EVALUATION OF BACTERIAL COMMUNITY AND OF ACTINOMYCETES COMPOSITION FROM THE *ALLIUM URSINUM* L SPECIES RHIZOSPHERE

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Abstract

Soil s microbiota is an effective recycling instrument of organic matter, of nutritional resources providance for living organisms, an instrument of atmospheric nitrogen fixation in forests biomes. This one changes under the action of some abiotic and biotic factors.

In this research, we have studied the impact of the plant of *Allium ursinum* L, the soil moisture and the way to prepare the soil in the laboratory condition, upon bacterial community and the composition of actinomycetes from the *Streptomyces* genus, from the forest soil.

The soil studied belongs to a deciduous forest, located in the western part of the country. The soil s samples have been taken from the top layer of the soil (0-20 cm), from the *Allium ursinum* L. plants rhizosphere, plants which have been in their blossoming period. The microbian groups of interest have been isolated in specific environments resulted from soil s samples prepared beforehand by plants residue removal (SC) and sifting, as well as from original soil s samples which had no prior preparation (SN).

The results show that the bacteria in the soil sample SC compared to the bacteria in the soil sample SN has dominated numerically. The bacterian species which predominated in both types of soil was *Bacillus cereus* var. *mucoydes*. The actinomycetes have been equal numerically in both soil' samples mentioned below, but their diversity has been reduced. The highest number of species belonging to the genus of *Streptomyces* was isolated from soil sample SC. The species of actinomycetes, common for both soil samples were: *S. albosporeus* and *S. albus*. In the soil sample SC, *S. albosporeus* species dominanted, and in the soil sample SN, *S.chromogenes* was the most representative.

Key words: bacterial community, actinomycetes, *Allium ursinum* L., rhizosphere, forest soil

After Hiltner L. (1904), the rhizosphere is the soil area with intense biological activity, characterized by the interaction of soil-root-microorganisms. According to J.D. Bever (2003), studies, this interaction has impact on soil processes, its biodiversity and on plants. Control of these interactions is done through radicular exudates, which modify the physical, chemical and biological soil properties (Bais H.P. *et al*, 2004; Sanon A. *et al*, 2011). In support of this assertion is Hackl E. *et al* (2004) who observed that forest soils studied in some European countries differ in chemical, nutrient content and microbial biomass.

Allium ursinum L. belongs to the *Amaryllidaceae* family (Friesen N. *et al, 2006;* Chase M.W. *et al*, 2009; Govaerts R., 2011; Sobolewski D. *et al*, 2015) and is one of the plants found in the forest areas of Europe and Asia (Oborny B. *et al, 2011).* According to studies conducted by Djurdjevic L. *et al.* (2004), *A. ursinum* has allelopatic effect.

An important part of the soil microflora is represented by bacterial community, which is estimated at several million or billion cells / g soil, which includes actinomycetes (Goodfellow M., 1983). The major component of the actinomycete population is represented by the genus *Streptomyces*, which is tolerant to acidic pH (Davies F.L., Williams S.T., 1970).

It is known that the microflora of the soil has an important role in the life of the soil, because is involved in the degradation processes of chemical compounds, the execution of the biogeochemical cycles, the humic matter forming, processes for nitrogen fixation and provides nutrients to living organims from this environment (Donnelly P.K. *et al*, 1990; Hu S., Van Bruggen A.H.C., 1997).

Given the role of microflora in the edaphical processes, and relatively little microbial research from forest soils, in this work we have studied bacterial community, including the actinomycetes amount and composition of *Streptomyces* genus from a forest soil, to observe the special influence

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of *A. ursinum* L, soil moisture, and the way of sample preparation procedures in laboratory, on microorganisms.

MATERIAL AND METHOD

Soil samples together with *A. ursinum* L. blooming plants, were collected from 0-20 cm depth, in a deciduous forest, located in the western part of the country. Soils samples preparation and conditioning was carried out in laboratory conditions. Bacteria and actinomycetes were isolated from soil samples prepared in advance by sieving and plant debris removal, but also from soil samples with plant debris. Before receiving the primary microbial isolates, moisture was determined using the thermobalance.

Bacteria were isolated on nutrient agar and actinomycetes on Gause's medium (Ștefanic G., 2006). Bacteria were incubated for 48 hours, and actinomycetes for 7 days at 28^{o}C temperature.

RESULTS AND DISCUSSIONS

The results of bacteria and actinomycetes abundance, as well as the diversity of *Streptomyces* genus species isolated from the rhizosphere of *Allium ursinum* L. after 48 hours and 7 days incubation of respectively are shown in figures 1-3.

After 24 hours, the differences between the two soil samples (SC, SN) concerning the number of bacteria are relatively small. After 48 hours of incubation, there is an evolution of the bacteria number in both soil samples, compared to 0-24 time interval. The highest number of bacteria was encountered in the SC soil sample, both after 24 hours and after 48 hours *(figure 1)*. These statements are supported by the results of Borozan A.B. *et al.* (2013), on medium soil extract agar.

According to Cesarz S. *et al*, (2013) studies, soil microbial processes depend on plant and are influenced by root exudates.

