GROWTH YIELD AND NUTRIENTS UPTAKE BY WHEAT PLANTS AS AFFECTED BY PHOSPHORUS LEVELS AND MYCORRIZAL INOCULATION UNDER SALINE CONDITIONS
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ABSTRACT
There is an inability in facilitating phosphorus under Egyptian soil conditions. For this purpose, a pot experiment was conducted at the experimental farm Faculty of Agric., El-Mansoura Univ. during the winter season of 2008-2009 to investigate the uptake of applied and residual phosphorus by wheat (Triticum aestivum L.) plants as affected by mycorrhizal inoculation under saline condition. Phosphorus was applied at four rates of 0, 50, 75 and 100 Kg/feddan as single super phosphate (15.5\%P2O5) under three levels of salinity (1, 6 and 9 dSm\(^{-1}\)). Arbuscular mycorrhizal (AM) fungi was used as a mixture including (Glomus mossea, Glomus intraradices and Glomus clarium). The results of this investigation revealed that: with AM inoculation, the mean values of N, P and K\% in shoot and roots and Na\% in roots were significantly increased. Also, with AM inoculation, the mean value of Na\% in shoots decreased. This trend was true during both stages of planting. On contrast, in non-mycorrhizal inoculated plants the illustrated average of N, P and K\% in wheat shoot and roots were significantly decreased as the level of salinity increased. On the other hand, the mean values of Na\% in wheat shoots and roots were significantly increased due to adding salinity levels over the control during both stage of growth. Concerning the effect of phosphorus application, the average of N, P and K\% in wheat shoots and roots were increased significantly over the control. Adding P\(_3\) level was superior for increasing aforementioned traits. Addition of P levels, however, had no significant effect on Na\% in wheat shoots. This trend was observed during both stages of the experiments. With respect to the interactive effect between adding AM, salinity levels and phosphorus application, it could be realized that adding P\(_3\) level with S\(_0\) level combined with I\(_1\) (AM inoculation) gave the highest value of N, P and K in wheat shoots and roots. However, the mean values of Na \% did not significantly affected by treatments of the experiment.

Keywords: Mycorrhizal inoculation, salinity conditions, phosphorus fertilization, wheat plants.

INTRODUCTION
Wheat is one of the most important grain crops. Raising wheat through increasing the productivity per unit area as well as expanding the cultivated area in newly reclaimed lands is a major important national target. Increasing productivity per unit area, particularly in saline conditions, could be achieved by cultivating high yielded cultivars along with importing agronomical practices. As a result, arbuscular mycorrhizal plants are often more competitive and better tolerant to the environmental stresses than non-mycorrhizal plants (Abdel-Fattah et al., 2002; Paradi et al., 2002; Kumar et al., 2010; Asrar and ElHindi 2011).
Arbuscular mycorrhizal fungi (AMF) are a main component of the soil edaphon in most agro ecosystems. These obligate mutualistic symbionts colonize the roots of the majority of crop plants (Smith and Read 1997). AMF can efficiently absorb mineral nutrients (George et al., 1995) by their extended hyphal network, especially from low fertile soils, and deliver them to their host plants in exchange for carbohydrates. Arbuscular mycorrhizal fungi employ different mechanisms to enhance salt tolerance of host plants such as enhancing nutrient acquisition (P, N and Ca) (Giri and Mukerji 2004; Sheng et al., 2009), inhibiting high uptake of Na and Cl and their transport to plant shoots (Daei et al., 2009) and improving water uptake and transport by plant xylem (Ruiz-Lozano and Azcon2000).

Phosphorus (P) is an essential nutrient element production of ATP, DNA, RNA, and other cell constituents of plants. It plays important roles in nearly all phases of the plant life, including photosynthesis, flowering, seed production, maturation, and root growth. Its deficiency can cause severe stunting and significant yield losses (Haven et al., 1999). Soil-P is often a limiting nutrient for plant growth. The P concentration in soil solution is typically very low because soluble forms of P are fixed by soil solid phase, making less than 0.01% of total soil P available to plants (Gallaher, 2007; Mengel and Kirkby, 2001). Phosphorus, therefore, is considered one of the least mobile plant nutrients in soil.

Soil salinity is a worldwide dilemma, restricting plant growth and production, especially in arid, semiarid and tropical regions through reducing nutrients uptake and increasing osmotic stress of plants (Apse et al., 1999; Abdel-Ghani 2009). Those regions are still increasing as a result of salt water irrigation and land degradation.

