



RESEARCH ARTICLE

Effect of various soil cultivation methods on some microbial soil properties

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Received: 10/01/2020

Accepted (Online first): 24/01/2020

Vol./Issue/Year: 1(1), 2020

Competing interests: Author(s) stated no compete of interest.

Edited by Lowy A. Daniel and Bence Mátyás

Grant information: The author(s) declared that no grants were involved in supporting this work.

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How to cite: Sándor, Zs. et al. "Effect of various soil cultivation methods on some microbial soil properties" *DRC Sustainable Future* 2020, 1(1): 14-20, DOI: 10.37281/DRCSF/1.1.3

ABSTRACT:

Cultivating the top 0-25 cm soil layer by ploughing cultivating method requires considerable energy, labor, and additional costs. Also, the larger soil surface caused by cultivation, the moisture content of soil can be lost easier. Therefore, in recent years soil loosening cultivation has become gained popularity, particularly to protect the moisture content of soil and reduce the risk of desertification. At the Experimental Station of Debrecen University, known as Látókép (a name, which corresponds approximately to visual image), two cultivation methods have been applied for research: (i) conventional ploughing and (ii) strip and streaked loosening cultivation methods (and variation of this method applying satellite determination of position, RTK system). In this paper, total number of bacteria, soil respiration, biomass carbon and nitrogen, net nitrification, and dehydrogenase activity were measured under irrigated and non-irrigated conditions. The aim of the research was to evaluate the effects of the various cultivation methods. Soil samples were collected in spring (May) and autumn (September). Microbiological effects on the soil resulted from various cultivation methods were compared. Results demonstrate that the loosening cultivation method (strip tillage with loosening) exerts a more favorable effect on the parameters of soil biological activity than the conventional ploughing system. The most significant effect of loosening cultivation system was experienced in the increase of microbial biomass carbon (MBC) and nitrogen (MBN) by over 80%, along with an intensified dehydrogenase activity. Loosening system yielded positive effects on the other examined biological parameters, except for the total bacteria number and soil respiration.

Keywords: soil cultivation methods, number of soil bacteria and fungi, soil respiration, microbial biomass carbon (MBC), and microbial biomass nitrogen (MBN).

1. Introduction

Our most important natural resource is the soil representing the basic medium for crop production;

therefore, its protection is vital. In Hungary, almost 50% of the land is cultivated for agricultural production. As part of agro-ecosystems, agricultural land is a special

type of natural resource, as it satisfies human needs and can be renewed with reasonable cultivation (Csete and Láng, 2004). Food production is realized in the agro-ecosystems, which depend on soil cultivation methods. Over agricultural processes large amount of carbon dioxide emission or absorption occurs, which affect the oxygen production, as well. Soil is a spot of biodiversity, which sensitively responds to the environmental changes (Fekete et al. 2012).

In recent years, knowledge of microbiological processes in the soil has expanded. Scientists' main goal is to understand the role of microbial community in the soil and in the nutrient cycles (Cho and Tiedje 2000; Fekete et al. 2017). Different cultivation technologies exert significant influence on the microbiological activity in soils (Kátai, 2006; Lajtha et al., 2018). Frey and co-workers examined the effects of no-tillage and conventional tillage on the bacterial and fungal abundance, on biomass, total C- and N, organic carbon and nitrogen and some other soil properties (Frey et al., 1998). These authors performed 6 long-term tillage comparison experiments. Bacterial abundance was not affected to a significant extent by tillage treatment.

Soil cultivation is an important element of arable land production, which consists of mechanical shaping of the soil with various means. The primary task of tillage is to improve the air and water management of soils to regulate their air and water content, to control the heat and nutrient management, and also the physical, chemical, and biological soil properties (Kocsis et al., 2018).

Two different methods are generally used in soil cultivation: with ploughing and without ploughing. In Hungary the ploughing still represents the most common cultivation method. For ecological reasons, there is a growing trend of cultivation without ploughing (untreated tillage systems) and combinations of the two types of basic cultivation (Birkás, 2006).

Intensive soil cultivation can activate soil life, can drastically change the natural communities of flora and fauna, and on the long shot it can favor soil degradation. With the quantitative reduction of organic matter, viability of soils varies, usually deteriorates, which can impact negatively soil fertility. Conservation of the living space of soil creatures can contribute to the improvement of soil condition, since they exert indirectly and directly a positive effect on several soil properties. In order to maintain biodiversity and multifunctionality of soils, appropriate cultivation methods should be applied. Essential for cultivation is to secure less disturbance of soil and use techniques that provide soil protection (Gyuricza and Mikó, 2015). Biological soil properties largely determine its renewal and affect important quality indicators of soil (Stott et al., 1999; Pabar et al., 2019). Microorganisms in soil play major

role in the conversion of organic matter and nutrient cycle. The biological system of the soil responds sensitively to environmental changes, especially to the loss of organic matter, which can be caused by soil cultivation. Generally, larger biological activity is measured, when soil cultivation is done without rotary tillage ploughing relative to cultivation with ploughing (Doran, 1980; Linn and Doran, 1984).

