

TILLAGE EFFECT ON SOIL MICROBIOTA IN WINTER WHEAT (*TRITICUM AESTIVUM* L.) AT EZARENI RESEARCH STATION

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Abstract

The study was carried out at UASMV Iasi Research Station – Ezareni Farm, on a cambic mesocalcaric regraded chernozem. To highlight the influence of tillage (no-tillage, conventional tillage) on soil microbiota in winter wheat (*Triticum aestivum* L.), soil samples were taken in 3 growth stages (tillering, stem elongation and flowering) in March-June 2017, from 7-10 cm depth soil layer. Subsequently, within 24 hours from sampling, the samples were analyzed by dilution method and seeded on four different culture media: PDA, PDA + Streptomycin, PDA + Rose Bengal and ASHBY. The number of bacteria was determined after 24 hours, and filamentous fungi microscopic identification was done 5 days after medium seeding.

Results show major differences regarding soil microbiota for both tillage treatments and in all three crop growth stages. At tillering, microbial activity was dominated by *Penicillium* and *Trichoderma* fungi found in different proportions; *Penicillium* fungi also dominated at stem elongation in no-tillage, while in conventional tillage *Ulocladium* fungi was also present into the seeded medium.

At flowering, *Penicillium* fungi had almost the same proportion both in no-tillage and conventional tillage. *Aspergillus* fungi registered also almost the same proportion in both tillage treatments but were dominant over *Penicillium* genus.

The number of genera was almost the same in conventional and no-tillage, in all three crop growth stages, but their proportions had a significant variation.

In no-till, the main genera of micromycetes were found in smaller proportions in comparison to conventional tillage. Microbiological analysis shows that soil microbiota is directly affected by tillage and crop growth stage in winter wheat.

Key words: no-tillage, soil microbiota, nutritive media

The soil represents an excellent environment for the development of microorganisms, a fact proven both by their number and by their great diversity (Zarnea G., 1994). Due to its heterogeneity, this natural environment carries populations of microorganisms with very different biological and biochemical particularities, between which are established extremely complex relations.

Soil microflora is a sensitive indicator of soil functionality when investigating the impacts of management practices, such as fertilization (García-Ruiz *et al*, 2008) and land use systems (Alvear *et al*, 2005).

Biomass of all microorganisms living in soil play an important role in the functioning of entire soil ecosystems because their enormous biochemical activity. Soil microflora is involved in biochemical transformations of fertilizers, synthesis of biologically active substances and nitrogen fixation from the air. Because of active role of soil micro-population, the diminution in microbial activity is always alarming (Barabasz *et al*, 2002).

Carter M.R., (1991, quoted by Kabiri *et al*, 2016) mentions that the response of soil microorganisms and biochemical properties to tillage practices has been measured by estimating the size and activity of the microbial community and the activities of various soil enzymes.

According no-tillage, by leaving all residues on the soil surface, we obtain a series of biological, chemical and physical characteristics, which are opposite to the ones of a ploughed soil. A comparison between the no-tillage and conventional tillage will include differences regarding the microbial environment, the number and the activity of the microorganisms from the soil (Phillips R.E. and Phillips S.H., 1983).

The aim of the present study was to highlight the effect of tillage in winter wheat soil microbiota.

MATERIAL AND METHOD

In order to highlight soil microbiological activity in winter wheat, soil samples have been

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taken for analysis during three vegetation stages in March – June 2017, from two tillage treatments: no-tillage and conventional tillage.

For sampling, there was made an initial delimitation of the area from which were chosen 3 points in diagonal for each tillage treatment. The soil samples were taken from a depth interval of 7 – 10 cm from around the roots. After being taken, the soil samples were transferred in sterile paper bags and then transported to the laboratory in order to be analyzed. The soil samples were ground up with a mortar and pestle, processed through the dilution method and sown in four different media: PDA (Potato-dextrose-agar), PDA + Streptomycin, PDA + Rose Bengal and Ashby, for the identification of Gram-positive bacteria, Gram-negative bacteria, molecular nitrogen fixation bacteria and of actinomycetes.

