THE EFFECT OF SOME RHIZOBACTERIAN STRAINS ON SOLUBLE PROTEINS CONTENT IN SOYBEANS (*GLYCINE MAX* L. MERR.)

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Abstract: Now it is an accepted fact that plant growth-promoting rhizobacteria (PGPR) can increase the productivity of several crops. The main objective of the present study was to find if there are any differences in protein content in the seeds of soybean (*Glycine max* L. MERR.). Using spectrophotometric methods for analyzing the protein contents and electrophoretic methods for qualitative analysis it was observed that no major modifications occur in protein spectrum. Looking at the quantitative side there was a small improvement in protein quantity.

INTRODUCTION

Considering the benefits of intensive agriculture practice in our time and the negative impact of chemical fertilizers and pesticides against the environment, usage of PGPR like biofertilizers is one of the most promising biotechnologies for growing the primary production with less quantity of chemical fertilizers (Bashan, 1998). Usage the isolated bacteria from crop plant's rhizosphere for productivity increase may be an alternative to chemical fertilizers. The main goal is to reduce the pollution and to preserve the environment in the spirit of practicing an ecological agriculture.

Soybeans represent a crop of major economic importance and pesticides are commonly applied to soybeans before planting and during vegetation period. A study was performed to analyze the impact of colonization of the soybean rhizosphere with some rhizospheric bacteria during soybean development. The main purpose was to study the influence of some PGPR like biofertilizers on the soluble protein contents in soybeans.

MATERIAL AND METHODS

From the rhizosphere of soybean (at the beginning of fructification) were isolated, according to morphological characteristics, 8 different bacteria strains (for convention Rs1-Rs8). From the studied strains it was prepared a suspension in sterile distilled water that served as inoculum. The concentration of biopreparation was determined with counter colonies technique (Wistreich, 1997), and it was estimated at 64 x 10^6 CFU/ml. After a previous sterilize of all beans, it was realized an inoculation using a biopreparation immersion only for the beans of the probe lot. Both lots (control and probe) were inseminated using a small experimental seeding machine. The experiment was made at "Ion Ionescu de la Brad" Didactic Station of USAMV, within the "Ezăreni" Didactic Farm, Iaşi, 2005, on a cambic cernoziom with adobe clay texture and good fertility, with moderate humus and highly nitrogen content, with moderate mobile phosphor supply, with highly potassium content and with a very low acid reaction, almost neutral. During vegetation period no chemical fertilizers were supplied to the plants. Only current maintenance operations were performed on the culture. The resulted soybeans were the material on which were performed all the analysis. Soluble protein content was determined according to Bradford method and electrophoretic analysis has been done using SDS-PAGE techniques according to Laemli. The images of the gels were analyzed using specialized software, ImageQuant, from Amersham / GE HealthCare.

RESULTS AND DISCUSSIONS

The initial inoculation with bacterial strains induces in the plants of *Glycine max* L. MERR. an increased capacity of synthesis of proteins. All these proteins were stored in the beans. That was revealed by the analyses that were performed on this material. In this aspect it was observed an increase from 505.59 mg of protein % in the probe to 611.04 mg of protein to the probe in the case of extraction made with distilled water.(Figure 1) When the extraction was made with saline solution (NaCl 1% in distilled water) on the same material that was previously extracted with distilled water the results were in the same shape. Even the quantities of the proteins were almost the same as follows: in control 651.18 mg of protein % and in the probe 699.31 mg of protein %. (Figure 1)

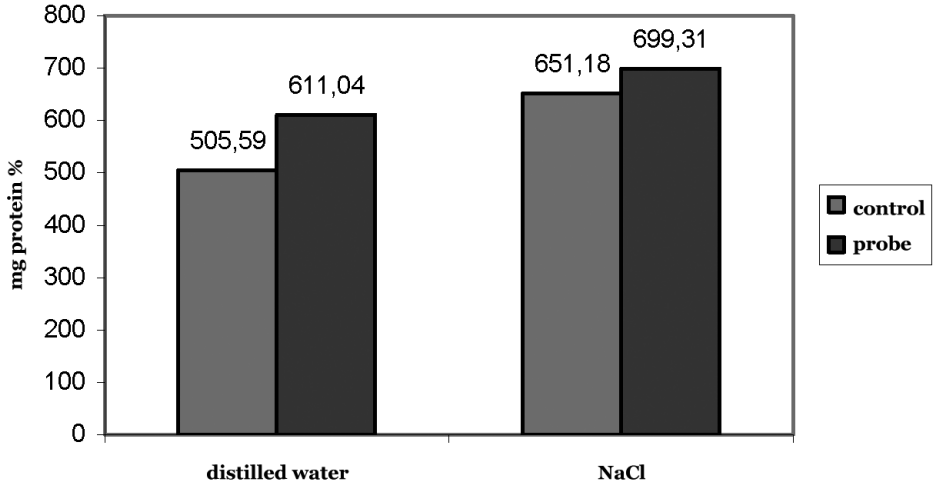

Figure 1 – The total protein content in the beans of *Glycine max* L. MERR. form the „in vivo" crops of the control and probe

