*Почвознание агрохимия и екология, 57, 3/2023*
*Bulgarian Journal of Soil Science Agrochemisty and Ecology, 57, 3/2023*

*DOI: https://doi.org/10.61308/JPYA6605*

# Impact of advanced soil conservation technologies for growing maize on sloping terrains on soil microbiota

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**Citation**: Ivanova, I., & Kuncheva, G. (2023). Impact of advanced soil conservation technologies for growing maize on sloping terrains on soil microbiota. *Bulgarian Journal of Soil Science Agrochemisty and Ecology, 57*(3), 9-15.

**Abstract**

Water erosion leads to the loss of soil, nutrients, organic matter, disruption of soil structure, reduction of moisture capacity, loss of biodiversity, and is the most significant soil degradation process when growing agricultural crops on sloping terrain. Soil microorganisms are the basis of the functioning of ecosystems, the circulation of substances, soil-forming processes and the formation of the soil structure. Microbiological indicators, such as numbers, microbial biomass, enzyme activity, etc., are a sensitive indicator of the changes that have occurred in the soil, under the influence of anthropogenic or natural impacts. The present study is on the abundance of major groups of soil microorganisms, when applying different tillage systems in growing maize on sloping terrain. The study was carried out on Carbonate Black Earth, at the Experimental Station for Combating Soil Erosion, at IASSPP "N. Pushkarov", in the village of Trastenik, Ruse, Agricultural Academy - Sofia. A field experience with corn for grain, applying traditional and anti-erosion technologies (surface and vertical mulching) for growing corn, is presented. The obtained results show a positive influence of the applied soil protection technologies on the number of saprophytic bacteria, mold fungi, actinomycetes, nitrogen-fixing and cellulose-degrading microorganisms.

**Key words**: soil microorganisms, microbiological indicators, anti-erosion technologies, vertical mulching, surface mulching

## Introduction

Soil biota is the "biological engine of the earth, associated with driving and transforming physical, chemical, biological and ecological processes" (Ritz et al., 2004). Soil microorganisms are the basis of soil fertility, the formation of a sustainable structure, the transformation of organic matter, the cycle of biogenic elements, etc. (Fortuna, 2012; Malcheva, 2020; Petkova et al., 2015; Petkova et al., 2022; Perfanova & Lyubenova, 2017; Perfanova & Ivanova, 2019).

The distribution by type and activity of soil microorganisms: bacteria, actinomycetes and microscopic fungi, depends on the soil type, the content of organic matter, etc., and changes under the influence of abiotic and biotic factors, and depending on the way of land use (Blazevska, 2016; Blazevska et al., 2021; Perfanova et al., 2021).

Indicators, characterizing the structure and activity of soil biota, are the subject of research and an indicator of the ecological balance in the soil and its productivity (Kennedy & Papendick, 1993; Islam & Weil, 2000; Avidano et al., 2005; Usman et al., 2016).

The ecological plasticity and adaptability of microorganisms determines the possibility that indicators characterizing their number and activity can be used to assess soil quality, as well as indicators of adverse changes in the soil (Bending et al., 2004; Schloter et al., 2018; Petkova et al., 2022).

Soil degradation is the result of natural processes and human activity, as a result of which the structure, composition, structure deteriorate and fertility decreases. Soil erosion is the most significant degradation process (Dimitrov et al., 2014; Malinov, 1999; Malinov et al., 2014; Ruseva, 2006). The loss of soil, the reduction of soil organic matter, the deterioration of physical indicators and moisture retention as a result of water erosion, has a serious impact on natural and agrarian ecosystems, with economic and social consequences (De Vries et al., 2006; Panagos et al., 2020). In these ecosystems, the total biomass of flora and fauna, biological diversity and yields of cultivated crops decrease (Usman et al., 2016). Soil is a limited resource and this requires maintaining it in a state of sustainable system (Dinev et al., 2017). The area of arable land is decreasing as a result of the growing pressure of society, which forces scientific research to be aimed at compensating the loss of soil by improving its quality. A key role is played by the maintenance of favorable chemical, physical and biological properties of the soil (Dimitrov et al. , 2019; Ivanova, 2021; Kolchakov et al., 2019; Kuncheva, 2016; Malinov et al., 2007; Mitova et al., 2015), for which the application of soil protection technologies is necessary for growing agricultural crops.

The purpose of the present study is to determine the influence of traditionally applied and anti-erosion technologies for growing corn on sloping terrain, on the number of main groups of soil microorganisms.

