THE INFLUENCE OF PHOSPHORUS AND RHIZOBACTERIA ON SOYBEAN (*GLYCINE MAX. L.*) ROOT GROWTH UNDER SUBOPTIMAL MOISTURE REGIME

ACȚIUNEA FOSFORULUI ŞI RIZOBACTERIILOR ASUPRA CREŞTERII SISTEMULUI RADICULAR LA SOIA (*GLYCINE MAX. L.*) ÎN CONDIȚII SUBOPTIMALE DE UMIDITATE

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Abstract. *The effect of phosphorus and suspension of microorganism's Azotobacter chroococcum and Pseudomonas fluorescens on root growth and phosphorus concentration in soybean plants exposed to a short drought stress was studied in a greenhouse experiment. The suspension of microorganisms was administrated in soil without or with P supply. Control plants were grown at normal moisture conditions 70% WHC (water holding capacity) and other part of plants at flowering stage was subjected to water deficit - 35% WHC. The results shown that drought reduced the morphological root parameters irrespective of level of P. Dry mass of roots, total root length, specific root length and fine roots increased in treatments with microorganisms application, in particular in treatment without P. The content of total P in soybean organs din not change significantly but it was observed an increase of Pi concentration due to P and microorganisms supply.*
Key words: *Glycine max*., drought, rhizobacteria, root growth, phosphorus

Rezumat. *S-a efectuat un studiu în scopul elucidării efectului fosforului şi a suspensiei tulpinilor bacteriene Azotobacter chroococcum şi Pseudomonas fluorescens asupra modificărilor caracterelor morfologice a sistemului radicular şi conținutului de fosfor (P) la plantele de soia în condiții de secetă. Fosforul şi suspensia de microorganisme s-a aplicat în sol înainte de semănat. Plantele martor s-au cultivat la umiditatea optimă (70% din capacitatea pentru apă a solului, CTA). În faza înfloritului un lot de vase a fost supus regimului suboptimal de umiditate (35% CTA) pe o perioadă de două săptămâni. Seceta a condus la reducerea parametrilor morfologici a rădăcinii indiferent de regimul de nutriție cu P. Masa rădăcinilor, lungimea totală a rădăcinilor, lungimea specifică, cota rădăcinilor fine au înregistrat valori mai mari în varianta cu aplicarea microorganismelor. Concentrația fosforului total din organe nu s-a modificat substanțial însă a crescut conținutul fosforului anorganic în urma aplicării fosforului şi microorganismelor.*
Cuvinte cheie: *Glycine max*., rizobacterii, rădăcini, fosfor, secetă

INTRODUCTION

Phosphorus (P) is an essential macronutrient required for plant growth and development, but plants have to cope with limiting soil P availability in many

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terrestrial ecosystems (Schachtman et al., 1998). In the context of increasing international concern for food and environmental quality, the use of plant growth-promoting rhizobacteria (PGPR) for reducing chemical inputs in agriculture is a potentially important issue. PGPR have been applied to various crops to enhance growth, seed emergence and crop yield (Dey et al., 2004; Herman et al., 2008; Minorsky, 2008). Biofertilizers affect the root growth which in turn influences the uptake capacity of nutrients and water (Adesemoye et al., 2009). It is well known that microorganisms (MO) are involved in a large range of biochemical processes and they have direct implications in transformation of phosphorus compounds in soil and they are integrated components of P cycle (Deubel et al., 2000). In addition to improve plant growth, PGPR are involved in the increase of nitrogen uptake, synthesis of phytohormones, solubilization of minerals such as phosphates and production of siderophores that chelate iron and make it available to the plant root (Lalande et al., 1989; Glick, 1995; Bowen and Rovira, 1999). It has also been reported that PGPR is able to solubilize inorganic and/or organic phosphates in soil (Liu et al., 1992). The rhizobacteria are considered the effective pathway of mineral nutrition improvement of plants. It is documented that *Pseudomonas flouresecence* and *Azotobacter chroococum* are mainly components of microbial community of rhizosphere. Among legumes soybean is more sensitive to shortage of moisture and phosphorus deficit. Under such environmental conditions the soybean yield is reduced substantially. In period of energetic crisis and exhaust of non-renewable recourses it is very important to indentify the means for improvement of plant nutrition. Little information is available concerning morphological changes of root in soybean in relation to rhizobacteria and P supply under limited water conditions. One of the research objectives was to determine the impact of phosphorus and rhizobacteria (*Pseudomonus fluroucense* and *Azotobacter chroococcum*) supply on modification of morphological characters of roots and phosphorus concentration in soybean plant parts under suboptimal moisture regime of soil.