**MATERIALS AND METHODS**

**Sowing:** A pot experiment was conducted at the experimental farm of Faculty of Agric., El-Mansoura Univ. during the winter season of 2008-2009 to investigate the uptake of applied and residual phosphorus by wheat (*Triticum aestivum* L.) plants as affected by mycorrhiza inoculation under saline condition. 24 treatments were arranged in split-split plots design, which were the simple possible combination between two treatments of mycorrhizal inoculation, three levels of salinity and four levels of P-fertilization. The following treatments were used:

- The mycorrhizal inoculations were adopted in main plots as follows: (I₀) Control (without inoculation) and (I₁) mycorrhizal inoculation (*Glomus mossea*, *G. intraradices* and *G. clarium*)
- The salinity levels were arranged in sub plots as follows: (S₀) soil salinity of 1 dSm⁻¹ as the control treatment, (S₁) salinity level of 6 dSm⁻¹ and (S₂) salinity level of 9 dSm⁻¹.
- P-fertilization levels were adopted as sub-sub plots as follows: (P₀) without P-fertilization, P₁, P₂ and P₃ as 50, 75 and 100% from the recommended P fertilization dose. The recommended dose of P fertilization is 100 Kg/fed
super phosphate (15.5% P$_2$O$_5$), these levels equal (0.27, 0.405 and 0.54 g/pot respectively).

Each treatment was replicated four times. 72 large plastic pots (20 cm. diameter and 50 cm. depth) were used. Each pot was filled with 5 kg air dried soil taken from the surface layer of a private farm located in Talkha city, Dakahlia Governorate. The soil was analyzed for some physical and chemical properties as shown in Table 1.

Wheat seeds were obtained from seeds production unit; Agriculture Research Center, Egypt. Ten seeds c.v. Sakha 93 were sown on November 27, 2008. 18 days after sowing, plants were thinned to five plants per pot. Two salt solutions were prepared at the rate of (6 and 9 dSm$^{-1}$) of commercial salt i.e. (3.84 and 5.76 g/l.), respectively. The chemical analysis of the used commercial salt is illustrated in table 2.

Table 1: Some physical and chemical properties of the experimental soil.

<table>
<thead>
<tr>
<th>Soil characteristics</th>
<th>values</th>
</tr>
</thead>
<tbody>
<tr>
<td>Particle size distribution</td>
<td></td>
</tr>
<tr>
<td>Sand (%)</td>
<td>23.50</td>
</tr>
<tr>
<td>Silt (%)</td>
<td>26.50</td>
</tr>
<tr>
<td>Clay (%)</td>
<td>50.00</td>
</tr>
<tr>
<td>Soil texture</td>
<td>Clay</td>
</tr>
<tr>
<td>Some physical and chemical properties</td>
<td></td>
</tr>
<tr>
<td>Field capacity (%)</td>
<td>35.0</td>
</tr>
<tr>
<td>Saturation percentage</td>
<td>70.0</td>
</tr>
<tr>
<td>Calcium carbonate (%)</td>
<td>4.00</td>
</tr>
<tr>
<td>O.M. (%)</td>
<td>1.10</td>
</tr>
<tr>
<td>pH (1:2.5)</td>
<td>7.80</td>
</tr>
<tr>
<td>EC (dSm$^{-1}$) soil paste</td>
<td>1.00</td>
</tr>
<tr>
<td>Soluble cations (meq. L$^{-1}$)</td>
<td></td>
</tr>
<tr>
<td>Ca$^{2+}$</td>
<td>2.50</td>
</tr>
<tr>
<td>Mg$^{2+}$</td>
<td>0.70</td>
</tr>
<tr>
<td>Na</td>
<td>3.10</td>
</tr>
<tr>
<td>K</td>
<td>1.00</td>
</tr>
<tr>
<td>Soluble anions (meq. L$^{-1}$)</td>
<td></td>
</tr>
<tr>
<td>CO$_3^{2-}$</td>
<td>-</td>
</tr>
<tr>
<td>HCO$_3^{-}$</td>
<td>0.40</td>
</tr>
<tr>
<td>Cl$^{-}$</td>
<td>3.40</td>
</tr>
<tr>
<td>SO$_4^{2-}$</td>
<td>3.80</td>
</tr>
<tr>
<td>Olsen-p (mg/kg soil)</td>
<td>3.87</td>
</tr>
<tr>
<td>Available nutrients (ppm)</td>
<td></td>
</tr>
<tr>
<td>Olsen-p (mg/kg soil)</td>
<td>305</td>
</tr>
<tr>
<td>Total nutrients (ppm)</td>
<td></td>
</tr>
<tr>
<td>N (mg/kg soil)</td>
<td>32</td>
</tr>
</tbody>
</table>

