

THE EFFECT OF DIFFERENT TILLAGE PRACTICES ON THE SOIL BACTERIA MICROFLORA IN WINTER WHEAT CULTIVATION

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Abstract

Soil microorganisms play an important role in the sustainability of soil vitality and productivity. In agricultural production, nutrients needed by plants are converted into useful form for plants by microorganisms. Intensive agricultural activities on agricultural soils are a serious threat to soil microorganisms and threaten the sustainability of agriculture in the long run. Soil treatment practices are known to be effective on soil microbial flora. This study was carried out in vertisol soil in Diyarbakir to investigate the effectiveness of conventional tillage, reduced tillage and direct seeding practices on soil microbial flora in the 2016-2017 winter wheat sowing period. Soil samples were taken from the rhizosphere of the wheat plant during the sowing time and flag leaf phenological period. Dilution technique was applied to the samples to determine live bacteria quantities. As a result of the study, it was determined that different tillage practices have a significant effect on soil bacterial count. It was determined that direct seeding (NT) application was significantly increased the number of soil bacteria compared to conventional tillage (CT) and reduced tillage (RT) treatments ($p<0.001$).

Key words: soil bacteria, wheat, soil tillage, no-till.

INTRODUCTION

Soil microorganisms play an important role in the continuation of soil fertility and productivity. Soil bacteria microflora is responsible for many processes that are necessary for the agricultural productivity of the soil. These processes include the recycling of plant nutrients, the protection of soil structure, the degradation of agricultural chemicals and pollutants, and the control of plant pathogenic microorganisms (Parkinson and Coleman, 1981; Lupwayi et al., 1998). Bacterial microorganisms in the soil are usually colonized on the soil surface, near the surface and on organic tissues (Chaudhry et al., 2015). The most important factors affecting both the intensity and composition of soil microbial flora are; environmental and soil temperature, amount of organic matter, inorganic nutrients, pH, climatic conditions and tillage activities. The tillage activities included in these factors have a direct impact on all other factors, since they affect the aeration of soil, the distribution of organic matter in the top profile of soil, and moisture capacity. For this reason, the chosen

tillage method affects the type and number of soil microorganisms. The widespread use of intensive and unconscious agricultural practices that involve too much intervention in the soil is among the greatest threats to soil ecosystem and biodiversity.

It is clear that intensive farming practices such as soil cultivation, monoculture production and chemical fertilizer application are detrimental effects on the formation of microbial flora in a wide range of soil bacteria (Gosling et al., 2006; Nyberg, 2006).

Wheat crop in the world and Turkey in terms of creating the raw materials for essential nutrients, is of a special importance compared to other agricultural products. GAP region with an area of 75 358 km² which is located in the Southeastern Anatolia Region of Turkey is corresponding to 9.7% of the total area of the country (Sessiz et al., 2006).

With the increase in the amount of land opened to irrigation in the context of GAP, intensive agricultural systems are applied in areas where, the soil are tilled in conventional methods. In this region where intensive agricultural systems are applied, at least two different crops are

grown annually in plant rotation. In this rotation system, winter wheat + corn or winter wheat + lentil farming is widely used.

Approximately 1.2 million hectares of wheat are sown in the Southeastern Anatolia Region according to the data of year 2016, and it is the third most important production area after Central and Western Anatolia (TÜİK, 2017a).

This planting area corresponds to 15.79% of the total wheat cultivation area of Turkey (TÜİK, 2017b).

Conventional tillage methods are widely applied in the region, especially in wheat cropping after corn harvesting.

The field with corn stalk remaining from the previous product harvest on the surface is ploughed with a mouldboard plough and then tilled with secondary tillage implements. Although landfills have been abandoned in recent years due to increased fuel costs and accelerated loss of moisture from the soil, conventional tillage methods throughout the region are still widely preferred.

In conventional systems, intensive and deep tilling of the soil results in numerous adverse effects on the physical, chemical and biological properties of the soil, and significant improvements in both environmental and soil quality parameters are obtained in conservative soil treatments and direct seeding systems.

In this point, it is important to determine the level of soil cultivation and planting methods on the number of soil bacteria in terms of high yield and healthy plant growth.

Understanding the changes on the bacterial microflora and linking different tillage methods helps to ensure precisely regulated sustainable agricultural systems (Treonis et al., 2010; Wall et al., 2012; Zhang et al., 2015).

In this study, it was aimed to determine the effect of different tillage and sowing methods on the number of soil bacterial microorganisms, which is one of the most important of soil microorganisms during sowing and flag leaf phenology in winter wheat growing.

MATERIALS AND METHODS

The study was carried out within the period of 2016-2017 wheat cultivation season at Dicle University Research Farm in Diyarbakır province located in Southerneast of Turkey ($37^{\circ}53'22''$ latitude N, $40^{\circ}16'38''$ longitude E, 670 m above sea level).

