EVALUATION OF THE POTENTIAL OF DIFFERENT BACTERIAL STRAINS ISOLATED FROM THE RHIZOSPHERE OF PLANTS FOR FURTHER USE IN PLANT GROWTH PROMOTION

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Abstract. *In concordance with the application of environmental-friendly technologies to exploit the natural potential of plants it is necessary to have a deeply knowledge about relations between plant, their natural habitat and microorganisms from rhizosphere. In the last years researchers highlighted the importance of rhizosphere microorganisms in increase of plant fitness by different mechanism: providing a better nutrition of plant, producing plant growth regulators such phytohormone, ensuring protection of the roots of plant from pathogen attack. Our study followed to isolate bacterial strains from rhizosphere of canola (Bassica rape) and nettle (Urtica dioica) and testing their capacities in promoting the growth of wheat plants As a 6 total strains, 4 were isolated from canola and 2 from the nettle that have proven high frequency of appearances on media plates. The pure isolated strains, were tested for support of germination of wheat kernels, plant growth promoting and proven of traits to production of the indoleacetic acid (IAA). For screening of isolates for the production of IAA were used Salkowski regent. The results have revealed significant differences compared to the control in terms of wheat seed germination for strains R-IV, R-III, R-I, U-I. The bacterial strains U II and RI were highlighted in sustaining plant growth through the accumulation of dry matter and production of IAA.*

Keywords: *Plant growth promoting rhizobacteria, IAA, rhizosphere*

INTRODUCTION

Rhizosphere is the area where soil is in vicinity of plant roots. Here, edaphic microbial community is influenced by the metabolites synthesized and excreted by plants. These metabolites are diverse organic compounds which have a chemotactic effect on some beneficial microbes, enhancing plant growing (NANNIPIERI et al. 2007). The quantity and quality of metabolites varies depending of plant development phase, nutritional level, and environmental stress factors, directly influencing the abundance and community of rhizospheric microorganisms (BROECKLING, 2008). In the last 10 years scientists identified a large number of microorganisms with the ability to promote plant growing. These microorganisms were called plant growth promoting rhizobacteria - PGPR (DUTTA & PODILE, 2010). The microbes showed a capability to synthesize phytohormones: indolyl acetic acid (IAA), gibberellin or cytokinins (LENIN & JAYANTHI, 2012), or can assure the necessary nitrogen, solubilize phosphorus, produce siderophores (AHMAD ET AL 2008; RODRÍGUEZ & FRAGA, 1999) or have an indirect action by assuring plant protection against pathogens by synthesizing antibiotics (EL-TARABILY AND SIVASITHAMPARAM, 2006).

MATERIAL AND METHODS.

Rape and nettle plants were collected from field with roots and adherent soil at the end of March, and were taken in laboratory. To isolate and determine the total number of rhizoshere bacteria from rape and nettle, tehnique described as HUSEN et al, 2007 was used.

Rhizosphere bacteria were isolated on non selective media as Nutrient Agar by ten fold dilution series technique. Bacterial isolates that showed a great capacity of growth on culture media were further isolated in pure culture, and microscopically examined. Totally, three colonies from the rape roots surface: R-I, R-III, R-IV and another three from the nettle roots surface : U-I si U-II, U-V have been selected and were multiplied on liquid broth (50ml).

To test the efficiency of bacterial strains on the germination of wheat kernels were used whole wheat seeds from variety Alex put into Petri dishes padded with absorbent paper. It was used 10 seeds /Petri dish, in two repetitions and was distributed 2ml from liquid culture from each isolates on Petri dish. In control samples was distributed only steril lichid broth. The germination test was conducted at thermostat between 21-22 ° C in darkness for 4 days, after that were measured the length of coleoptile, and rootlets for each sample. The results were statistically analyzed.

The germinated wheat seeds were transferred into containers (of 500ml capacity) with perlite for highlighting the strains effect on plant growth. Each container was added with 5 ml of liquid bacterial isolate. The treatment was repeated in two moments for a month period. Between additional dates of bacterial isolates plants were kept wet by adding Hoghland solution (Dobrei Carmen and Sumalan R, 2003). For control sample was using the same quantity of sterile medium. After one month was determined the accumulation of dry matter on each wheat plant of sample. The results were statistically analyzed by ANOVA one way.

The production of indolicacetic compounds by each bacterial isolate was done using minimal medium with L –tryptophan added. After 4 days of incubation in dark condition (23^0C) the IAA produced was determined using Salkowski reagent by measuring the intensity of pink colours spectrophotometrically at 530 nm according to Husen, 2007.

RESULTS AND DISCUSSIONS

There are many papers related to the screening of rhizobacteria from different plant culture (AHMAD, 2008; KUMAR, 2012, UPADHYAY, 2009), but very little about screening of rhizobacteria from wild plant roots (LENIN, 2012).