Table 2: Soluble cations and anions of the commercial salt (meq/l):

<table>
<thead>
<tr>
<th>Na$^+$</th>
<th>Mg$^{2+}$</th>
<th>Ca$^{2+}$</th>
<th>SO$_4^{2-}$</th>
<th>Cl$^-$</th>
<th>CO$_3^{2-}$</th>
</tr>
</thead>
<tbody>
<tr>
<td>38</td>
<td>6</td>
<td>6</td>
<td>5</td>
<td>40</td>
<td>5</td>
</tr>
</tbody>
</table>

Plants were irrigated to catch the field capacity. Thereafter, soil moisture content was kept at 70% of soil field capacity by watering to the constant weight.

Mineral fertilization: Mineral NPK fertilizers were added according to the recommendation of the Ministry of Agriculture and Soil Reclamation. Nitrogen was added at 163 Kg/ fed. (0.88 g. / pot) in form of Urea (46%) in two equal doses after 25 and 60 days from sowing. Potassium was added at 50 days.
after sowing in form of potassium sulphate (48%K₂O) at 50 Kg/ fed (0.27 g. / pot). Phosphorus fertilizer was added as super phosphate (15.5% P₂O₅) at levels of 0, 50, 75 and 100 Kg/fed. i.e. (0, 0.27, 0.405 and 0.54 g. /pot respectively) with sowing.

**Bio fertilization:** a mixture of arbuscular mycorrhizal (AM) fungi including *(Glomus mossea, Glomus intraradices and Glomus clarium)* was used. The mycorrhizal inoculum consisted of root, hyphae, spores and growth media from pot culture of onion plants which was previously infected with *Glomus* spp. Grown for four months in pot culture. The inoculum dosage of AM was 2.0 g per pot containing approximately 250 spores. The inoculum was placed 3cm below wheat-germinated grains at sowing time. Two and three plants were taken after 60 days (tilling growth stage) and 124 days (filling stage) from wheat seeds sowing, respectively from each plot and carried immediately to the laboratory. Plant samples were separated; weight and oven dried at 70°C till constant weight was reached. The dried plant samples were thoroughly ground and stored for chemical analysis.

**Soil analysis:** Particle size distribution was determined using the International Pipette Method as described by Piper (1950). pH value was measured in the 1:2.5 soil water suspension, EC value was measured by electrical conductivity meter in soil paste extract as dSm⁻¹ as described by Jackson (1967). Water saturation percentage was determined by the method described by the U.S. Salinity Laboratory Staff (1954). Organic matter was determined according to Walkley and Black method, Black (1965). Calcium carbonate was determined using Collin's calcimeter according to Piper (1950). Soluble carbonate and bicarbonate were determined by the titration with a standard HCl solution, Jackson (1967). Soluble calcium, magnesium and sulfate in soil paste extract by the titration with a standardized versenate solution, Jackson (1967). Soluble sodium and potassium were determined by a flame photometer, Jackson (1967). Soluble chloride was determined by titration with a standard silver nitrate solution, Jackson (1967).

Available N was measured using the conventional method of Kjeldahl as described by Bremner and Mulvany (1982). Available phosphorus was extracted by 0.5 N sodium bicarbonate and determined following the method of Olsen and Sommers, (1982). Available potassium was determined by flame photometer according to Black (1965).

**Plant Analysis:** The oven dried plant samples were ground and wet digested by sulphuric / perchloric acid mixture as described by Peterburgski (1968). Total nitrogen% was determined by using microkjeldahl method as described by Pregle (1945). Total phosphorus% was determined calorimetrically using the scorbic acid method as described by Murphy and Riley (1962). Potassium and sodium were determined in the digested plant materials using a flame photometer as described by Black (1965).

**Statistical analysis:** All data were statistically analyzed according to the technique of analysis of variance (ANOVA) and the least significant difference (LSD) method was used to compare the differences between the means of treatment according to the methods described by Gomez and Gomez, (1984). All statistical analyses were performed using analysis of variance technique.
RESULTS AND DISCUSSION

Nitrogen, phosphorus and potassium concentration in wheat shoots:

Data illustrated in Figs. 1, 2 and 3 showed the effect of adding AM, salinity levels and phosphorus application on N, P and K concentrations in wheat shoots during both stage of planting. With regard to the effect of adding AM, the mean values of N, P and K% in shoot were significantly increased due to adding AM. On the contrary of this trend, the illustrated average values of N, P and K% in wheat shoots were significantly decreased as the level of salinity increased during both stages of plant growth.

