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# MICROBIOLOGICAL ACTIVITY IN THE SOIL OF VARIOUS AGRICULTURAL CROPS IN ORGANIC PRODUCTION<sup>1</sup>

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Summary: The purpose of this study is to investigate the microbial activity and the number of different groups of microorganisms in the soil under organic agricultural systems.

A range of analyses was conducted on soil samples taken from calcareous chernozem soils managed under organic (7 sites) and conventional agricultural systems (1 site). Laboratory measurements were performed in the Laboratory of Microbiology, Faculty of Agriculture, Novi Sad. The total number of bacteria, actinomycetes, fungi, aminoheterotrophs and azotobacters was determined using the dilution method. Soil dehydrogenase activity was measured spectrophotometrically.

The greatest number of the Azotobacter sp. bacteria was recorded in the soil devoted to pumpkins  $(132.61 \times 10^2)$  and in the soil devoted to apples (126.39  $\times$  10<sup>2</sup>). The greatest number of aminoheterotrophs (1786.05  $\times$  10<sup>6</sup>) and the total number of bacteria  $(1370.82 \times 10^6)$  and actinomycetes  $(235.45 \times 10^4)$  were determined in the soil devoted to carrots. Fungi were more abounded in the soil devoted to chard  $(36.82 \times 10^4)$  than in the soil devoted to other plants. The research results show that the soil devoted to wheat in organic production indicated a greater number of aminoheterotrophs, total bacteria, actinomycetes and fungi, whereas only the number of Azotobacter sp. was greater in the soil devoted to wheat in the conventional agricultural system. The highest dehydrogenase activity level was determined in the soil devoted to radishes, whereas the lowest dehydrogenase activity level was determined in the soil devoted to apples.

Key words: soil, microorganisms, rhizosphere, organic production

## INTRODUCTION

Soil is a highly complex system in which plants and macro-, meso- and micro-organisms compete for water and nutrient sources (Melo, 1994). All these forms of life interact with one another and with the soil to create continually changing conditions.

Soil microorganisms, such as bacteria and fungi, control the ecosystem via decomposition and nutrient cycling, and may serve as indicators of land-use change and ecosystem health (Doran and Zeiss, 2000, Waldrop et al., 2000, Yao et al., 2000). According to Powlson et al. (1987), the main function of microorganisms is to mediate soil processes and present high rates of turnover, being a sensitive indicator of changes in the soil organic matter. Moreover, the microbial biomass is a main living component of the soil organic matter, which is responsible for maintaining and improving the quality of soil. Although small in mass, the microbial biomass is among the most labile pools of organic matter, and thus serves as an important reservoir of plant nutrients, such as N and P (Jenkinson and Ladd, 1991, Marumoto et al., 1982). Therefore, the soil microbial biomass, activity and number have been suggested as indicators of the soil quality and fertility. Conversely, soil microbial activities, populations and communities are governed by the environmental variables (i.e. soil moisture, temperature, physical disturbance of the soil, etc.), interactions with soil fauna, amount and quality of carbon and other nutrients available from plant residues, root exudates, and agricultural systems (conventional or organic) (Melero et al., 2005, Araujo et al., 2008).

The purpose of this study is to investigate the microbial activity and the number of different groups of microorganisms in the rhizospheric soil of different plants managed under the organic agricultural system.

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#### MATERIAL AND METHODS

A range of analyses was conducted on soil samples taken from soils managed under organic and conventional agricultural systems (8 different sites). In addition to chard, carrots and radishes in a greenhouse, wheat, apples, cabbage and pumpkins in the open fields were also managed under the organic production system, whereas wheat in the open field was managed under the conventional farming system. Soil samples were taken from a depth of 0-30 cm. A total of four samples were taken from each site for analysis. The soil samples collected were placed in sterile polyurethane bags and kept at a temperature of 4°C. The samples were sieved through a 2 mm sieve and stored until used.