MATERIAL AND METHODS

In order to accomplish the research objective it was carried out a factorial pot experiment under semi-controlled environmental conditions with soybean (*Glycine max.L* cultivar Zodiac). The soil was represented by cernoziom carbonated with phosphorus deficit and it was mixed with sand (1:1 v/v). The treatments included variants with microorganisms supply in soil in combination without or with of phosphorus (100 mg P per kg of soil). Three plants were grown in each pot. There were four replications of each treatment. The soybean seeds were inoculated with bacteria strain *Bradyrhizobium japonicum*. Plants were cultivated at two water soil regime 70% WHC (soil water holding capacity) as normal level and 35% WHC as suboptimal level. The suboptimal moisture conditions were imposed at the beginning of flowering for two weeks. Plants were sampled from each treatment two weeks after starting the water stress. Root length was determined by line intersect method according to Tennant D. (1975). Phosphorus was determined calorimetrically by vanadomolybdophosphoric method (Murthy and Riley, 1962). The experimental data were analyzed by ANOVA and the differences were compared by least Significant Difference Test (p=0.05).

RESULTS AND DISCUSSION

Application of P along with PGPR resulted in significant difference for all morphological parameters that is shoot dry weight, root length, root dry weight and P content. Root biomass was significantly increased for all the treatments with P fertilization and inoculation over control (tab. 1, 2).

Table 1

Morphological changes of roots of soybean plants at normal water regime of soil (70% WHC) in relation to phosphorus and microorganisms (MO) supply

Treatments	Root mass, g plant^{-3}	Total root length m plant^{-3}	Mass of fine roots, g plant^{-3}	Fine roots, %
P0	1,71±0,14	136,3±7,4	1,36±0,10	79,5
P20 mg kg^{-1} soil	2,92±0,07	187,3±13,3	2,22±0,17	76,0
P100 mg kg^{-1} soil	4,21±0,12	263,8±11,8	3,71±0,13	88,7
P0 + MO	2,43±0,11	141,7±13,2	2,06±0,20	84,7
P100 mg kg^{-1}soil + MO	4,69±0,07	236,9±13,6	3,77±0,35	80,4

Moisture stress resulted in marked reduction in root growth. Plant fertilization with phosphorus increased the fraction of fine roots that played a major role in acquisition of mineral nutrients and water.

Inoculated plants significantly developed a greater root length and root mass of soybean than plants without inoculation (tab. 1, 2). This fraction was increased by 9,2% due to supplemental nutrition with phosphorus under normal water regime (70% WHC) in comparison to control (P0). The P effect on biomass accumulation of roots was highly significant when the soil had 100 mg P per kg of soil, registering 59,4% increase of biomass over “0” level of P. The amount of biomass accumulated in plants with administration of PGPR was significantly more than the non-inoculated plants.

The root growth was increased significantly in treatment with application of phosphorus in particular under water limiting conditions. Under optimal moisture level the positive effect of rhizobacteria application was established in treatment without P supply and provided an increase by 29,2%, but it was less pronounced in treatment with fertilizer application. The water stress diminished the fraction of fine roots evidently in unfertilized treatment. But the supplemental phosphorus nutrition (100 mg P kg of soil) increased the morphological parameters by two times in plants subjected to drought in comparison with reference treatment.

The largest root system was registered with alone phosphorus application under optimal conditions of humidity. It’s necessary to emphasize that similar effects of MO were revealed in investigations with corn plants and rape (Krey et al., 2011). The estimation of specific root length is a characteristic for rate of fine roots.

Table 2

Morphological changes of roots of soybean plants at suboptimal water regime of soil (35% WHC) in relation to phosphorus and microorganisms supply

Treatments	Root mass, g plant^{-3}	Total root length m plant^{-3}	Specific root length m g^{-1} d.w.	Mass of fine roots, g plant^{-3}	Fine roots, %
P0	1,36±0,15	94,2±2,9	69,3	0,91±0,21	66,9
P20 mg kg^{-1} soil	1,85±0,20	151,8±4,8	82,1	1,18±0,09	63,8
P100 mg kg^{-1} soil	2,73±0,38	246,5±17,5	90,3	2,09±0,26	76,5
P0 + MO	1,51±0,11	136,3±5,3	90,3	1,10±0,10	72,8
P100 mg kg^{-1}+ MO	2,98±0,19	229,5±11,5	76,9	2,36±0,18	79,2

The supply of rhizobacteria strains increased the specific root length under subnormal water conditions from 69,3 to 90 m g^{-1} dry weight in treatment without fertilizer supply (tab. 2).