## Materials and methods

The area on which the experience is displayed, is located on carbonate chernozem. The terrain is undulating with long gentle slopes, with a slope of 5° (8.7%). The territory has a southeast exposure. To realize the purpose of the research in the experimental field of IASSPP "N. Pushkarov", at the Experimental Station for Combating Soil Erosion land of the village of Trastenik, region Ruse, at the Agricultural Academy - Sofia, was performed in four variants in four repetitions, with one-factor. The factor is the soil protection technological operations, used in the proposed technologies (Shanin, 1977). Variants of the experiment are: $f_o$ - corn crop, grown according to traditional technology (with basic tillage turning the layer), applied on the slope of the slope - control; $f_1$ - maize, according to traditional technology (using basic tillage with inversion of the layer), applied across the slope; $f_2$ - maize, according to soil protection technology (using basic tillage with layer turning and surface mulching with manure), across the slope; $f_3$ - corn crop, with advanced soil protection technology for minimal and non-traditional processing (including basic tillage - loosening and the soil protection measures

vertical mulching with manure, cutting with furrowing with sowing and hoeing and furrowing with cutting and furrowing with necking), across the slope.

In the control option f, deep ploughing, pre-sowing treatments, sowing, vegetation treatments and harvesting are carried out along the slope of the slope, while in $f_1$ they are carried out in a transverse direction. Transversely on the slope of the slope, treatments are also carried out in variant $f_2$, and surface mulching with manure (450-500 kg/da) is also applied before the pre-sowing treatments. In variant $f_3$, the main tillage - deep plowing is replaced by the plowless technological operation of loosening (cultivation with a chisel cultivator), at a depth of 0.40 m. In this variant, with all the traditional treatments, across the slope, the anti-erosion methods of vertical mulching with manure at a depth of 0.40 m, cutting with furrow formation and furrow formation with cutting and furrow formation are carried out.

Traversing is carried out annually, twice at different stages of this advanced soil protection technology, with different technical means. It is applied at the same time as the sowing of the corn, across the slope in the formed interrows, at a depth of 0.25 m, with the distance between the slots being 1.4 m (across the interrow). The other anti-erosion method, furrowing with cutting and treading is carried out across the slope of the slope, and the technological operations of furrowing (hugging), and cutting with treading are carried out sequentially and simultaneously.

The microbiological analyzes of the soil samples were carried out in the laboratory for erosion studies of IASSPP "N. Pushkarov" in RU "An. Kanchev". During the research period, the number of soil microbiota was determined three times during the vegetation (before sowing, in maximum growth and after harvesting) of the crop, taking into account bacteria - spore-forming and non-spore-forming, nitrogen-fixing, cellulose-degrading microorganisms, actinomycetes, fungi, oligotrophic microflora on solid food media, MPA (mesopeptone agar) - for bacteria, CAA (starch ammonia agar) - for actinomycetes, Chapek's medium - for mold fungi, RPA (diluted soil agar) - for oligotrophic soil microflora, as well as nitrogen-fixing bacteria - Ashby's medium, cellulose-degrading bacteria on Hutchinson's nutrient medium. Samples were taken in the 0-20 cm layer, by averaging the first and third replicates of each trial variant three times per year (Grudeva et al., 2006). The obtained results of the conducted experiment were processed with software products Anova test and Statgraphics 2.1.

**Results and discussion**

The results, obtained for the amount of the main groups of soil microorganisms CFU (colony forming units) $10^6 g^{-1}$ absolutely dry soil are reflected in table 1.

In variant $f_3$ during the maximum growth phase, the amount of saprophytic bacteria was $2045.24 \times 10^6$ CFU/g, which was 2.88 times greater than the control variant $709.30 \times 10^6$ CFU/g. The combination of minimal tillage and the application of manure in the slits improves the development of the soil microflora. Bacteria show the greatest sensitivity to changes in soil conditions. The differences between the individual variants reach 4-5 times.

The results, obtained for the spore-forming microorganisms, indicate that their number is the highest in the $f_3$ variants before sowing, respectively, with a value of $84.28 \times 10^6$ CFU/g in maximum growth $115.65 \times 10^6$ CFU/g and after harvesting $60.82 \times 10^6$ CFU/g, compared to the control variant, where the values are $71.14 \times 10^6$ CFU/g, $17.99 \times 10^6$ CFU/g and $35.39 \times 10^6$ CFU/g. The introduction of organic matter has a positive effect on the soil microbiota.

Oligotrophs as microorganisms developing on mediums poor in organic matter before sowing were in the greatest amount $347.97 \times 10^6$ CFU/g, compared to variant $f_0$. The level of oligotrophs is high in the variants with the addition of manure.