Experiments performed on Brazilian Cerrado Oxisols indicate that microbiological soil properties have improved upon cultivation by tillage. Over the years natural nutrient flow and soil structure have improved, which increased the sustainability of agriculture (Greena et al., 2007). Franchini et al. (2007) showed that microbial biomass carbon and nitrogen content of the soil was higher in areas without ploughing as compared to ploughing the soil.

Pseudomonas bacteria commonly are known as the growth of the plant growth stimulus (PGPR) in the rhizosphere. It has increased considerably during non-rotating farming (Agaras et al., 2014), which has an effect on the transformation of organic matter and can therefore contribute to continuous plant nutrition as required (Dudás et al., 2017). In non-rotated tillage systems, mushroom hyphas are longer and larger than the biomass of fungi and bacteria than in conventional cultivated soils (Beare et al., 1997 and Frey et al., 1999), which is due to the increased soil moisture content as a result of decreased soil disturbance.

Protected and more favourable soil moisture can enhance the activity of soil enzymes and through of it many other soil biology processes such as soil temperature and respiration (Kotroczó et al., 2014). In the case of non-cultivated direct sowing of seeds, the diversity of microscopic fungi and the amount of soil aggregates increased, suggesting that if there is no tillage, the number and activity of the microscopic fungi can be greater due to the preservation of the organic matter in the soil. It indirectly promotes the soil structure, the aggregate stability and generally improves the soil quality (Wang et al., 2010).

2. Methods

Experiments were conducted in 2013 at the Debrecen-Látókép Plant Cultivation Experimental Station of Debrecen University. Needed technical background was provided by KITE Ltd. In the area, calcareous chernozem soil can be found. According to the International Classification (WRB), the soil of the experiment was Calcic Endofluvic Chernozem (Endoskeletal).

In the selected study area, several cultivation systems have been used, with plant stock variations and increasing fertilizer levels. In this paper we examine the effects exerted on soil properties by (1) ploughing and

(2) conventional loosening cultivation, respectively. Soil loosening is the conventional loosening with disc and its variation with the RTK (Real-Time Kinematic) system (3 and 4). The essence of the RTK system is that for determining location one can use satellites, which can determine the site with 2 cm accuracy. For root crop cultures, plant rows are cultivated by tillage, and there is not cultivation between the rows. Precise positioning allows the rows to shift each year by 30 cm, so that the same area will be loosened every three years. Experiments include irrigated and non-irrigated plots. Tests were set randomly, with four repetitions.

Soil samples were taken twice in 2018, in the first week of May and second week of September from the parcels of maize culture, treated with medium dose fertilizer. Stock number was 50,000. From parcels cultivated by loosening with RTK cultivation, soil samples were taken both from the loosened rows, both between the rows; average soil samples were taken from four soil samples. As a result, we have done four kinds of treatment:

- Treatment 1: ploughing, or Conventional Tillage (CT), this being the control;
- Treatment 2: loosening by disc;
- Treatment 3: loosening by RTK method, soil samples were taken from the plant rows;
- Treatment 4: loosening RTK, soil samples were taken between the rows.

In the work disclosed here effects exerted on microbial soil properties by the two cultivation methods (ploughing and loosening) varied with Real-Time

Kinematic system. These effects were evaluated, and the effectiveness of the cultivation methods compared. Results are from the third year after the beginning of the experiments. All plots represent a set of four repetitions. Obtained results are listed in three tables. Statistical and mathematical methods have been used to evaluate the differences between different cultivation methods. Significant differences at 5% level were calculated as generally accepted in agricultural experiences.

Moisture content of soil was measured by drying the samples at 105 °C for 24 h. Quantity and quality of humus was measured by colorimetric method, according to Hargitai (1988). Total number of bacteria and microscopic fungi was examined by plate dilution methods, using bouillon agar and peptone-glucose agar (Pochon, et al., 1962). Organic carbon (OC) was assessed as recommended by Székely et al. (1960), organic nitrogen (ON) by Kjeldahl method (Filep 1988), nitrate determination (net nitrification) was measured after incubating fresh soil samples at 28 °C for two weeks (Felföldi, 1987). Quantity of aerobic cellulose decomposition and nitrifying bacteria was measured with the MPN (Most Probable Number) method, in liquid culture media (Trolldenier, 1996). CO₂-production was monitored after 10-day incubation (Hu et al., 1997.) Microbial biomass carbon (MBC) and MBN were measured by fumigation-extraction methods (Vance et al., 1987). Out of soil enzymes, dehydrogenase activity was calculated according to the method reported by Mersi and Schinner (1990).