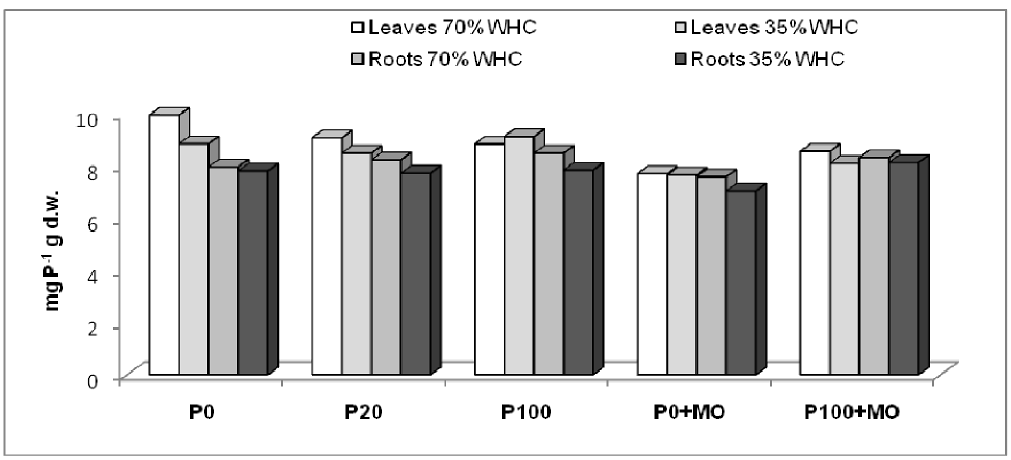

Fig. 1 - Concentration of total phosphorus in leaves and roots as affected by microorganisms (MO) and phosphorus supply in relation to soil water regime

The use of MO in combination with sufficient phosphorus nutrition did not increase this trait. Thus the results demonstrated that the application of PGPR had a stimulatory effect on accumulation of dry matter in roots under unfavorable conditions of soil moisture. The data established that the use of industrial fertilizers reduces the positive impact of bacteria strains. These data are consistent with findings of other researches (Adesemoye et al., 2009). There are a strong relationship between root development and rate of nutrients and water absorption from soil. The total P concentration in leaves and roots indicated poor changes in comparison with values registered in concentrations of inorganic phosphorus (data are not presented). Also there were not significant change in leaves but it was observed an accumulation of P in roots tissues in control plants by application of MO under normal moisture regime of soil (fig. 1). Marschner and Dell (1994) have noted that application of mycorhizal fungi increased the absorption and concentration of nutrients particularly in soil with phosphorus deficit, with positive impact on plant growth and development. In present experiment there was an increase of P concentration in plants in treatment with MO application in deficit P soil. Perhaps this result was caused by biological dilution effect because the leaves mass was lower in this treatment. It is necessary to note that the nutrient concentration in roots increased along with increasing the level of P in hydrated normally plants but there were not changes under water deficit (fig. 1).

Legumes have a higher demand for P nutrition comparative with cereals. There is a need to note that in some investigations was present a large number of bacteria with a solubilization potential of phosphorus compounds, and it can fulfils the nutritive needs with the interrelations established at root level. Therefore, it is considered that adequate P nutrition of plants is favorable for plant fortification and nodules formation (Muntean et al., 2003). That's why the establishment of specific relations between MO and root system are benefic for tolerance of organism to unfavorable conditions of water and phosphorus. The application of MO in soil with P deficiency contributes to large allocation of P to aboveground plant parts of soybean.

Further investigations, including efficiency test under field conditions, are needed to clarify the role of PGPR as biofertilizers that exert beneficial effects on plant growth and seed yield. Such researches will be useful for elaboration an integrated nutrient management in agricultural ecosystem through utilization of bacterial strains and fertilizers.

CONCLUSIONS

1. Our results suggest that plants benefit from the microorganisms symbiosis under dry soil conditions mainly in terms of an improved nutrient uptake.

2. The application of rhizobacteria had a stimulatory effect on root growth without fertilization and increases the soybean tolerance to drought conditions.

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