

## STUDY ON THE EFFECT OF *ALLIUM URSINUM* ON SOIL BACTERIA EVOLUTION

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### ABSTRACT

Romania is included among European countries where *Allium ursinum* species is present. This species has aroused the interest of the research team, because of the many positive aspects it shows, starting from the medical field to the food sector. One of the objectives we have set and managed to capture in this paper refers to soil microorganisms, the environment from which the plant takes the water and nutrients and whose fertility is provided by microbial processes.

The biological material was represented by soil and *Allium ursinum* plants corresponding to each soil sample. Source area of soil and garlic plant is the west part of Romania. Bacterial population was isolated from screened soil samples (without plant debris) but also unscreened (plant residues were not removed), on culture media: soil extract with added nutritive gelose.

Bacterial population abundance studies were performed after 24 and 48 hours of incubation, at the optimum temperature for mesophilic microorganisms.

Although there were no differences in the nutrient substrate used for the study of culture "*in vitro*", the results highlight that in the first 24 hours of incubation the bacterial population clearly dominate in the screened soil sample, compared with the unscreened sample. In the next 24 hours, the existing quantitative bacterial differences between the two samples were significantly reduced.

**Keywords:** bacteria, *Allium ursinum*, rhizosphere, soil, root exudates

### INTRODUCTION

*Allium ursinum* species is widespread throughout Europe, especially in the eastern part of the continent. It develops in forest areas (GRIME ET AL., 1988), in shady deciduous forests (beech, oak, hornbeam) with wet soils, slightly acidic and rich in humus. It is a perennial plant of the Alliaceae family, which blooms in April-June.

The herbaceous part and bulbs of *A. ursinum* are used in homeopathic medicine because of the germicide antiparasitic and antiseptic properties, (CAROTENUTO ET AL., 1996 CHYBOWSKI, 1997). It is also used as a condiment or in different food preparations (BOSS-TEICHMANN, 2009).

One of the important factors for this species is humidity (KOVÁCS, 2007). KESIK ET AL. (2011) revealed that optimal conditions for *A. ursinum* are ensured by mulching the soil with pine bark and moderate nitrogen fertilization.

Forest litter covering the soil has a protective role against thermal fluctuations, prevents water loss and positively influences soil aggregation state (BALEWICZ-WOZ NIAKI AND KESIK, 2010; KESIK ET AL., 2006).

Weight gain and elongation of this species leaves is influenced by nitrogen nutrition (MAIRAPETYAN ET AL., 1999; DZIDA AND PITURA, 2008). *A. ursinum* leaves are rich in nitrates. According to the literature there is a close temporal connection between photosynthetic process that involves the vegetation and activities that occur in forest soil (HÖGBERG ET AL., 2008). According to those authors, half of the biological activities occurring in forest soil are supported by photosynthetic process conducted by trees, but there are no studies on carbon flux from canopy to soil microorganisms.

The knowledge on the influence of plant roots on soil degradation processes are limited, but are required for evidence of soil carbon dynamics (CESARZ ET AL., 2013). The degradation of vegetal residues on the soil is initiated by their associated flora, especially by fungi. First are degraded the low molecular weight compounds by bacteria and fungi. Among fungi, an essential role in the degradation of plant structural components is held by basidiomycetes. This process is continued for additional saprophytic flora, represented by bacteria, fungi and by saprophagous fauna. Soil fauna and macroflora determine the soil structure and architecture (LAVELLE, 2002).

Mezofauna accelerates fragmentation of litter and creates favorable conditions for the development of microflora and consequently the mineralization process, leading to improvement of soil nutrient reserves. Decomposition is accompanied by the synthesis of soil organic matter (LAVELLE AND SPAIN, 2001; SCHAEFER, 2009).

In this work the evolution of bacterial microflora in forest soil under the direct influence of the *Allium ursinum* species root exudates, was studied.