After sowing, the plates were incubated at 28°C, for 24 hours, in order to determine the number of bacteria, and the microscopic identification of filamentous fungi was done after 5 days. The count

was done with the naked eye, using a marker, and in the case of big densities, the Wolfhügel plate was used. In order to accurately determine the fungi species, microscopic preparates were analyzed with the optical microscope (Larpernt J.P., Larpernt-Gourgand M., 1990).

RESULTS AND DISCUSSIONS

The results have shown major differences regarding the activity of microorganisms on each of the three vegetation stages.

The analysis of the values demonstrates that the numeric distribution of microorganisms is directly influenced both by tillage, and by the vegetation stage of winter wheat.

In *Table 1* are presented the results of the investigations regarding the bacterial activity from the soil samples taken from both tillage treatments.

Table 1

Dynamics of soil microbiota in winter wheat crop in the agricultural year 2016-2017

Growth stages	NO-TILL				
	Total number of bacteria	G ⁺ Bacteria	G ⁻ Bacteria	Nitrogen fixation bacteria UFC/g	
	UFC/g	UFC/g	UFC/g	Anaerobe	Aerobe
Tillering	239 x 10 ³	60 x 10 ³	179 x 10 ³	2.4 x 10 ³	1.6 x 10 ³
Stem elongation	348 x 10 ³	15 x 10 ³	133 x 10 ³	4.2 x 10 ³	6.2 x 10 ³
Flowering	282 x 10 ³	21 x 10 ³	261 x 10 ³	7.8 x 10 ³	15 x 10 ³
	CONVENTIONAL				
Tillering	411 x 10 ³	11 x 10 ³	400 x 10 ³	2.2 x 10 ³	2.4 x 10 ³
Stem elongation	504 x 10 ³	41 x 10 ³	463 x 10 ³	2.2 x 10 ³	7.8 x 10 ³
Flowering	347 x 10 ³	23 x 10 ³	324 x 10 ³	8.8 x 10 ³	16.4 x 10 ³

Analyzing the results obtained regarding the abundance of bacteria, it was noticed that the number of Gram-positive bacteria is higher in no-tillage at "tillering" (60 x 10³ UFC/g), and in the next growth stages, the Gram-positive bacteria were predominant in conventional tillage, a situation that we get to observe also in the last vegetation stage. These results demonstrate the fact that the bacterial activity was more intense at first in the stage of "tillering" in the case of the no-tillage system, after which the situation changed in the case of the other two growth stages, the number of Gram-positive bacteria being higher in conventional tillage (41 x 10³ and 23 x 10³ UFC/g).

Bacteria participate in the mineralization processes, which are essential for plant nutrition and for insuring soil fertility. They are the only organisms that are capable to enrich soil nitrogen-, by fixating the molecular nitrogen from the atmosphere. Through extracellular polysaccharides,

bacteria participate to the aggregation of soil particles and humus formation (Ştefan M., 2006).

The values obtained have shown the presence of bacteria that fixate molecular nitrogen, i.e. *Clostridium pasteurianum* and *Azotobacter chroococcum*, in much greater amount A-T "flowering", in both tillage treatments.

The results of microscopic identification of micromycetes have shown that the species of the *Penicillium* genus were the most widespread in all three vegetation stages.

The analysis of the soil samples taken at "tillering" of the wheat have indicated the presence of eight genera of fungi in no-tillage system (*fig. 1*) and seven genera of fungi in conventional tillage system (*fig. 2*). In the case of both tillage treatments, the species of the *Penicillium*, *Trichoderma* and *Aspergillus* genera were predominant.

Clear differentiations have been observed in the case of species from *Penicillium* genus, where

the lowest value was recorded in the conventional tillage, in a percentage of 64%, while in the no-tillage system, these bacteria were found in a percentage of 77%.