In variant $f_3$, it was significantly higher $578.01 \times 10^6$ CFU/g and $484.46 \times 10^6$ CFU/g during maximum growth of maize and after its harvest.

Actinomycetes are actively involved in the final stages of decomposition organic matter and

**Table 1.** Amount of the main groups of soil microorganisms CFU (colony forming units) $10^6$/gr Absolutely dry soil

| Variant | Total number of saprophytic bacteria | Sporeforming | Oligo trophic | Actino mycetes | Mold fungi | Nitrogen fixatives | Cellulose degraders |
|---|---|---|---|---|---|---|---|
| ***2017*** | | | | | | | |
| Before sowing | | | | | | | |
| $f_0$ | 186.20 | 4.18 | 296.40 | 0.152 | 0.00140 | 2.20 | 1.26 |
| $f_1$ | 210.93 | 26.36 | 380.43 | 0.452 | 0.00090 | 2.15 | 0.90 |
| $f_2$ | 421.80 | 47.50 | 296.40 | 0.228 | 0.00210 | 3.72 | 1.29 |
| $f_3$ | 2990.00 | 6.90 | 195.50 | 1.840 | 0.00320 | 4.26 | 1.41 |
| Maximum growth | | | | | | | |
| $f_o$ | 627.20 | 29.12 | 0 | 1.083 | 0.00120* | 0.90 | 2.36 |
| $f_1$ | 417.60 | 46.40 | 0 | 1.102 | 0.00140** | 1.39 | 2.40 |
| $f_2$ | 620.00 | 88.00 | 0 | 1.100 | 0.00090 | 26.73* | 3.14 |
| $f_3$ | 1482.00 | 11.40 | 0 | 1.633 | 0.00190* | 2.85 | 2.36 |
| After harvesting | | | | | | | |
| $f_o$ | 6.00 | 13.81 | 3.73 | 0.280 | 0.00130 | 0.57 | 1.46 |
| $f_1$ | 162.40 | 23.97 | 42.53 | 0.440 | 0.00200 | 0.67 | 0.46 |
| $f_2$ | 275.00 | 11.10 | 40.33 | 1.160* | 0.00110 | 1.21 | 1.28 |
| $f_3$ | 121.60 | 12.16 | 7.60 | 1.140* | 0.00160 | 1.06 | 2.36 |
| ***2018*** | | | | | | | |
| Before sowing | | | | | | | |
| $f_0$ | 2702.80 | 175.90 | 0 | 0.46 | 0.0044 | 0.46 | 0.23 |
| $f_1$ | 2469.87 | 193.80 | 11.40 | 2.28 | 0.0027 | 1.14 | 0.23 |
| $f_2$ | 3765.80 | 175.93 | 0 | 0.93 | 0.0045 | 1.39 | 0.70 |
| $f_3$ | 3696.50 | 218.80 | 11.60 | 0.81 | 0.0045 | 2.67 | 0.93 |
| Maximum growth | | | | | | | |
| $f_o$ | 832.20 | 0 | 23.40 | 1.05 | 0.00012 | 2.73 | 0.23 |
| $f_1$ | 558.20 | 0 | 11.40 | 1.56 | 0 | 2.51 | 0.35 |
| $f_2$ | 1014.00 | 17.10 | 7.60 | 1.59 | 0 | 5.02 | 0.68 |
| $f_3$ | 1016.38 | 1.14 | 91.36 | 2.78 | 0.00015 | 1.83 | 1.03 |
| After harvesting | | | | | | | |
| $f_o$ | 408.27 | 89.32 | 582.67 | 0.35 | 0.00035 | 6.82 | 0.38 |
| $f_1$ | 436.60 | 36.97 | 545.20 | 0.63 | 0.00035 | 12.99 | 1.04 |
| $f_2$ | 490.67 | 69.38 | 582.13 | 1.93** | 0.00046 | 13.14 | 2.12* |
| $f_3$ | 538.87 | 125.87 | 1038.40* | 1.14* | 0.00063 | 16.56 | 1.18 |
| ***2019*** | | | | | | | |
| Before sowing | | | | | | | |
| $f_0$ | 149.50 | 33.35 | 747.50 | 0.345 | 0.0014 | 4.95 | 0.12 |
| $f_1$ | 440.70 | 8.29 | 252.37 | 0.790 | 0.0010 | 1.88 | 0.47 |
| $f_2$ | 393.30 | 12.98 | 401.20 | 1.92* | 0.0007 | 2.64 | 7.95** |
| $f_3$ | 472.00 | 27.14 | 413.00 | 1.180* | 0.0014 | 4.17 | 6.26* |
| Maximum growth | | | | | | | |
| $f_o$ | 668.50 | 24.85 | 476.00 | 2.52 | 0.0020 | 3.89 | 1.79 |
| $f_1$ | 583.00 | 59.36 | 273.00 | 4.06 | 0.0015 | 2.73 | 1.16 |
| $f_2$ | 773.50 | 117.60 | 293.27 | 3.71 | 0.0028 | 6.43 | 1.38 |
| $f_3$ | 3637.33* | 334.40* | 1642.67 | 13.35** | 0.0025 | 10.93 | 3.96 |
| After harvesting | | | | | | | |