Laboratory measurements were performed in the microbiological laboratories of the Faculty of Agriculture, Novi Sad. The microbiological analyses included determining the total number of bacteria, actinobacteria, fungi, aminoheterotrophs and azotobacters. The number of microorganisms was determined using the dilution method (Trolldenier, 1996). The following (appropriate) nutrient media were used (Hi Media Laboratories Pvt. Limited, Mumbai, India): nutrient agar for the total number of bacteria, synthetic agar for the number of actinomycetes, potato dextrose agar for the number of fungi, meat peptone agar for the number of aminoheterotrophs, and mannitol salt agar for the number of azotobacters. After incubation, the plates were inspected. The dehydrogenase activity of soil was determined spectrophotometrically (Lenhard, 1956, Thalmann, 1968).

The number of microorganisms was calculated in one gram of absolutely dry soil and presented as the CFUg<sup>-1</sup> soil. The number of microorganisms is expressed in a *logarithmic* graph. The significance of the difference between the organic and conventional agricultural production was determined using the Fisher's LSD test. The data were statistically processed by means of the Statistics StatSoft 13.2 program. The dehydrogenase activity recorded is presented graphically.

#### RESULTS AND DISCUSSION

This research was focused on examining the microbial activity and the number of microorganisms in the rhizospheric soil of different plants. The results obtained showed that the number of microorganisms, with the exception of *Azotobacter* sp. and fungi, was the highest in the rhizospheric soil of carrots (Figure 1).

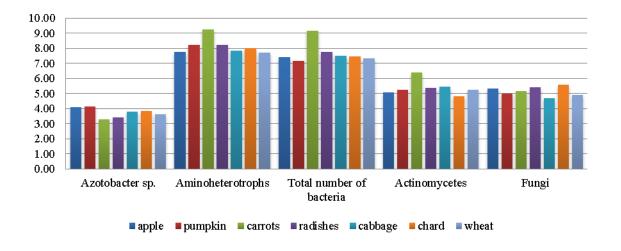


Figure. 1. The number of different groups of microorganisms in the soil managed under the organic agricultural system (Log Num)

Azotobacter is a genus of usually motile, oval or spherical bacteria that form thick-walled cysts, and may produce large quantities of capsular slime. Azotobacter is an aerobic, free-living soil microbe which fixes nitrogen from the atmosphere. Results of many studies indicate positive effects of Azotobacter sp. on plant growth (Stamenov et al., 2012). In this study, the number of Azotobacter sp. was between  $132.61 \times 10^2$  and  $18.96 \times 10^2$  CFUg<sup>-1</sup> soil. The largest number of this species was detected in the soil devoted to apples and pumpkins, whereas the smallest number of Azotobacter sp. was determined in the soil devoted to carrots.

Aminoheterotrophs comprise a large group of bacteria and fungi that transform proteins and other organic nitrogen compounds. As ammonia is released during the course of these processes, aminoheterotrophs are also referred to as ammonifiers, and the process of transformations in which they are involved as ammonification. The number of aminoheterotrophs in the soil samples analyzed was between  $1786.05 \times 10^6$  and  $67.73 \times 10^6$  CFUg<sup>-1</sup> soil. The greatest number of aminoheterotrophs was recorded in the soil devoted to carrots, whereas the lowest in the rhizospheric soil of wheat.

The total number of bacteria is a good indicator of soil fertility. This research showed that the total number of bacteria was also the greatest in the soil devoted to carrots (13.  $71 \times 10^8 \, \text{CFUg}^{-1}$  soil), whereas the lowest number of bacteria was detected in the rhizospheric soil of pumpkins.

Actinomycetes constitute a major constituent of the microbial populations present in the soil. They belong to an extensive and diverse group of Gram-positive, aerobic, mycelial bacteria that play important ecological roles in soil nutrient cycling (Halder et al., 1991). Most actinomycetes in the soil belong to the genus *Streptomyces* (Suzuki et al., 2000) and 60% of most biologically active compounds, such as antifungal and antibacterial compounds or plant growth promoting substances, that have been developed for agricultural purposes originate from this genus (Ilic et al., 2007). In this paper, the number of actinomycetes was between 235.45 x 10<sup>4</sup> and 6.69 x 10<sup>4</sup> CFUg<sup>-1</sup> soil. The greatest number of actinomycetes was determined in the rhizospheric soil of carrots, whereas the smallest was determined in the soil devoted to chard.