Table 1. Number of bacteria, microscopic fungi, and two physiological groups of bacteria in a soil cultivation experiment, performed under irrigated and non-irrigated conditions (Látókép, Debrecen, April 1 and September 9, 2018.)

	Total number of bacteria *10 ⁵ *g soil ⁻¹		Number of microscopic fungi *10 ³ * g soil ⁻¹		Cellulose decomposing bacteria *10 ³ * g soil ⁻¹		Number of nitrifying bacteria *10 ³ *g soil ⁻¹		
	spring	fall	spring	fall	spring	fall	spring	fall	
Irrigated	Ploughed	6,13	8,35	21,13	29,33	0,28	2,59	0,60	0,92
	Loosened	3,83*	5,10*	22,19	56,36*	0,23	0,99	0,36	0,56
	RTK in rows	9,91*	5,20*	32,46*	31,97	0,57	3,19	0,84	1,56
	RTK between r.	6,46*	5,81*	27,53*	51,38*	0,50	1,30	0,47	1,78
	LSD5%	0,31	1,22	2,61	5,72				
Non-irrigated	Ploughed	4,15	8,71	24,29	31,44	0,36	0,60	0,70	0,76
	Loosened	2,07*	2,47*	23,83	62,13*	0,09	3,67	0,17	0,28
	RTK in rows	5,23*	2,19*	32,59*	33,43	0,19	0,86	0,72	1,12
	RTK between r.	5,0*	2,08*	28,66*	52,26*	0,35	1,62	0,62	1,75
	LSD5%	0,16	0,48	2,56	5,87				

3. Results and Discussion

Along the evaluation, results have been compared to traditional (rotational) ploughing; therefore, results from ploughing plots serve as the control. Humus content of soil varied between 2.6-2.8%. Significant differences were not found between the 2 kinds of treatment. Nevertheless, one should stress again that these are only results obtained in the second year after the start of experiment, so the time span is insufficient for humus content to change.

The number of bacteria and microscopic fungi along with two important physiological groups of bacteria – cellulose decomposing and nitrifying bacteria – are listed in Table 1. Bacterial population decreased significantly in the loosened plots (2nd treatment) in both irrigated and non-irrigated conditions, as compared to the ploughed plots. Regarding to the RTK treatments (3, 4), bacteria number in samples increased significantly over spring, but in fall it decreased relative to the control. Under non-irrigated conditions over spring, a smaller number of bacteria was measured in all treatments. Higher total bacteria number was extracted from plots of conventional tillage. This indicated that the quantity of bacteria was mostly affected by soil aeration. Higher amount of microscopic fungi was measured in loosened parcels, and in most cases the increase was significant. Generally, in fall a larger sum of fungi was

measured in both irrigated and non-irrigated conditions. Mycelia of microscopic fungi and the formation of spores was mainly determined by the rotation of soil, in agreement with results by Beare et al. (1997) and Frey et al. (1999).

For cellulose decomposition and nitrifying bacteria no significant differences were counted, given that the principle of determination is based on a statistical method (Most Probable Number). Over fall a greater number of cellulose decomposing bacteria was measured, as a result of greater quantities of plant residue. Foregoing statement is true for the number of nitrifying bacteria, particularly the loosened plots by RTK. Best results were obtained in soil found between plant rows, according to treatment 4. In most cases the amount of MBC increased significantly in the 3 loosened treatments as compared to the control ploughing. Results of microbial biomass nitrogen (MBN) showed a similar tendency to the one experienced in MBC, indicating that loosened soil conditions were favorable on microbial activity of soil across the higher amount of MBC and MBN. Soil respiration generally increased, but the increase was significant only for one loosened treatment, at spring sampling.

Table 2. Some parameters of soil biological activity in a soil cultivation experiment, under irrigated and non-irrigated conditions (Látókép, Debrecen, April 1 and September 9, 2018.)