Regarding the effect of phosphorus application, data at the same Figures reflected that; the average values of N, P and K% in wheat shoots were increased significantly over the control treatment by 1.44, 3.24 and 4.68% for N, 3.25, 7.47 and 9.42% for P and 0.93, 1.56 and 2.50% for K during the first stage. The same trend was true in the 2nd stage.

The difference comparison between the average values of N, P and K% in wheat shoots as affected by adding AM, salinity levels and phosphorus application were significantly increased. The highest values were 3.87, 0.424 & 3.98 for N, P and K in the 1st stage and 1.10, 0.155 & 1.35 for N, P and K in the 2nd stage and recorded with I1, S0 level and P3 level.

Figure 1: Interaction effect of adding AM, salinity levels and phosphorus fertilization on nitrogen (N) concentration in wheat shoots after 60 and 124 days from sowing.
Figure 2: Interaction effect of adding AM, salinity levels and phosphorus fertilization on phosphorus (P) concentration in wheat shoots after 60 and 124 days from sowing.

Figure 3: Interaction effect of adding AM, salinity levels and phosphorus fertilization on potassium (K) concentration in wheat shoots after 60 and 124 days from sowing.

Nitrogen, phosphorus and potassium concentration in wheat roots:

Effect of adding AM, salinity levels and phosphorus application and their interactions on N, P and K concentration in wheat roots are presented in Figs. 4, 5 and 6 during 1\textsuperscript{st} stage (after 60 days) and 2\textsuperscript{nd} stage (after 124 days) from sowing.

The effect of adding AM on N, P and K % in wheat roots were significantly increased with inoculating the soil with AM. The highest mean values of N, P and K % in wheat roots were 1.02, 0.175 and 1.82% in the 1\textsuperscript{st} stage and 1.08, 0.178 & 1.83 in the 2\textsuperscript{nd} stage. Optioned results are confirmed with those reported by Abo-Ghalia and Khalafallah (2008), Sharif et al., (2010) and Singh and Singh (2007). The average values of N, P and K % in wheat roots present in the same figures were significantly decreased due to salinity levels was increased from $S_0$ level up to $S_3$ level during both stage of growth. Optioned results are confirmed with those reported by El-Etreiby (2000), Kaya \textit{et al.}, (2001) and El-Arqan \textit{et al.}, (2002).
Adding phosphorus fertilization significantly gave the higher magnitudes of N, P and K % in wheat roots. Adding P3 level was superior for increasing aforementioned traits. For example the rate of increases over the untreated plants for N% were accounted to be 3.45, 5.75 and 8.05% in the 1st stage and 3.32, 6.45 and 9.68% in the 2nd stage for the treatment of P1, P2 and P3 levels, respectively. The results are confirmed with those reported by Chaturvedi (2006), You et al., (2007), Bonde et al., (2008) and Sher et al., (2010). Concerning the effect of the interaction between adding AM, salinity and phosphorus applications levels, it can be noticed that adding P3 level and S0 level combined with I1 (AM inoculation) gave the highest value of N, P and K % in wheat roots. The highest values were recorded as 1.23, 0.193 and 1.98% in the 1st stage and 1.29, 0.212 & 2.00% in the 2nd stage for N, P and K %, respectively.

Soil salinity significantly reduces the absorption of the mineral nutrients, mainly P, N and K in non-mycorrhizal plants. AM-inoculated plants had significantly greater concentrations of (P) Azcon and Atrash (1997) and Roychoudhury et al., (2010), (N) Founoune et al., (2002) and K than those of non-mycorrhizal plants. These findings allow us to deduce that the increased salinity tolerance in mycorrhizal plants is based on P nutrition and other minerals (Azcon and Atrash, 1997). In saline soil, higher absorption of P in AM-inoculated plants may improve growth rate, salt tolerance and suppress the adverse effect of the salinity stress Kumar et al., (2010). In addition, Duke et al., (1986) concluded that in addition to P uptake enhancement, there were some other mechanisms such as induction of the osmotic materials that led to osmotic adjustment and improved salt tolerance in mycorrhizal plants.

Figure 4: Interaction effect of adding AM, salinity levels and phosphorus fertilization on nitrogen (N) concentration in wheat roots after 60 and 124 days from sowing.
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**Figure 5:** Interaction effect of adding AM, salinity levels and phosphorus fertilization on phosphorus (P) concentration in wheat roots after 60 and 124 days from sowing.

**Figure 6:** Interaction effect of adding AM, salinity levels and phosphorus fertilization on potassium (K) concentration in wheat roots after 60 and 124 days from sowing.