The study carried out on trial plots of 10 x 14 m, was planned in completely randomized parcel design with three replications.

The soil structures of the study area was given in Table 1.

Table 1. The soil structures of the study area

Depth (cm)				pH	Salinity	Organik Material	CaCO ₃	P ₂ O ₅	K ₂ O	Fe	Zn	Mn	Cu
	Sand (%)	Silt (%)	Clay (%)	Texture	(1: 2.5) (mmhos cm ⁻¹)	%	%	kg da ⁻¹	mg kg ⁻¹				
0-30	4.8	39.1	56.1	C	7.3	0.1	1.0	11.8	3.9	184.9	5.9	0.5	9.6
													1.4

As is seen in Table 2, three different tillage systems were applied in the study for wheat cultivation: conventional tillage (CT), reduced tillage (RT) and direct seeding (NT). The working width and the working depth of the

equipments are in Table 3. Mouldboard plough with a 400 mm working depth is the main equipment of conventional tillage and is frequently used after especially after corn harvest.

Table 2. The systems and the treatments

Systems	Treatments
Conventional Tillage (CT)	<ul style="list-style-type: none"> • Stubble chopper • Mouldboard plough • Disc harrow (2 times) • Scraper • Seeding
Reduced Tillage (RT)	<ul style="list-style-type: none"> • Cultivator • Disc harrow (2 times) • Scraper • Seeding
Direct Seeding (NT)	<ul style="list-style-type: none"> • Stubble cutting • Seeding

Table 3. The sizes of the equipments used in the study

Soil tillage equipment	Working width (mm)	Working depth (mm)
Mouldboard plough	1200	400
Cultivator	1700	350
Discharrow	2100	210
Scrapper	2000	50
Drill (24 row)	4800	5

Commonly used bread wheat variety in the region was used as wheat seed. Irrigation of the wheat was carried out by sprinkler irrigation system. For the fertilizer requirement of wheat plants, $15\text{-}18 \text{ kg N da}^{-1}$, $8 \text{ kg P}_2\text{O}_5 \text{ da}^{-1}$ and $15\text{-}20 \text{ kg K}_2\text{O da}^{-1}$ were given. No pesticides were used during the growing season. In the parcels that contains excessive amount of weeds, only the weeds were removed by hand and removed from the parcel.

Soil samples were obtained from two different periods; during the sowing and in the flag leaf phenological period of winter wheat. According to the simple random sampling method in each parcel, 27 soil samples were taken for each period in three replicates.

Soil samples were collected from 5-20 cm depth of points determined according to simple random sampling method with the help of a shovel, and is placed in a polyethylene bag (Bora and Karaca, 1970; Saygili et al., 2006). The obtained soil samples were dried in room conditions and then passed through a 2 mm sieve to prepare for dilution technique. The reason for the selection of the dilution technique in the study is that it is only possible

to count live and active bacteria in the dilution technique. Before studying, a preliminary study performed to determine the amount of dilution suitable for determining the number of soil microbial populations in the dilution technique; 1/100000 and 1/1000000. In addition to Nutrient Agar (NA) medium used as a general medium for bacteria (Johnson and Curl, 1972), Potato Dextrose Agar (PDA) medium was used for each sample in order to understand the effect of the medium in the study.

Before the analysis, Nutrient Agar (NA) and Potato Dextrose Agar (PDA) commercial formulations were prepared and autoclaved at 121°C for 20 minutes and sterilized under 1 atm. pressure. Later, the medium were poured into Petri dishes wrapped in parafilm and stored at +4°C in the refrigerator until the analysis was carried out. Soil samples were dried and passed through a 2 mm sieve and soil particles under the sieve were used in the study. The soil sample obtained by sieving in this way was pounded into a sterile ceramic mortar and came to henna consistency. After weighing 10 g on a precision scale, was put in a 250 ml volume of sterile Erlenmeyer. The dilution rate here is

1/10. 1 ml of the soil-water suspension in the prepared Erlenmeyer is transferred to a test tube containing 9 ml sterile water and mixed. 1ml of suspension in this test tube transferred to a test tube containing 9 ml sterile water and mixed again. This procedure was repeated in the same manner for 5 test tubes containing 9 ml of sterile water. The starting dilutions of the last two dilutions will be diluted to 1/100000 and 1/1000000.

100 μ l (microliter) of the last two dilutions (1/100000 and 1/1000000) were pre-prepared with a micropipette using a sterile tip for each use, transferred to sterile Petri dishes containing rested NA and PDA medium, and spread through the sterile glass stick.

Thus, for every soil sample; A total of 4 Petri dishes were inoculated to a Petri dish containing 1/100000 dilution 1 NA and 1 PDA media and to a Petri dish containing 1/10000 dilution 1 NA and 1 PDA media. The inoculated Petri dishes were wrapped with parafilm, incubated at $24 \pm 1^\circ\text{C}$, and counts of bacterial colonies were recorded every day for 10 days (Çınar and Biçici, 1991; Saygılı et al., 2006). All laboratory studies were carried out in a sterile sowing cabinet. In this respect, it is aimed to prevent contamination from air or other sources.