	Total number of bacteria *10 ⁵ *g soil ⁻¹		Number of microscopic fungi *10 ³ * g soil ⁻¹		Cellulose decomposing bacteria *10 ³ * g soil ⁻¹		Number of nitrifying bacteria *10 ³ *g soil ⁻¹		Dehydrogenase enzyme INTF µg *g ⁻¹ 2h ⁻¹		
	spring	fall	spring	fall	spring	fall	spring	fall	spring	fall	
Irrigated	Ploughed	117,9	80,6	9,7	2,6	102,3	118,0	9,7	15,5	43,7	39,2
	Loosened	313,8*	189,0*	20,7*	18,8*	112,7	92,4	120,7*	29,1*	111,6*	45,3*
	RTK in rows	319,8*	245,3*	37,1*	14,2*	140,6*	103,9	54,3*	16,4	61,7*	37,8
	RTK between r.	145,7*	84,4	13,6	10,7*	112,9	97,5	14,7*	14,7	62,6*	33,7
	LSD5%	21,8	11,3	5,6	1,4	12,8	3,5	9,3	3,7	3,8	2,4
Non-irrigated	Ploughed	125,2	73,5	9,8	3,2	93,9	110,6	15,4	15,4	52,7	36,7
	Loosened	305,1*	123,5*	20,7*	19,3*	99,8	101,7	29,4*	21,2*	96,8*	37,6
	RTK in rows	284,8*	178,0*	12,3	18,5*	124,1*	114,8	43,2*	11,4	66,3*	40,5*
	RTK between r.	171,8*	64,3	18,2*	15,5*	110,9*	101,7	14,5	11,5	71,6*	36,9
	LSD5%	13,5	8,7	4,3	2,0	11,8	4,5	4,6	2,3	3,8	3,3

Table 3. Significant changes in microbial parameters for loosened parcels, as compared to conventional ploughing tillage.

Correlation		Irrigated	Non-irrigated	Total
number of total bacteria	+	1	2	3
	-	4	4	8
	0	1	0	1
number of microscopic fungi	+	4	4	8
	-	0	1	1
	0	2	1	3
cellulose decomposing bacteria	+	3	1	4
	-	3	4	7
	0	0	1	1
nitrifying bacteria	+	2	3	5
	-	4	2	6
	0	0	1	1
microbial biomass carbon	+	5	5	10
	-	0	1	1
	0	1	0	1
microbial biomass nitrogen	+	5	5	10
	-	0	0	0
	0	1	1	2
soil respiration	+	1	2	3
	-	3	2	5
	0	2	2	4
net nitrification	+	4	3	7
	-	0	2	2
	0	2	1	3
dehydrogenase activity	+	4	4	8
	-	2	0	2
	0	0	2	2
Total	+	29	29	58
	-	16	16	32
	0	9	9	18

+: positive effect relative to conventional tillage; -: negative effect compared to conventional tillage; 0: there were no significant differences

By evaluating net nitrification of soil samples in a 2-week incubation, one can conclude that there is a significant increase in nitrate content of the loosened plots as compared to the conventional ploughing. Higher nitrate production was measured in soil samples over spring, both in irrigated and not irrigated treatments. Activity of dehydrogenase enzyme is an indicator of the microbial redox system and the oxidative activity of the soil. Since dehydrogenase enzymes are active only within the living cell, the activity shows the activity of living microbial biomass. Dehydrogenase is essential for the metabolic reactions of cells, thus reflecting the state of metabolic activity. Generally, the smallest dehydrogenase activity was measured in plots of conventional ploughing, except for one treatment in fall. Activity increased in loosened plots, especially in spring, when significant growth was achieved in all treatments. In fall, dehydrogenase activity was stable in both irrigated and non-irrigated parcels.

According to the summary in Table 3, in the third year of the experiment over 50% of the measured microbial parameters increased significantly in the loosened plots relative to ploughing (CT); only one third of the parameters decreased significantly. Particularly the amount of microscopic fungi, the MBC and MBN content and dehydrogenase activity responded positively and changed significantly in most cases on the plots of the loosened cultivation tillage. Regarding the number of total bacteria and cellulose decomposing bacteria, significant decrease was recorded in the loosened plots, and higher numbers were measured in the ploughed plots. On the bases of yearly average results, in most cases higher activity was observed in the irrigated plots as compared to non-irrigated plots.

Conclusions

Based on measuring nine important soil microbial parameters, significantly higher biological activity was observed in plots of soil samples from loosened cultivation, and its combination with RTK-system, relative to conventional ploughing tillage. This applies for the third year after starting the experiment. Positive effects of loosening tillage were most prevalent in the amount of microscopic fungi, the increase of microbial biomass carbon and nitrogen content; altogether they accounted for nearly 75% of increase. In addition, dehydrogenase activity rose significantly. In plots of ploughing tillage, only the total number of bacteria and soil respiration was higher, indicating the positive activating effect of this cultivation method.

Generally, microbiological activity increased owing to the loosening conservation tillage, which enhanced the microbial biomass via larger production of organic matter. In general, it can be stated that cultivation

methods exert direct and indirect effects on the measured microbial soil parameters. Further research is needed at controlled experimental sites to gather more information about tillage-induced changes of organisms living in soil and their biological activity. These properties affect indirectly soil structure, aggregate stability, and overall soil quality.

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