzed by standard procedures as given in Ryan et al. (2001). Among the organic wastes used in this study, wheat straw had the highest organic matter while that of tobacco waste was the lowest. Regarding N content, tobacco waste had the highest N content (1.93%) and the lowest N content belong to wheat straw (0.48%). C:N ratio of the organic wastes ranged from 22 to 100 and the highest level C:N ratio observed in wheat straw while that of lowest is tobacco waste. The order of organic waste associated with C:N ratio was Tobacco waste > Rice waste > Soybean waste > Wheat straw. In addition these OW contained major nutrients such as P, K and Ca which are agronomically important.

<table>
<thead>
<tr>
<th>Organic material</th>
<th>C/N</th>
<th>N, %</th>
<th>P, %</th>
<th>K, %</th>
<th>Ca, %</th>
</tr>
</thead>
<tbody>
<tr>
<td>Wheat straw</td>
<td>100</td>
<td>0.48</td>
<td>0.10</td>
<td>2.81</td>
<td>0.41</td>
</tr>
<tr>
<td>Rice waste</td>
<td>87</td>
<td>0.52</td>
<td>0.08</td>
<td>2.17</td>
<td>0.21</td>
</tr>
<tr>
<td>Tobacco waste</td>
<td>22</td>
<td>1.93</td>
<td>0.18</td>
<td>3.66</td>
<td>2.86</td>
</tr>
<tr>
<td>Soybean waste</td>
<td>88</td>
<td>0.58</td>
<td>0.06</td>
<td>3.77</td>
<td>0.55</td>
</tr>
</tbody>
</table>

2.2. Azotobacter chroococcum

Indigenous Azotobacter chroococcum RK49 strain was provided by the Soil Microbiology laboratory in Ondokuz Mayis University, Samsun, Turkey. This organism was repeatedly tested for their for nitrogen fixation capacity and the effects on yield of plant (Kızılkaya, 2008, 2009) and stored in ultralow temperature freezer. Before inoculation, the indigenous Azotobacter chroococcum RK49 strain were cultivated by Nitrogen-free Ashby medium (5g glucose, 5g mannitol, 0.1 g CaCl2, 2 H2O, 0.1 gr MgSO4, 7H2O, 5 mg Na2MoO4, 2 H2O, 0.9 g K2HPO4, 0.1 g KH2PO4, 0.01 g FeSO4 .7H2O, 5 g CaCO3, 15 g agar in 1 L distilled water, pH 7.3). Pure culture of indigenous Azotobacter chroococcum RK49 strain used for inoculation were grown in N-free Ashby agar at 30 °C. A single colony from each strain was transferred to a 50 mL flask, containing nitrogen and agar free Ashby medium, and grown aerobically in flask 72 hour, on a rotating shaker (125 rpm) at 30 °C. A chroococcum strain grown liquid Ashby medium was then diluted with sterile distilled water, containing 0.025% Tween 20 to a final concentration of 10⁹ CFU mL⁻¹. For seed treatments, wheat seeds were placed in bacterial suspensions of 10⁹ CFU mL⁻¹ for 30 min before sowing under sterilized conditions and then transferred unsterilized soil.

2.3. Experimental soil

Surface soil (0-20 cm) was taken from the agricultural field in Bafr, Samsun. Some physico-chemical properties of soil were determined according to Rowell (1996). The soil used in this experiment contained 34.6 % clay, 31.93% silt, and 33.47% sand. Soil texture can accordingly be classified as a clay loam. Chemical properties were measured as follows: pH in water: 7.52, oxidizable organic matter content: 2.44%, CaCO3, 6.27% and Electrical Conductivity (EC) 0.84 dSm⁻¹. The experimental field had been under arable agriculture for 35 years. The site is located in the Black Sea Region, Northern Turkey (Latitude, 41°21'N; longitude, 36°15'W). The climate is semi-humid, the annual mean temperature is 13.6 °C and annual mean precipitation is 764.3 mm.

2.4. Greenhouse experiment

A pot experiment was carried out in the greenhouse with the spring wheat (Triticum aestivum) PAN-DAS in order to investigate the effects of inoculation with indigenous Azotobacter chroococcum RK49 strain with different organic wastes. Soil without organic waste addition and A. chroococcum inoculation was used as a control. A randomized complete plot design with three replicates per treatment and soil was used. The experiment was performed with the following 10 treatments:

1) control
2) + wheat straw (%5)
3) + rice straw (%5)
4) + tobacco waste (%5)
5) + soybean waste (%5)
6) + wheat straw (%5) + indigenous Azotobacter chroococcum RK49 inoculation
6) + rice straw (%5) + indigenous *Azotobacter chroococcum* RK49 inoculation

7) + tobacco waste (%5) + indigenous *Azotobacter chroococcum* RK49 inoculation

8) + soybean waste (%5) + indigenous *Azotobacter chroococcum* RK49 inoculation

9) + indigenous *Azotobacter chroococcum* RK49 inoculation

10) This soil was filled in 5L pots. Thirty seeds were sown in each pot and thinned to fifteen plants per pot after the full emergence of the first leaf. The pots were regularly irrigated to maintain a proper moisture level. Plants in pots were harvested 83 days after sowing. At the end of the experiments, plant were collected from the pots. Studied plant parameters were grain yield at the end of the both pot experiments.