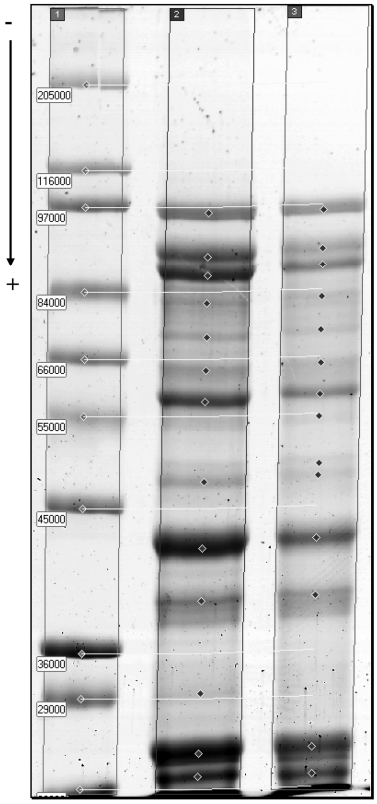

Figure 2 – The electrophoregram of the proteins extracted from the soybeans (1. wide range molecular marker from Sigma; 2. probe; 3. control)

Gel electrophoresis revealed a large number of proteins distributed into a wide area of molecular masses. In both control and probe molecular masses were from 31 kDa up to 105 kDa. (Figure 3 and Figure 4)

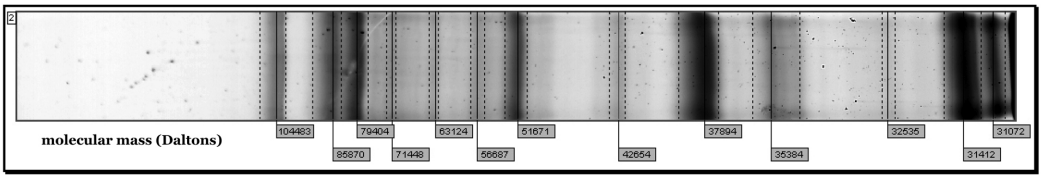

Figure 3 – The spectrum of protein fractions in soybeans - probe

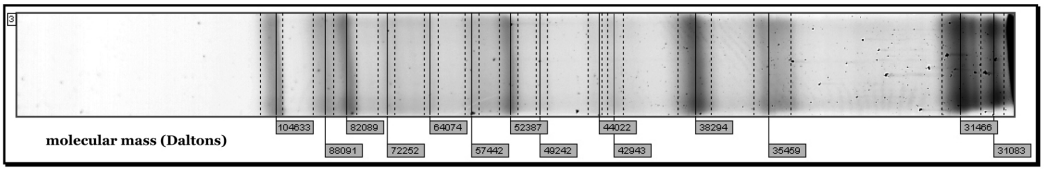

Figure 4 – The spectrum of protein fractions in soybeans – control

The only differences between control and probe were at the quantitative level of the proteins. The analysis didn't revealed many differences in qualitative composition of the extracts, just few fractions which could be artifacts in our determinations.

The quantitative differences observed at the fractions corresponding to the molecular masses of 105, 86, 80, 52, 38, and 31,5 kDa from the probe could be a direct result of the activity of the bacterial strains from the inoculum.

Overall the results that were obtained suggest that at the proteins level there are modifications induced by the bacterial strains selected from the rhizosphere and used as an inoculum. The modifications did not include large changes in the type of the proteins that were synthesized but modifications at the quantitative level. The conclusion is that at the quantitative level there are modifications that include, among others, an increase in the biosynthetic processes which are recovered in the beans as an increased content of proteins.

From the economic perspective, using rhizobacteries selected from the rhizosphere of the culture of *Glycine max* L. MERR. and tested for efficiency could be a measure to improve the nutritional and, finaly, economic value of the crop.

Another point of view could be that the beans with more proteins inside could be a better start for a new crop because more proteins means more energy for the future plant in the process of germination – that is at the time when the plants aren't able to synthesize their own proteins.

CONCLUSIONS

The presence (and activity) of the selected rhizobacterial strains induce an increased biosynthesis of proteins which are stored in the beans of *Glycine max* L. MERR.

The inoculum containing selected rhizobacterial strains doesn't induce any major modification in the spectrum of the proteins from the soybeans.

The rhizobateries selected from the rhizosphere of the roots of *Glycine max* L. MERR. could be used as a mean to increase the quantity and the quality of the crop, with direct, positive, economic implications.

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