Regarding the presence of the *Trichoderma* genus, in conventional tillage there have been recorded values of 19%, in comparison with no-tillage, where these species were found in a lower percentage (8%).

The species of the *Aspergillus* genus were found in the same percentage (6%) in the case of both tillage systems.

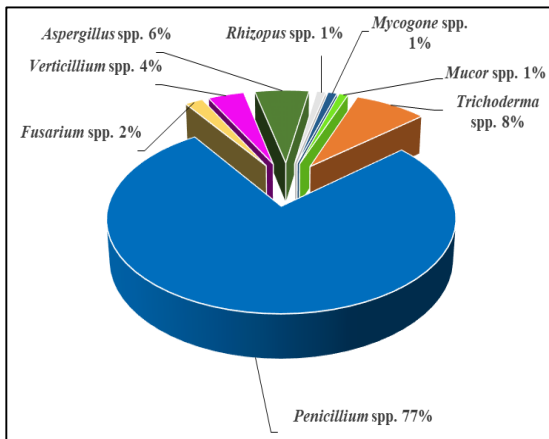


Figure 1 – Frequency of isolated fungi „tillering” stage, no-tillage

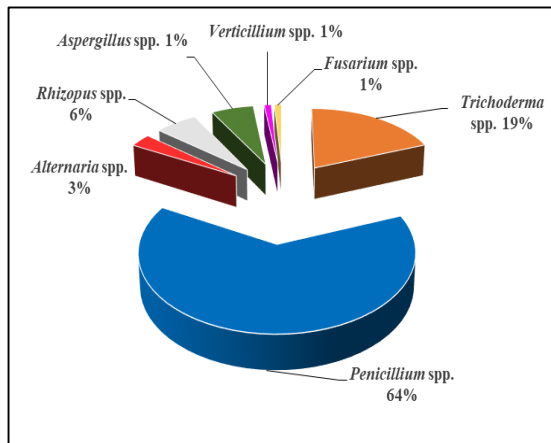


Figure 2 – Frequency of isolated fungi „tillering” stage, conventional tillage

From the analysis of the samples taken in the “stem elongation” stage, a smaller presence of the main fungi genera was observed, compared with the “tillering” stage, more reduced in no-tillage system (fig. 3), compared to conventional tillage (fig. 4).

Judging about the main fungi genera identified, it was noted that the species of the *Penicillium* genus were more numerous in the case of the no-tillage (74%) compared to conventional tillage variant (48%).

The species of the *Trichoderma* genus were represented in a percentage of 23% in conventional tillage, while in no-tillage, they have been represented in a lower percentage (5%).

In addition to the other stages analyzed, in conventional tillage, there were found species that pertain to the *Ulocladium* genus.

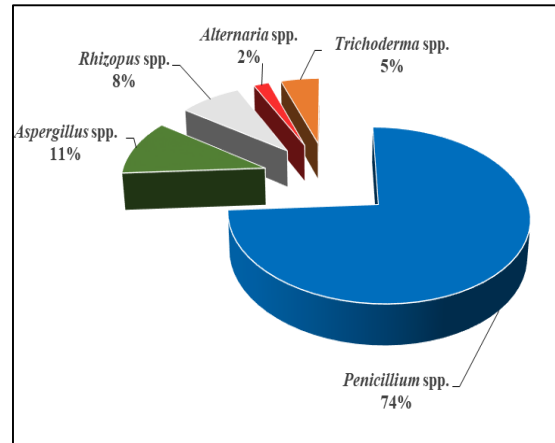


Figure 3 – Frequency of isolated fungi „stem elongation” stage, no-tillage

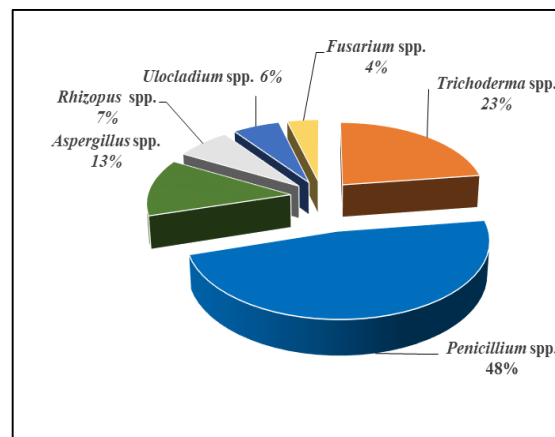


Figure 4 – Frequency of isolated fungi „stem elongation” stage, conventional-tillage