## MATERIAL AND METHOD

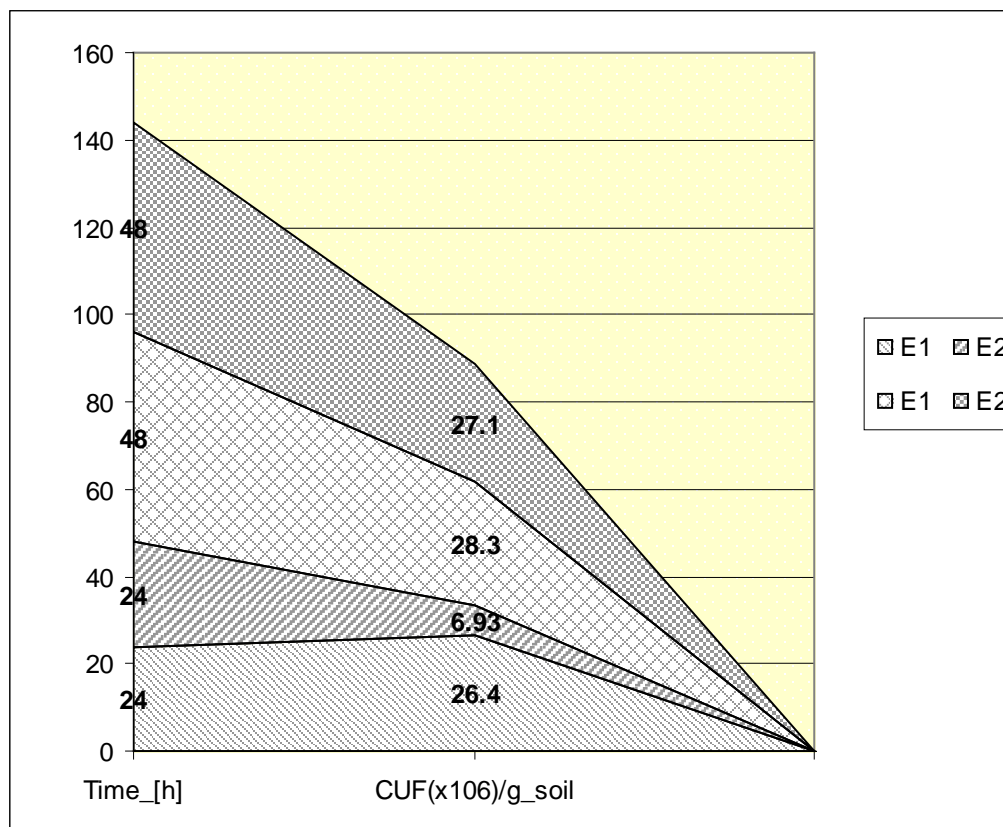
The biological material was represented by ten soil samples accompanied by corresponding plants from a deciduous forest, located in the western part of Romania. Sampling was conducted in the fall season, from the depth of 0-20 cm. The processing of soil samples was conducted under laboratory conditions.

Isolation and quantitative determination of bacterial populations in soil was done from processed soil, screened and without plant debris, and also from the unprocessed soil, unscreened, with plant debris that have been not removed. The nutrient medium used for bacterial seeding was represented by a mixture: soil extract and nutritive gelose (STEFANIC, 2006). The bacterial inoculum was prepared following dilution method. Inoculation was performed at a 1:6 dilution. Bacterial cultures were incubated at 28°C for 24 and 48 hours, their abundance being measured and statistically interpreted.

For statistical interpretation of data the statistical software package PAST 2.14, was used (HAMMER ET AL., 2001).

## RESULTS

Results regarding bacterial abundance and also distribution from *Allium ursinum* species rhizosphere in the two types of samples after 24 and 48 hours of incubation are emphasized in Figures 1 and 2.



**Figure 1. Evolution of bacterial population depending on time interval**

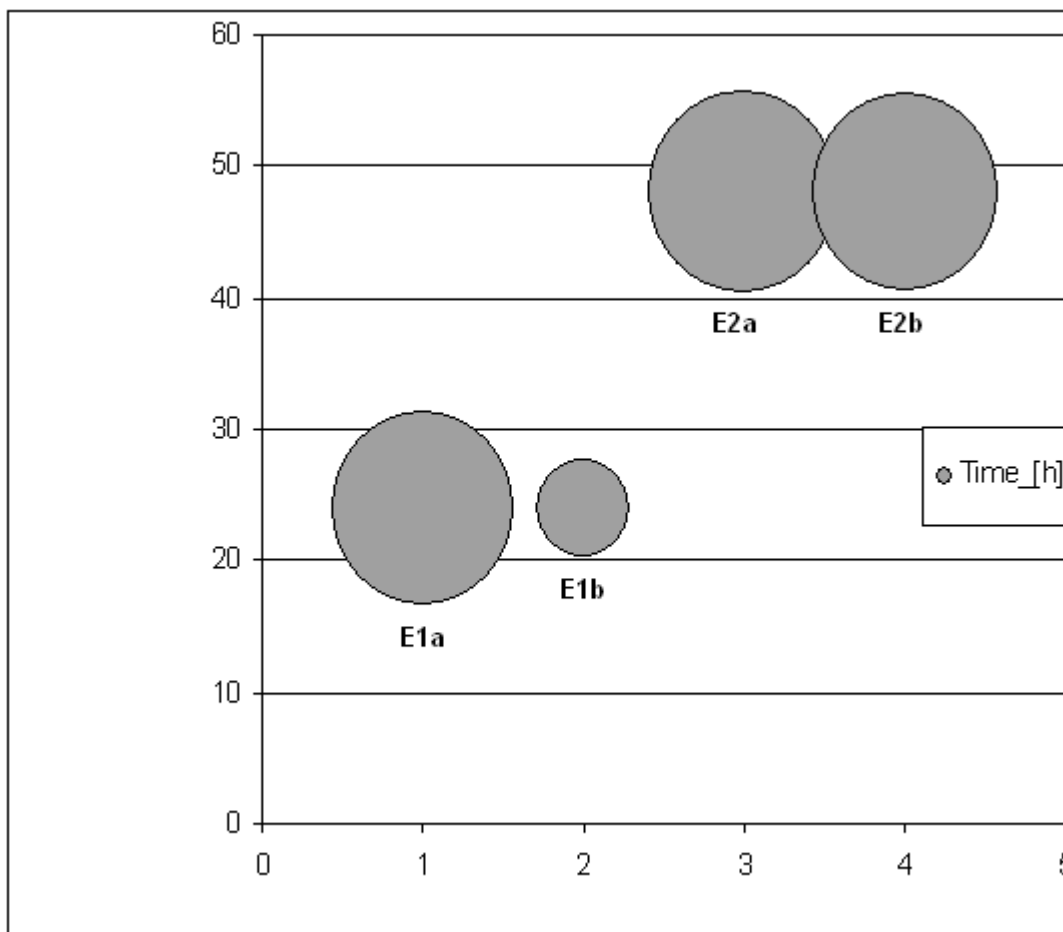
(24 hours: E<sub>1</sub>- screened ground, E<sub>2</sub>- unscreened ground, 48 hours: E<sub>1</sub>- screened ground, E<sub>2</sub>- unscreened ground)