Fungi play a very important role in immobilizing and retaining nutrients in the soil. They produce organic acids which facilitate creating the soil organic matter resistant to degradation. Many fungi develop a mutually beneficial relationship with plants. The results obtained show that the largest number of fungi was in the rhizospheric soil of chard  $(36.82 \times 10^4 \text{ CFUg}^{-1} \text{ soil})$ . The smallest number of fungi was detected in the soil devoted to cabbage.

Upon comparing the organic and conventional production of wheat, it was noticed that the number of microorganism groups, with the exception of the Azotobacter sp., was greater in the soil managed under the organic farming system (Figure 2). However, these differences proved to be statistically non-significant at the level p<0.05.

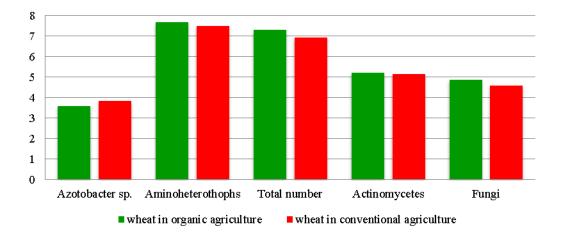


Figure. 2. The number of different groups of microorganisms in the rhizospheric soil of wheat managed under organic and conventional agriculture systems (Log Num)

The number of certain groups of microorganisms and their enzymatic activity depend on the variability of soil properties, as well as the level of anthropogenic influences (Nannipieri et al., 2003). Previous results suggest that different practices in the conventional production system can disrupt microbial activity and abundance. This is in accordance with the results of Cinnadurai et al. (2013). These authors showed that cultural practices can affect the function, community structure, and population of soil microorganisms.

Dehydrogenase is an enzyme that occurs in all viable microbial cells. This enzyme defines the metabolic state of soil microorganisms (Watts et al., 2010). Dehydrogenase activity (DHA) is one of the most adequate, important and sensitive bioindicators related to soil fertility (Wolinska and Stepniewska, 2012). The dehydrogenase activity values obtained in this study ranged between 3853.09 and 305.39 µg TPF 10g<sup>-1</sup> soil (Figure 3).

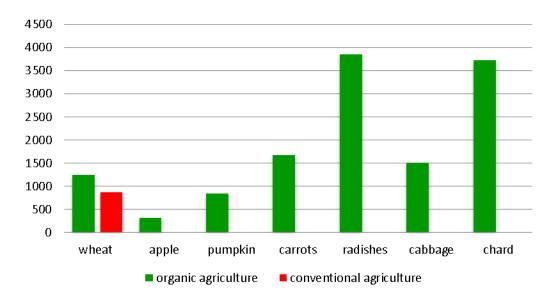


Figure. 3. Dehydrogenase activity in the rhizospheric soil of different plants (µg TPF/10g dry soil)

The highest value of dehydrogenase activity was measured in the rhizospheric soil of radishes (3853.09  $\mu$ g TPF  $10g^{-1}$  soil) and chard (3721.78  $\mu$ g TPF  $10g^{-1}$  soil), whereas the lowest value was measured in the soil devoted to apples (305.39  $\mu$ g TPF  $10g^{-1}$  soil) and pumpkins (836.26  $\mu$ g TPF  $10g^{-1}$  soil).