The data obtained on the basis of ten day observations were recorded. The logarithmic transformation was performed to the obtained data because of a positive correlation between the variance of the groups and averages and statistical analysis was then performed by the SPSS program. The results were evaluated according to the statistical analysis of raw data.

RESULTS AND DISCUSSIONS

According to the results of analysis, the effects of soil tillage method, sampling time, medium and dilution on the number of bacteria were found statistically significant.

The treatment of CT is mostly used for seedbed preparation in wheat cultivation in the region. It is known that CT method has negative effects on soil and environmental protection. In this study, negative effects of the conventional soil tillage treatment on the number of soil bacteria was determined as predicted. On the other hand NT was significantly increased the number of

soil bacteria compared to CT and RT treatments (Figure 1). Sirazuddin et al. (2016), determined that NT recorded maximum population of bacteria followed by RT and CT. Similar results showed that the abundance of soil bacteria founded greater in NT and RT than CT, even in different climates (Helgason et al., 2010; Kuntz et al., 2013; Shixiu et al., 2016).

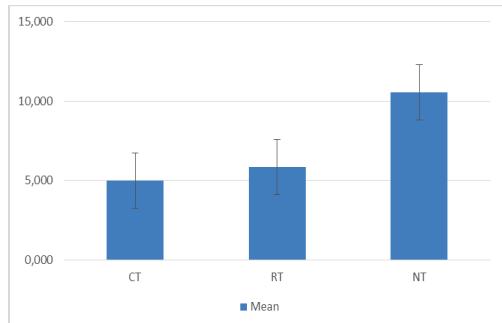


Figure 1. The means of bacteria after tillage practises
 $F = 9.509$, $df = 2,192$, $P < 0.001$

In the conventional tillage method, there was a significant decrease in the number of bacteria from the time of sowing until the time of flag leaf (Figure 2). Among the methods, statistically different and high values were determined for the number of bacteria in direct seeding method.

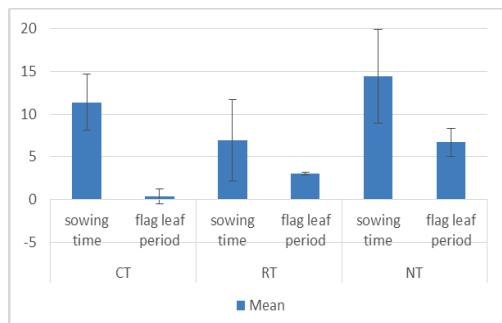


Figure 2. Effects of sampling time on the number of bacteria $F = 6.288$, $df = 2,192$, $P < 0.005$

The influence of tillage on microbial diversity was more prominent at the flag leaf period than at planting time. When examined according to the methods, it is seen that the difference between the numbers of bacteria in the flag leaf period is fully opened between NT and CT.

As seen in Figure 3, the NA medium is directly effect on bacteria formation. As expected, the development of bacteria has become more in NA because it is the suitable medium for bacteria according to the PDA.

The number of bacteria in reduced tillage took the third place in the PDA medium, although it was the second in terms of bacterial count on the NA medium. The most serious decline in bacterial quantities in NA and PDA medium occurred in the samples taken from direct seeding parcels.

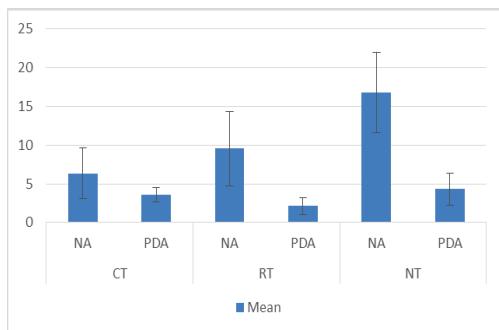


Figure 3. Effects of medium on the number of bacteria
 $F = 4.387$, $df = 2,192$, $P < 0.05$

CONCLUSIONS

In this study, as expected, the number of total bacteria is positively influenced by NT and RT. Also NT and RT systems prevent the number of bacteria from significantly decreasing from the time of sowing until the time of flag leaf. Recorded significant decrease in the number of bacteria from the time of sowing until the time of flag leaf in conventional tillage system may result in reductions in diversity of soil organisms due to loss of the soil moisture, increased soil compaction, reduced pore volume and degradation of the rhizosphere layer by overtopping the soil.

Especially using of mouldboard plough in every cultivation season for various reasons, causes this negative factors.

Conservation tillage systems including direct seeding has been proposed to farmers, have a major role in abundance and richness of soil bacteria.

Considering the results of this study, adoption of the conservation tillage systems in wheat farming, which is one of the major sources of

income for farmers in the region, supports bacterial growth and maintains soil sustainability.

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