Marschner P. *et al.* (2001), argue that the diversity of bacterial microflora is influenced by plant species, soil type, plus soil sampling points in the rhizosphere. On the other hand, Garbeva P. *et al* (2004), adds to all the above, the interconnections microorganism-soil and microorganism -plant.

There is the possibility that the plant rhizosphere phenolic acids, to influence microorganisms in the ecosystem. Schmidt S.K., Ley R.E. (1999) and Sanon A. *et al.* (2011), argue that the microorganism used phenolic compounds as an energy source, and quickly mineralized them.

Cross T. (1982), argues that humidity is an important factor of the soil, affecting microbiota. Furthermore, Donnelly P.K. *et al* (1990), remind in addition to humidity, two other factors, temperature and acidity, which may influence the degradation processes of cellulose and lignin from forest soils.

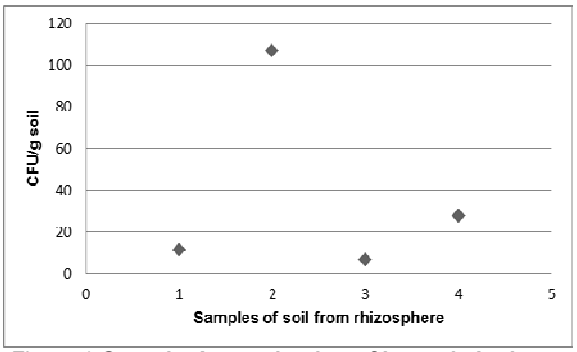

Figure 1 **Quantitative evaluation of bacteria in the *A. ursinum* L plants rhizosphere** (Legend: 1,2 sieved soil; 3,4 un-sieved soil; 1,3-after 24 hours; 2,4-after 48 hours)

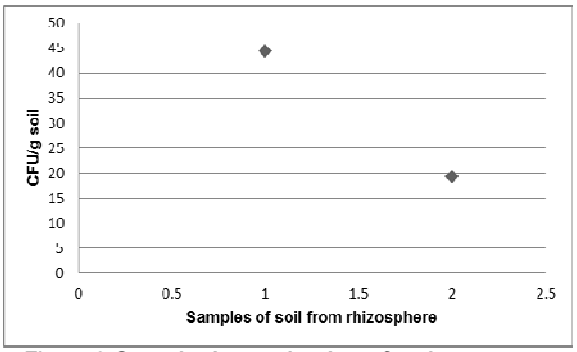

Figure 2 **Quantitative evaluation of actinomycetes from the *A. ursinum* L plants rhizosphere** (Legend: 1-sieved soil; 2-un-sieved soil)

After 7 days of incubation, was observed a significant evolution of actinomycetes amount in SC soil sample, compared to the SN soil sample *(figure 2*).

Range of species belonging to the genus *Streptomyces* is higher in SC soil sample; *S. albus*, and *S. albosporeus* species were isolated from both soil samples. In the SC soil sample prevails *S. albosporeus* species, while in the SN soil sample, is found mainly *S. chromogenes* species *(figure 3).*

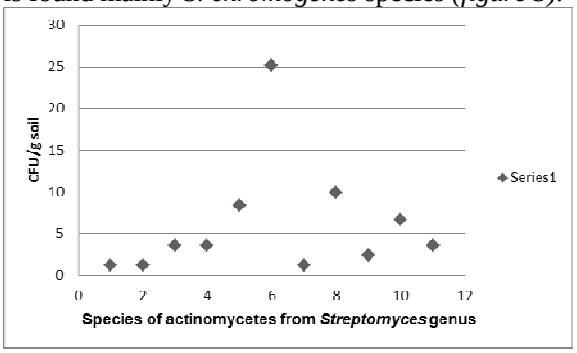

Figure 3 **Species diversity of Streptomyces genus from the *A. ursinum* plant rhizosphere** (Legend: 1,7-sieved soil; 8-11- un-sieved soil; *1.S.olivochromogenes; 2. S.griseus; 3. S.chrysomallus; 4. S.chromogenes; 5. S.albus;6. S.albosporeus; 7. S.aureus; 8. .chromogenes;9. S.albus; 10. S. albosporeus; 11.S.helvolus)*

Some authors noted in their research that the number and composition of actinomycetes depends on geographic area, soil properties and climate conditions (Goodfellow M., O Donnel A.G., 1989; Ghorbani-Nasrabadi R. *et al*, 2013). Brant J.B. *et al* (2006), states that higher differences in the composition of the microbial community in the different forest soils from Europe and the U.S., are seasonal. Many researchers have observed that the population of actinomycetes is higher in agricultural soils compared with forest soils, pastures, etc. (Burck I.C. *et al*, 2003; Bossio D.A. *et al*, 2005; Fierer N. *et al*, 2009). Vieira F.C.S., Nahas E. (2005), showed that nutrient medium and the incubation temperature influence the number of microorganisms in the soil.

CONCLUSIONS

Bacteria and actinomycetes from the studied forest soil, has a significant quantitative evolution in SC soil samples. Differences also exist in species composition belonging to the *Streptomyces* genus. These differences may be due to plant, soil type, moisture, geographical location, season, and the method of soil samples preparation.

The data provided by this research is an important step for quantitative and qualitative evaluation of soil microflora from *A. ursinum* L. species rhizosphere, since such information is hard to find in literature.

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