Different numbers of microorganisms and different microbiological activities in the rhizosphere of the crops analyzed suggest the influence of root exudates on the number, activity and variety of microorganisms in the soil. A number of studies have assessed the diversity of microbial populations in the rhizosphere of different plant species, including wheat (Miller et al., 1989) clover and ryegrass (Darbyshire and Greaves, 1967, Stamenov et al., 2012), grasses (Lawley et al., 1983), maize (Hajnal, 2010), cabbage and peppers (Simonida et al., 2005). In the study of Simonida et al. (2005), the total number of microorganisms in the rizospheric soil of cabbage was 1.2 x 10<sup>8</sup> CFUg<sup>-1</sup> soil, which is lower than the value obtained in our study (cabbage 7.45 x 10<sup>8</sup> CFUg<sup>-1</sup> soil). This difference can be accounted for by different agricultural production systems and plant varieties. In several other studies, the influence of plant species on the rhizosphere microbial communities was also argued (Briones et al., 2002, Graner et al., 2003, Milling et al., 2004). Plant rhizosphere is the soil nearest to the plant root system where roots release large quantity of metabolites from living root hairs or fibrous root systems. These metabolites act as chemical signals for motile bacteria to move to the root surface, and represent the main nutrient source available to the plant to support its growth and persistence in the rhizosphere. At the moment of exudation and thereafter, organic materials are subject to microbial attacks. According to Grayston et al. (1998), the specific content of root exudates may create a niche that influences which microorganisms are to colonize the rhizosphere, thereby altering the composition and diversity of microorganisms colonizing the rhizosphere. Therefore, plant species, plant developmental stages, soil types and production systems have been indicated as major factors in determining the composition of rhizosphere microbial communities (Broeckling et al., 2008).

The results of these studies contribute to a better understanding of the rhizosphere and the influence of the plant variety, as well as the type of agricultural system of production, on the number and activity of microorganisms. A better understanding of the basic principles of the rhizosphere ecology, including the function and diversity of inhabiting microorganisms, would be a good base for an appropriate use of microorganisms in the sustainable production of different agricultural plants.

### **CONCLUSION**

The greatest number of the *Azotobacter* sp. bacteria was recorded in the soil devoted to pumpkins and apples. The greatest number of aminoheterotrophs, the total number of bacteria and actinomycetes was determined in the soil devoted to carrots. Fungi were more abundant in the soil devoted to chard than in the soil devoted other plants.

Upon comparing the organic and conventional production of wheat, it was noticed that the number of microorganism groups, with the exception of the *Azotobacter* sp., was greater in the soil managed under the organic farming system.

The highest dehydrogenase activity was determined in the soil devoted to radishes, whereas the soil devoted to apples indicated the lowest dehydrogenase activity.

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#### REFERENCES

ARAUJO, A.S.F., SANTOS, V.B., MONTEIRO, R.T.R.: Responses of soil microbial biomass and activity for practices of organic and conventional farming systems in Piauí state. Brazil. *European Journal of Soil Biology*, 44: 225-230, 2008.

BRIONES, A.M., OKABE, S., UMEMIYA, Y., RAMSING, N., REICHARDT, W., OKUYAMA, H.: Influence of different cultivars on populations of ammonia-oxidizing bacteria in the root environment of rice. Applied Environmental Microbiology, 68: 3067–3075, 2002.

BROECKLING, C.D., BROZ, A.K., BERGELSON, J., MANTER, D.K., VIVANCO, J.M.: Root exudates regulate soil fungal community composition and diversity. *Applied* and *Environmental Microbiology*, 74: 738-744, 2008.

CINNADURAI, C., GOPALASWAMY, G., BALACHANDAR, D.: Diversity of cultivable Azotobacter in the semi-arid alfisol receiving long-term organic and inorganic nutrient amendments. Annals of *Microbiology*, 163: 1397–1404, 2013.

DARBYSHIRE, J.F., GREAVES, M.P.: Protozoa and bacteria in the rhizosphere of *Sinapis alba* L., *Trifolium repens* L., and *Lolium perenne* L. Canadian Journal of Microbiology, 13: 1057-1068, 1967.

DORAN, J.W. and ZEISS, M.R.: Soil health and sustainability: managing the biotic component of soil quality. *Applied Soil Ecology*, 15(1): 3–11, 2000.