**Sodium concentration in wheat shoots and roots:**

Sodium concentration in wheat shoots and roots as influenced by adding AM, salinity levels and phosphorus applications as well as interactions were presented in Figs. 7 and 8 during 1st and 2nd stages of growth.

The mean values of Na % in wheat shoots and roots were significantly affected by inoculating AM. In this respect, as inoculation AM the mean value of Na% in shoots decreased on contrary of the mean value of Na% in roots was increased. This trend was true during both stage of planting.

On the other hand, the mean values of Na % in wheat shoots and roots were significantly increased due to adding salinity levels over the control. In other words, the rate of increase over the control for Na% by wheat shoots and roots were accounted to be 12.82, 25.64% and 30.77, 65.38% in the 1st stage and 13.14, 28.95% and 21.43, 42.86% in the 2nd stage, respectively.
Such data reveal that; using phosphorus application had no significant effect on Na % in wheat shoots and roots during 1st and 2nd stages of planting.

With respect to the interactive effect between inoculating AM, as well as salinity levels and phosphorus application, inoculating AM under any level of salinity with using phosphorus fertilization had no significant effect on Na % in wheat shoots and roots during 1st and 2nd stage of planting.

Obtained results are in agreement with those obtained by Ali et al., (2000), Akbarimoghaddam et al., (2011), Tahir et al., (2011) Also, Abdel-Fattah and Asrar (2012) Mycorrhizal colonization of a plant with AMF can reverse the effect of salinity on K⁺ and Na⁺ Nutrition. Mycorrhizal colonization can enhance K⁺ absorption under saline conditions Alguacil et al., (2003); Giri et al., (2003) and Zuccarini and Okurowska (2008) while preventing Na⁺ translocation to shoot tissues. Na⁺ uptake may also be influenced by the synthesis and storage of polyphosphate Olrovich and Ashford (1993) as well as by other cations, particularly K⁺; Giri et al., (2003). The uptake of K⁺ increased in shoot tissues of mycorrhizal plants even at a high salinity level (9.5dSm⁻¹). This increases the K⁺: Na⁺ ratio in roots and shoots of AM plants Giri and Mukerji (2004).

Figure 7: Interaction effect of inoculating adding AM, adding salinity levels and phosphorus fertilization on sodium (Na) concentration in wheat shoots after 60 and 124 days from sowing.
CONCLUSION

Based on the obtained results of this study it could be concluded that using mycorrhiza inoculation coupled with mineral fertilization increase the availability of plant nutrients and production and reduces the soil pollution resulted from mineral fertilizers.

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النمو والمحصول وامتصاص العناصر بواسطة نباتات القمح متأثرة بمعدلات الفوسفور واللتقيح بالميكروهيزا تحت الظروف المحلية.

خالد حسن الحامدي، أحمد علي موسى ومحمد السيد راضي.

قسم الأراضي – كلية الزراعة – جامعة المنصورة.

نفذت تجربة أُجريت في المزرعة التجريبية، كلية الزراعة، جامعة المنصورة في خلال الموسم العضوي 2008-2009 وذلك لتقييم تأثير إمتصاص العناصر المعتمدة بواسطة نباتات القمح وتأثيرها بالتقيح بالميكروهيزا تحت الظروف المحلية. وتم توزيع التجربة على أربعة عوامل معنوية في تصميم طبقات تحت منشقة وتم تفعيل كل الطرق الممكنة بين معالجات من سلالات الميكروهيزا وعوامل مكونات من الاضافات إلى 4 معالجات من التسديد الفيسيولوجي. وتم توزيع التجربة كالتالي:

- سلالات الميكروهيزا كعمالات رئيسية: (S1) سلاسلة (بندون)، (S2) سلاسلة من النوع (G. clarium و G. intraradices و Glomus mossea).
- نسب من مجموعة أنانية عامة: (P0) مجموعة الفيسيولوجية، (P1) مجموعة الفيسيولوجية ب 50% من المعاملات، (P3) و100% من المعاملات.
- النتائج: تم تقييم نباتات الفيسيولوجية التي تم تصنيفها بالميكروهيزا في معالجات الفيسيولوجية في التجربة وتم حساب النتائج من خلال مقايضة النتائج. ووجد أن مع الزراعة وزيادة متوسطات الفيسيولوجية الفيسيولوجية الميكروهيزا الفيسيولوجية كانت تتأثر بشكل إيجابي على نباتات الفيسيولوجية والميكروهيزا، حيث أن الفيسيولوجية الميكروهيزا الفيسيولوجية كانت تتأثر بشكل إيجابي على نباتات الفيسيولوجية الميكروهيزا.

قام بتحقيق البحث

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