*Table 1. Continue*

| | | | | | | | |
|---|---|---|---|---|---|---|---|
| $f_o$ | 16.83 | 3.03 | 0 | 0.34 | 0.0006 | 0.30 | 0.88 |
| $f_1$ | 108.80 | 4.08 | 183.60 | 0.41 | 0.0020 | 1.01 | 0.67 |
| $f_2$ | 170.00 | 4.00 | 360.00 | 0.97 | 0.0011 | 1.41 | 4.83** |
| $f_3$ | 249.13 | 44.44 | 407.37 | 0.98 | 0.0026*** | 3.10 | 2.62 |
| ***Average for the period*** | | | | | | | |
| Before sowing | | | | | | | |
| $f_o$ | 1012.83 | 71.14 | 347.97 | 0.32 | 0.00240 | 2.54 | 0.54 |
| $f_1$ | 1040.50 | 76.15 | 214.73 | 1.17 | 0.00153 | 1.72 | 0.53 |
| $f_2$ | 1526.97 | 78.80 | 232.53 | 1.03 | 0.00243 | 2.58 | 3.31 |
| $f_3$ | 2386.17 | 84.28 | 206.70 | 1.28 | 0.00303 | 3.70 | 2.87 |
| Maximum growth | | | | | | | |
| $f_o$ | 709.30 | 17.99 | 166.47 | 1.55 | 0.00111 | 2.51 | 1.46 |
| $f_1$ | 519.60 | 35.25 | 94.80 | 2.24 | 0.00097 | 2.21 | 1.30 |
| $f_2$ | 802.50 | 74.23 | 100.29 | 2.13 | 0.00123 | 12.73 | 1.73 |
| $f_3$ | 2045.20 | 115.65 | 578.01 | 5.92 | 0.00152 | 5.20 | 2.45 |
| After harvesting | | | | | | | |
| $f_o$ | 160.37 | 35.39 | 195.47 | 0.32 | 0.00075 | 2.56 | 0.91 |
| $f_1$ | 235.93 | 21.67 | 257.11 | 0.49 | 0.00145 | 4.89 | 0.72 |
| $f_2$ | 311.89 | 28.16 | 327.49 | 1.35 | 0.00089 | 5.25 | 2.74 |
| $f_3$ | 303.20 | 60.82 | 484.46 | 1.09 | 0.00161 | 6.91 | 2.05 |

HSD: For saprophyte.bact. before sowing P≤0.05-3024.32; P≤0.01 -4400.59; P≤0.001-6611.68; max. growth- P≤0.05-1336.17; P≤0.01-1944.21; P≤0.001-2921.08. after harvesting P≤0.05-364.422; P≤0.01-530.258; P≤0.001-796.687. sporulating - before sowing P≤0.05-188.175; P≤0.01-273.807; P≤0.001-411.381; max. growth- P≤0.05-277.653; P≤0.01-420.686; P≤0.001-676.15. after harvesting P≤0.05-79.914; P≤0.01-116.281; P≤0.001-174.406. oligotrophs - before sowing P≤0.05-463.376; P≤0.01- 702.083; P≤0.001-1128.43; max. growth- P≤0.05-1631.23; P≤0.01-2705.01; P≤0.001-5058.74. after harvesting P≤0.05-812.022; P≤0.01-1201.74; P≤0.001-1857.09.actinomycetes - before sowing P≤0.05-1.319; P≤0.01- 1.920; P≤0.001-2.885; max. growth- P≤0.05-6.446; P≤0.01-9.379; P≤0.001-14.092. after harvesting P≤0.05-0.500; P≤0.01-0.728; P≤0.001-1.094; mold fungi - before sowing P≤0.05-0.00299; P≤0.01-0.0043; P≤0.001-0.0065; max. growth- P≤0.05-0.00233; P≤0.01-0.0035; P≤0.001-0.0056. after harvesting P≤0.05-0.0014; P≤0.01-0.0020; P≤0.001-0.0030. nitrogen fixing - before sowing P≤0.05-2.587; P≤0.01-3.765; P≤0.001-5.657; max. growth- P≤0.05-12.461; P≤0.01-18.132; P≤0.001-27.242. after harvesting P≤0.05-12.645; P≤0.01-18.400; P≤0.001-27.645. cellulose-degrading - before sowing P≤0.05-4.746; P≤0.01-6.905; P≤0.001-10.375; max. growth- P≤0.05-2.313; P≤0.01-3.366; P≤0.001-5.058. after harvesting P≤0.05-1.976; P≤0.01-2.876; P≤0.001-4.321.
***- many well-proven differences; **- well-proven difference; *-proven difference