In Fig. 1 it can be noticed an evolution of CUF/g soil in 0-48 hours time interval in both types of samples.

Bacteria from the screened sample, free of plant debris dominated numerically compared with raw sample in which plant residues were not removed. The large number of CUF/g from screened soil sample was kept high after 24 hours and also after 48 hours compared the unscreened soil sample.

After CESARZ ET AL (2013), soil processes are controlled by rootlet exudates and are differentiated by the plant type (for wood species *Fagus sylvatica L.* and *Fraxinus excelsior L.* studied by the mentioned authors). Of the two woody plants, *Fagus sylvaticus* has a greater impact on processes in the soil. Changes in soil layer are associated with microflora from plants rhizosphere and carbon dynamics.

Besides reducing rhizosphere microbial activity is not always accompanied by changes in microbial community composition. Sometimes the presence of groups of organisms (e.g. arbuscular mycorrhizal fungi) can stimulate or inhibit some bacterial groups (VESTERGARD ET AL., 2008).



**Figure 2. Distribution of bacterial populations in the two types of samples after 24 and 48 hours of incubation**

(E<sub>1</sub>a- screened ground, 24 hours incubation and E<sub>1</sub>b- unscreened ground, 24 hours of incubation, E<sub>2</sub>a- screened ground, 48 hours incubation and E<sub>2</sub>b-unscreened ground, 24 hours of incubation)

Although CUF/g soil values were maintained in larger numbers in screened sample, even after 48 hours, quantitative bacterial growth is significant in the 24-48 hours interval for unscreened soil sample compared with screened sample, in which there is an insignificant increase in this period. In *Figure 2*, it is shown that at the end of the incubation interval, very small quantitative differences between the two types of samples are unnoticed.

## CONCLUSIONS

There was an increase in CUF/g soil in both soil samples with increasing the incubation interval.

Although in 0-24 hours time interval CUF/g from unscreened soil sample were numerically reduced, after another 24 hours of incubation a quantum leap is observed, close to that present in the sieved soil sample.

After 48 hours of incubation, bacterial microflora shows no significant differences in terms of quantity, even if the sample preparation protocol is different.

Influence of rootlet exudates on soil microflora has been reported by many authors, some of whom are mentioned in the pages of this work (e.g. CESARZ ET AL., 2013, RANGEL-CASTRO.ET AL., 2005).