3. RESULTS AND DISCUSSION

The changes on the grain yield of spring wheat in the addition of 5% dosage of different organic wastes, in addition of indigenous *A.chroococcum* RK 49 inoculation together with 5% dosage addition of different organic wastes and the only the inoculation of indigenous *A.chroococcum* RK 49 inoculation, are shown in Figure 1. The increases that occurred in these applications compared to the controlled application are given in Figure 2. It is determined that there are significant increases in the productivity of wheat plant in both by applying indigenous *A.chroococcum* RK 49 inoculation and organic wastes separately and applying indigenous *A.chroococcum* RK 49 inoculation and organic wastes together. While the yield of controlled application is 237.8 kg/da, the yield has increased by 16.5% to 277 kg/da.

![Figure 1. Effects of different organic wastes and the effects of indigenous A.chroococcum RK 49 inoculation made together with those organic wastes on spring wheat yield](image)

Similarly, the yield of the wheat has increased by 19.4% to 283.9 kg/da by 5% of wheat straw application to the soil, it has increased by 4.8% to 249.1 kg/da by tobacco waste application, it has increase by 0.5% to 239.1 kg/da by rice straw application, it has increased by 15.9% to 275.5 kg/da by soybean waste application. On the other hand, the increase on the yield of wheat which the *A.chroococcum* RK 49 inoculation made together with application of organic wastes made on has occurred much more effectively. The highest increase that has occurred in the yield of grain of the wheat plant is gathered by *A.chroococcum* RK 49 inoculation made together with mixing the wheat straw to the soil at the rate of 5%.

It is determined that increase on the productivity of wheat by the application of *A.chroococcum* RK 49 inoculation made together with application of wheat straw soybean waste is much more that the increase
occurred by the application of other wastes. Even, it is determined that the increase on the yield of wheat gathered by the application of only *A. chroococcum* RK 49 inoculation is more that the application of tobacco waste + *A. chroococcum* RK 49 inoculation and rice straw + *A. chroococcum* RK 49 inoculation applications. In fact, it is expected that the addition of *A. chroococcum* RK 49 inoculation to tobacco waste and soybean waste application should make more increase on the yield more than the application of those wastes alone, but it did not occur. Beyond the doubt, this situation can be related to the chemical composition of the wastes. Some organic compositions that are produced during decomposition of tobacco waste in the soil may limit the *A. chroococcum* population and the fixation of nitrogen by Azotobacter. Similarly, it is thought that because of its wider C/N rate than the other wastes that have and its resistant feature against to decomposition arising from the existence of high silicum content in its composition, it does not decompose during the experiment and can’t contribute to the productivity as expected. The studies have shown that in order to *A. chroococcum*, which are heterotrophic, can continue their lives, can be active and being able to contribute to productivity of the plant, the organic substance in the soil must be at adequate level. It is determined that, if the organic substance level is not in adequate level, the level of the organic substance in the soil must be increased with plant and animal based organic compounds, and the C/N rate of that organic compound must be greater than 33/1. In the cases, where the C/N rate of organic material is smaller than 33/1 (Alexander, 1977), the Azotobacteria that inoculated to the environment uses the nitrogen in organic material instead of fixation the nitrogen in the atmosphere, thus their contribution to plant yield become very limited.

![Graph showing percentage increase in yield](image)

**Figure 2. Percentage in increase on spring wheat yield made by different organic wastes and the indigenous *A. chroococcum* RK 49 inoculation made together with those organic wastes**

As a result, one of the most fundamental factors for increasing the productivity of wheat cultivation is the *Azotobacter* cultivation of the soil which supports the plant’s feeding. In addition to, in *A. chroococcum* RK 49 inoculation, adding the plant based wastes to the soil both promotes to development of the plant’ root by increasing the organic substance level, and increases the efficiency of *Azotobacter*. Besides those that the same results found in this study also, it is determined that the efficiency of *Azotobacter* changes according to organic material. It is determined that, the *A. chroococcum* RK 49 inoculation made together with the wheat wastes that are left over at the end of the wheat cultivation increases the plant product yield.

**References:**


**BIOLOGICAL ATTRIBUTES IN SOIL QUALITY FOR SUSTAINABLE LAND MANAGEMENT**

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