activate the processes of formation and accumulation of humus in the soil. They have the lowest quantitative indicators in the first two variants of the experiment with the applied traditional treatments. The higher the content of organic matter in the variants, the greater the amount of actinomycetes. Their maximum value was observed in variant $f_3$ during the maximum growth of corn – respectively, 5.92x10$^6$ CFU/g.

The amount of mold fungi in a large part varies less, but is highest in the variant with minimal treatments and vertical mulching, and the variant with surface mulching $f_3$ with values for the three phases of the experiment, respectively, 0.00303, 0.00152 and 0.00161x10$^6$ CFU/g and $f_2$ 0.00243, 0.00123 and 0.00089x10$^6$ CFU/g, respectively.

Nitrogen-fixing microorganisms were most abundant in variant $f_2$ -12.73 at maximum growth of maize. The results obtained for the group of cellulose-degrading microorganisms show that, before sowing the corn, they are the most in variant $f_2$ -3.31x10$^6$ CFU/g, followed by variant $f_3$ – 2.87x10$^6$ CFU/g. During the next phase, maximum growth, the highest number of 2.45x10$^6$ CFU/g was reported for variant $f_3$, followed by $f_2$.

In the last phase, after harvesting the corn, the

number of cellulose-degrading microorganisms was the highest in variant $f_2$ -2.74x$10^6$ CFU/g, followed by $f_3$. Option $f_1$ is characterized by the lowest number. The control variant $f_0$ with traditional treatment along the slope of the slope was characterized by the lowest numbers of the main groups of microorganisms, in general. This is due to soil compaction. In contrast, the microbiota of the variant with traditional treatment across the slope of the $f_1$ slope, was more abundant than that of the null variant. For the variant with surface application of manure, the introduction of organic matter has a favorable effect on the amount of microorganisms.

In variant $f_3$, with vertical mulching with manure, the soil microbiota is more strongly affected compared to variant $f_2$. This is explained by the greater depth of application and the combination with minimal tillage.

In the variant with surface application of manure, $f_2$ the introduction of organic matter increases the number of microbiota. The amount of spore-forming bacteria on average in this variant is the highest.

Over the three years, this variant showed a 1.13 - 1.94 higher amount of saprophytic bacteria compared to $f_0$, as well as the highest content of spore-forming bacteria. The amount of nitrogen-fixing bacteria increased 1.45 to 2.7 times on average over the three years, and cellulose-degrading bacteria was 5 times higher. From the data in table 1 shows, that cellulose-degrading microorganisms in variants $f_2$ and $f_3$ are 5.31 and 6.12 times higher compared to variant $f_0$.

The increased amounts of plant residues, root mass and organic matter content stimulate the activity of soil microorganisms, which has a positive effect on the structure, composition and properties of the soil.

As a result of the studies, the applied manure has a positive effect on the microflora and related processes in the soil, activating the microbiological activity. Under the influence of fertilization, changes occur not only in the microflora, but also in the enzyme activity of the soil.

## Conclusion

Genetic specificities, topographic features and land use have an impact on the physical properties of the soil, as well as on the development and number of soil microflora. Microbiological analyzes showed that minimum tillage, combined with vertical mulching, had a positive effect on soil microbiological properties when growing maize on moderately eroded carbonate chernozem on sloping terrain.

The low number of microorganisms in option $f_0$ is due to the lower porosity, lower moisture content and compaction of the soil. Soil microflora is a very sensitive indicator, that can be used to determine the erosion processes taking place in the soil, soil treatments, etc.

***Acknowledgments***

The authors thank FNI-MONS for funding the scientific research under Contract KP-06 H46/1 2020 on the topic "Effectiveness of anti-erosion agrotechnologies for improving the quality and hydrological regime of the soil and limiting greenhouse gas emissions".

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