## REFERENCES

- BAŁEWICZ-WOŹNIAK, K., (2010): Indicators of soil aggregation and their changes in conservation tillage for onion. *Acta Sci. Pol., HortorumCultus* 9(1), 73–85.
- BOSS-TEICHMANN, C. (2009): *Czosnek niedźwiedzi*. Wyd. Bellona, Warszawa, ISBN 978-83-11-11472-2, 24–31.
- CAROTENUTO A., DE FEO V., FATTORUSSO E., LANZOTTI V., MAGNO S., CICALA C. (1996): The flavonoids of *Allium ursinum*. *Phytochemistry* 41(2), 531–536.
- CESARZ, S., FENDER, A., BEYER, F., VALTANEN, K. PFEIFFER, B., GANSERT, D., HERTEL, D., POLLE, A., DANIEL, R., LEUSCHNER, C., SCHEU, S., (2013): Roots from beech (*Fagus sylvatica* L.) and ash (*Fraxinus excelsior* L.) differentially affect soil microorganisms and carbon dynamics, *Soil Biology and Biochemistry*, Vol. 61, 23–32
- CHYBOWSKI, J. (1997): Badanie aktywności przeciwbaczej wyciągów czosnkowych. *Herba Polonica* 43, 4, 383–387.
- DZIDA, K., PITURA, K., (2008): The influence of varied nitrogen fertilization on yield and chemical composition of Swiss chard (*Beta vulgaris* L. var. *cicla*L.). *Acta Sci. Pol. HortorumCultus*, 7(3), 15–24.
- GRIME, J.P., HODGSON, J.G., HUNT, R. (1988): *Comparative plant ecology – a functional approach to common British species*. – Unwin Hyman, London.
- HAMMER, Q., HARPER, D.A.T., RYAN, P.D. (2001): PAST: Paleontological Statistics Software Package for Education and Data Analysis. *Palaeontologia Electronica* 4(1): 9pp.
- HÖGBERG, P., HÖGBERG, M.N., GÖTTLICHER, S.G., BETSON, N.R., KEEL, S.G., METCALFE, D.B., CAMPBELL, C., SCHINDLBACHER, A., HURRY, V., LUNDMARK, T., LINDER, S., NÄSHOLM, T., (2008), High temporal resolution tracing of photosynthate carbon from the tree canopy to forest soil microorganism, *New Phytologist*, 177: 220–228
- KĘSIK, T., KONOPIŃSKI, M., BŁAŻEWICZ-WOŹNIAK, M., (2006 a): Wpływ uprawy przedzimowej i mulczu roślin okrywających na retencję wody, zagęszczenie i porowatość dyferencyjną gleby poprzemulczaniu. *Acta Agrophysica* 7(4), 135, 915–926.
- KĘSIK, T., KONOPIŃSKI, M., BŁAŻEWICZ-WOŹNIAK, M. (2006 b): Influence of plantmulch and prewinter soil tillage on physical properties of soil, emergence and yielding of onion. IX ESA Congress 4–7 september 2006, Warszawa, Poland. *Bibl. Fragm. Agron.* 11, 2, 545–546.
- KĘSIK, T., BŁAŻEWICZ-WOŹNIAK, M., MICHOWSKA, A.E. (2011) Influence of mulching and nitrogen nutrition on bear garlic (*Allium ursinum* L.) growth *Sci. Pol., Hortorum Cultus* 10(3), 221-233
- KOVÁCS, J.A. (2007): Data on the vegetation biology and coenological relations of *Allium ursinum* L. stands in Eastern Transsylvania. – *Kanitzia* 15: 63–76.
- LAVELLE, P., SPAIN, V.A. (2001): *Soil ecology*. Kluwer Academic
- LAVELLE, P. (2002): FUNCTIONAL DOMAINS IN SOILS. *ECOLOGICAL RESEARCH*, 17, 4, PP. 441–450
- MAIRAPETYAN, S.K., TADEVOSYAN, A.H., ALEXANYAN, S.S., STEPANYAN, B.T., (1999): Optimalization of the N:P:K ratio in the nutrient medium of some soilless aromatic and medicinal plants. *Acta Hort.*, 502, 29–32.

RANGEL-CASTRO IGNACIO , KILLHAM KEN, OSTLE NICK, NICOL GRAEME, ANDERSON IAN, SCRIMGEOUR CHARLIE, INESON PHIL, MEHARG ANDY AND PROSSER JIM, (2005): Stable isotope probing analysis of the influence of liming on root exudate utilization by soil microorganisms, *Environmental Microbiology* 7(6), 828–838

SCHAEFER, M., MIGGE-KLEIAN, S., SCHEU, S. (2009): The Role of Soil Fauna for Decomposition of Plant Residues, *Ecological Studies* Vol. 208, pp 207-230

STEFANIC, G. (2006): Metode de analiza a solului (biologica, enzimatica si chimica). Probleme de agrofitotehnie teoretica si aplicata. Institutul de Cercetare-Dezvoltare Agricola Fundulea, XXVIII, pp. 5-25.

VESTERGARD, M., FREDERIC, H., RANGEL-CASTRO, J.I., MICHELSEN, A., PROSSER, J., CHRISTENSEN, S., (2008): Rhizosphere bacterial community composition responds to arbuscular mycorrhiza, but not to reductions in microbial activity induced by foliar cutting, *FEMS Microbiol Ecol* 64, 78–89.