The analysis of the results from the “flowering” stage of the wheat shows the fact that in both no-tillage (fig. 5) and conventional tillage (fig. 6), there were found seven genera of fungi. Just as in the case of the previous stages, the species of the *Penicillium* genus were predominant, representing 77% in no-tillage and 62% in conventional tillage.

The species of the *Aspergillus* genus were found in similar percentages in the case of both no-tillage systems, namely 17% in no-tillage and 18% conventional tillage.

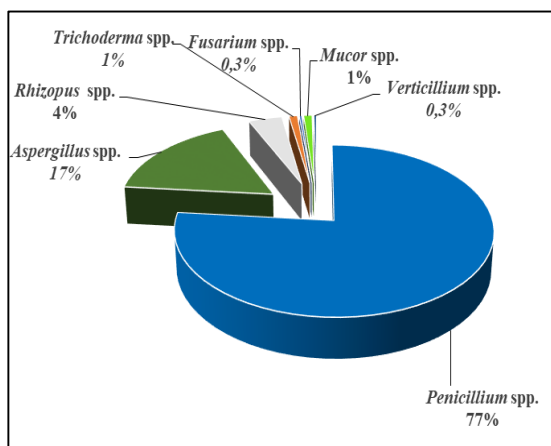


Figure 5 – Frequency of isolated fungi „flowering” stage, no-tillage

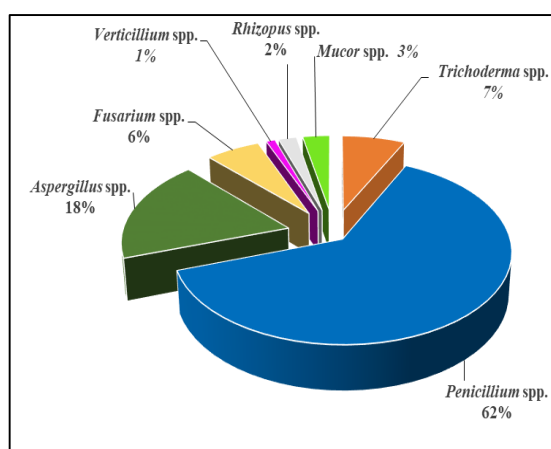


Figure 6 – Frequency of isolated fungi „flowering” stage, conventional tillage

The number of fungi genera have varied according to the growth stages, in no-tillage having found eight genera in the “tillering” stage, that were reduced to five genera in the “stem elongation” stage, which then grew to a number of seven genera in the “flowering” stage.

In conventional tillage there were found seven genera of fungi in the “tillering” and “flowering” stages, being reduced to only six genera in the “stem elongation” stage.

The fungi actively participate to the formation of the soil structure through the effect of mechanical immobilization and aggregation of soil particles, thus ensuring their stability.

CONCLUSIONS

In no-tillage and conventional tillage variants, the dominant genus was represented by *Penicillium*, but it was found in higher proportions in no-tillage.

The presence of the species from the *Aspergillus* genus among the main fungi genera is a good indicator because it is well known that these species have a role in the humus formation, creating aromatic substances that are similar to the lignin's that are introduced in the soil reserve.

Regarding the bacterial activity, it is considered that Gram-negative bacteria are predominant in the soil, and the analyzed soil samples have confirmed this situation.

In all three growth stages, in the case of conventional tillage system, the number of Gram-negative bacteria was visibly higher than the number of the Gram-negative bacteria found in no-tillage.

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