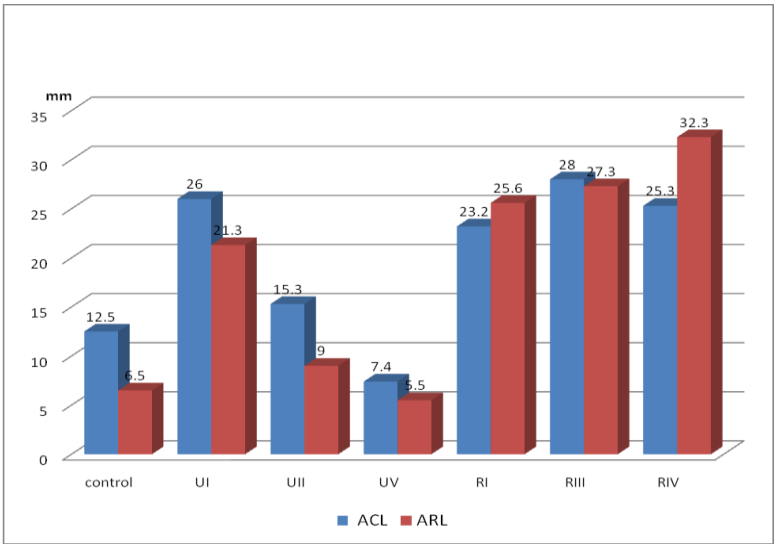

Figure 1. The mean value of coleoptile length (ACL), and rootlet length (ARL) for seedlings after 4 days from bacterial treatments

In present study, beneficial rhizobacteria were isolated and screened for traits related to wheat germination test. As overview from Figure 1, we can observe that U-I, R-I, RIII si R - IV recorded a positive effect comparative with control concerning coleoptile length (ACL) and rootlet length (ARL). However, it seems that R-I and R-IV promotion especially the rootlet growth because ARL values are bigger than ACL values. Bacterial isolate U-V recorded the lowest values while U-II performances are similar to control.

Concerning dry matter accumulation we can observe, from Figure 2, that all bacterial isolate are promoting the growth of plants. If some bacterial isolate promotes the growth of plant roots, as we can observed immediately after germination, later other bacterial strains reveals performance in dry matter accumulation (shoot and root biomass). From this point of view the bacterial isolate U- II recorded the best performance, the difference compared to control is statistically assured. Increases in shoot and root biomass were reported previously with co-inoculation of *B simplex, B. subtilis* and *B. cereus* of tomato plants (HASSEN, 2010).

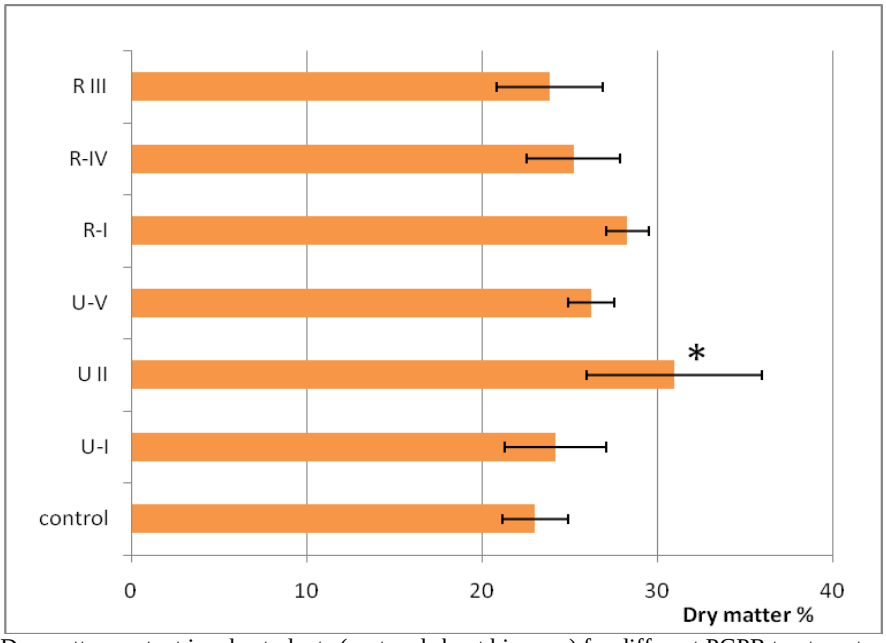

Figure 2. Dry matter content in wheat plants (root and shoot biomass) for different PGPR treatments

IAA is one of the most important phytohormone, and it was reported that IAA production by rhizobacteria can vary among different species and strains, and also can be influenced by culture condition, growth stage and substrate availability (KUMAR, 2012)

Table 1.

The amount of IAA production of bacterial isolates

Bacterial isolates	source	IAA (µg/ml)
U- I	nettle	19.07
U -II	nettle	14.90
U- V	nettle	9.70
R-I	rape	24.80

R-III	rape	16.45
R -IV	rape	23.05

Most of the bacterial strains produced plant growth promoting hormone like IAA (table 1). The range of IAA compounds was between 9.7 – 24.8 µg/ml. Among all six bacterial strains R I produced high quantity of IAA (24.8 µg/ml) followed by R -IV, U-I, R-III, U-II, and at last U-V.

CONCLUSIONS

The bacterial strains U-II and R-I were highlighted in sustaining plant growth through the accumulation of dry matter and production of IAA.

The result obtained by inoculation of single strain of rhizobacterial isolates demonstrates the potential of these isolates in the wheat growth promotion. Application of the isolates in the future, as commercial inoculants, require additional studies for traits of solubilising of phosphorus, assure the nitrogen consumption or producing of siderophores.

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