ĐURIĆ, S., NAJDENOVSKI, O., ĐORĐEVIĆ, S., MITKOVA, T., MARKOSKI, M.: Microbiological activity in rhiyospheric soil of different cultures, Letopis naučnih radova, 28, 1, 110-115, 2004.

GRANER, G., PERSSON, P., MEIJER, J., ALSTROM, S.: A study on microbial diversity in different cultivars of *Brassica napus* in relation to its wilt pathogen, *Verticillium longisporum*. FEMS Microbiol Letter, 29: 269–276, 2003.

GRAYSTON, S.J., WANG, S.Q., CAMPBELL, C.D., EDWARDS, A.C.: Selective influence of plant species on microbial diversity in the rhizosphere. *Soil Biology* and *Biochemistry*, 30: 369-378, 1998.

HAJNAL, T.: Uticaj inokulacije na prinos i mikrobiološku aktivnost u zemljištu pod usevom kukuruza, Doktorska disertacija, Poljoprivredni fakultet, Novi Sad, 2010.

HALDER, A.K., MISHRA, A.K., CHAKARBARTHY, P.K.: Solubilization of inorganic phosphates by *Bradyrhizobium*. Indian Journal of Experimental Biology, 29: 28–31, 1991.

ILIC, S.B., KONSTANTINOVIC, S.S., TODOROVIC, Z.B., LAZIC, M.L., VELJKOVIC, V.B., JOKOVIC, N., RADOVANOVIC, B.C.: Characterization and antimicrobial activity of the bioactive metabolites in streptomycete isolates. Microbiology, 76: 421-428, 2007.

JENKINSON, D.S. and LADD, J.N.: Microbial biomass in soil: measurement and turnover. In: Soil Biochemistry (E. A. Paul and J. N. Ladd, Marcel Dekker, eds.). New York, 5, 415-471, 1991.

LAWLEY, R.A., CAMPBELL, R., NEWMAN, E.I.: Composition of the bacterial flora of the rhizosphere of three grassland plants grown separately and in mixtures. Soil Biology & Biochemistry, 15, 605-607, 1983.

LENHARD, G.: The dehydrogenase activity in soil as a measure of activity of soil microorganisms, Z. Pflanzenernaehr. Dueng. Bodenkd, 73: 1-11, 1956.

MARUMOTO, T., ANDERSON, J.P.E., DOMSCH, K.H.: Mineralization of nutrients from soil microbial biomass. Soil Biology and Biochemistry, 14: 469-475, 1982.

MELERO, S., PORRAS, J.C.R., HERENCIA, J.F., MADEJON, E.: Chemical and biochemical properties in a salty loam soil under conventional and organic management. *Soil* and *Tillage* Research, 90: 162-170, 2005.

MELO, W.J.: Manejo: aspectos biológicos. In: PEREIRA, V.P. (Ed). Solos altamente susceptíveis à erosão: manejo visando à recuperação dos solos altamente susceptíveis à erosão. Jaboticabal, FCAV/UNESP/SBCS, 123-148, 1994.

MILLER, H.J., HENKEN, G., VAN VEEN, J.A.: Variation and composition of bacterial populations in the rhizospheres of maize, wheat and grass cultivars. Canadian Journal of Microbiology, 35, 656-660, 1989.

MILLING, A., SMALLA, K., XAVER, F., MAIDL, K., SCHLOTER, M., MUNCH, J.C.: Effects of transgenic potatoes with an altered starch composition on the diversity of soil and rhizosphere bacteria and fungi. Plant Soil, 266: 23–39, 2004.

NANNIPIERI, P., ASCHER, J., CECCHERINI, M.T., LANDI, L., PIETRAMELLARA, G., RENELLA, G.: Microbial diversity and soil functions, *European Journal of Soil Science*, 54: 665-670, 2003.

POWLSON, D.S., BROOKES, P.C., CHRISTENSEN, B.T.: Measurement of soil microbial biomass provides an early indication of changes in total soil organic matter due to straw incorporation. Soil Biology and Biochemistry, 19: 159-164, 1987.

STAMENOV, D., JARAK, M., ĐURIĆ, S., HAJNAL-JAFARI, T., ANĐELKOVIĆ, S.: The Effect of Azotobacter and Actinomycetes on the Growth of English Ryegrass and Microbiological Activity in Its Rhizosphere, Research Journal of Agricultural Science, 44 (2): 93-99, 2012.

SŪZUKI, S., YAMAMOTO, K., OKUDA, T., NISHIO, M., NAKANISHI, N., KOMATSUBARA, S.: Selective isolation and distribution of *Actinomadura rugatobispora* strains in soil. Actinomycetologic, 14: 27-33, 2000.

THALMANN, A.: Zur methodik des bestmmung des dehydrogenase activitat im bodenmittels TTC. Landwirtschaftliche Forschung, 21: 249-258, 1968.

TROLLDENIER, G.: Plate Count Technique. In Methods in Soil Biology (Franz Schinner, Ellen Kandeler, Richard Ohlinger, Rosa Margesin, eds.). Springer-Verlag, Berlin Heildeberg, 20-26, 1996.

WALDROP, M.P., BALSER, T.C., FIRESTONE, M.K.: Linking microbial community composition to function in a tropical soil. Soil Biology and Biochemistry, 32(13): 1837–1846, 2000.

WATTS, D.B., TORBERT, H.A., FENG, Y., PRIOR, S.A.: Soil microbial community dynamics as influenced by composted dairy manure, soil properties, and landscape position. Soil Science, 175: 474–486, 2010.

WOLINSKA, A., ŚTÉPNIEWSKA, Z.: Dehydrogenase activity in the soil environment. In: Canuto R.A. (ed.): Dehydrogenases. Intech, Rijeka. 2012.

YAO, H., HE, Z., WILSON, M.J., CAMPBELL, C.D.: Microbial biomass and community structure in a sequence of soils with increasing fertility and changing land use. *Microbial Ecology*, 40(3): 223–237, 2000.

# MIKROBIOLOŠKA AKTIVNOST U ZEMLJIŠTU RAZLIČITIH POLJOPRIVREDNIH KULTURA U ORGANSKOJ PROIZVODNJI

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**Izvod:** Cilj ovih istraživanja bio je ispitivanje mikrobne aktivnosti i brojnosti različitih grupa mikroorganizama u zemljištu pod organskim sistemom poljoprivrede.

Istraživanja su sprovedena na karbonatnom černozemskom zemljištu, u organskom poljoprivrednom (7 lokacija) i konvencionalnom sistemu (1 lokacija). Laboratorijska istraživanja su vršena u Laboratoriji za mikrobiologiju Poljoprivrednog fakulteta u Novom Sadu. Ukupan broj bakterija, broj aktinomiceta, gljiva, aminoheterotrofa i azotobaktera određen je metodom agarnih ploča. Aktivnost dehidrogenaze u zemljištu je merena spektrofotometrijski.

Najveći broj Azotobacter sp. je zabeležen u zemljištu pod bundevom  $(132.61 \times 10^2)$  i u zemljištu pod jabukom  $(126.39 \times 10^2)$ . Najveći broj aminoheterotrofa  $(1786.05 \times 10^6)$ , ukupan broj bakterija  $(1370.82 \times 10^6)$  i aktinomiceta  $(235.45 \times 10^4)$  utvrđen je u zemljištu pod šargarepom. Utvrđena je veća brojnost gljiva u zemljištu pod blitvom u odnosu na zemljište pod drugim biljkama. Istraživanje je pokazalo da je zemljište pod pšenicom u organskoj proizvodnji imalo veći broj aminoheterotrofa, ukupan broj bakterija, aktinomiceta i gljiva, dok je samo broj Azotobacter sp. bio je veći u zemljištu pod pšenicom u konvencionalnoj poljoprivredi. Najveća dehidrogenazna aktivnost utvrđena je u zemljištu pod rotkvicama, dok je u zemljištu ispod jabuke bila najniža.

Ključne reči: zemljište, mikroorganizmi, rizosfera, organska produkcija

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