

EFFECTS OF IRRIGATION AND AMELIORATION TECHNOLOGY ON MICROBIOLOGICAL AND ENZYMATIC PROPERTIES OF SOIL

IRINA SENIKOVSKAYA, V. FILIPCHIUK

“Nicolae Dimo” Institute of Soil Science, Agrochemistry and Soil Protection of Chisinau

Keywords: *irrigation, microorganisms, enzymes, amelioration, wastes*

Abstract

The significant negative effects on the microbiological and enzymatic properties of typical chernozem located in the Northern zone of the Republic of Moldova were observed as a result of long-term irrigation with saline water. The stocks of microbial biomass in the 0 - 160 cm layer were of 8073,5; 5274,2 and 4486,9 kg ha⁻¹ in the fallow chernozem (natural standard), arable un-irrigated and arable irrigated chernozems respectively. A similar trend has been noticed in the vertical distribution of enzymatic activities. There was a 3,6 - fold increase in the amount of humus - mineralizing microorganisms.

A soil management with the application of wastes (sugar industry sugar lime, manure) created conditions for the improvement of the microorganisms' vital activity in the degraded chernozem. Microbiological and enzymatic soil parameters depended on the doses of sugar lime material and the content of Na⁺ in the absorbing complex. A scale for assessing the ameliorative status of irrigated soils and for developing national standards of soil quality was elaborated. The use of sugar industry waste (the reserve of five years) with manure 60 t ha⁻¹ has been recommended for the regeneration of microbial communities, immobilization of enzymes and improvement of soil quality.

INTRODUCTION

The degradation of soils in the Republic of Moldova is the most critical, threatening problem for agriculture, for environment and people's habitat. Significant negative influence of the irrigation management using unsatisfactory water quality on soil's physical and chemical properties was observed. The mitigation of negative consequences caused by irrigation may be achieved by applying different wastes. Annually, about 250-300 thousand of sugar industry waste materials (sugar lime) accumulate in the country [1]. This waste is the liming material with an alkaline reaction (pH=7.75-8.05), it has a moderate organic matter content (3.8-4.2%). It has been determined that sugar lime is a biologically active material, in which bacteria are predominant among microbial population. Preliminary researches demonstrated that the biota's status of irrigated soils depends on the irrigation cycles, water quality, peculiarities of plants and other factors. On the other hand, microbiological processes are important for the transformation of wastes of different compositions in soils. That is why the

problem of the evaluation of the new reclamation technology to reduce the degradation of irrigated chernozem is so up-to date. The purpose of the research was to determine the influence of the irrigation management and amelioration technology by applying wastes (sugar industry waste material, manure) on the change of the microbiological and enzymatic properties of soil and to develop scale parameters of the stability of the microbial community.

MATERIAL AND METHODS

Experimental site. The experimental site is located in the northern zone of the Republic of Moldova. For a period of 23-25 years, the irrigation was carried out using poor water quality from the Reut river. The mineralization of irrigation water fluctuated between 0.780 and 1.200 mg dm⁽³⁾⁻¹; pH=8.4-8.6; SAR=4-6. The experiment was replicated four times in a randomized split-plot design without using waste management treatments and by applying sugar lime material annually, by dose with a reserve for three years and dose with a reserve for five years (24.9; 26.8 and 28.8 t ha⁻¹ accordingly). The dose of the ameliorant was calculated taking into account the full replacement of the exchangeable Na⁺ and the content of this element in the irrigation water. Reclaimed plots received 60 t ha⁻¹ of manure. The research was conducted in 2007-2009. Before the experiment, the soil indices in the 0-30 cm layer were as it follows: humus content - 4.21%; P₂O₅ - 5.30 mg 100 g⁻¹; K₂O - 30.4 mg 100g⁻¹; water-soluble salts content - 0.064 %; exchangeable Na⁺ was 4.73 me 100g⁻¹ or 12% from the total exchangeable cations; pH=7.6.

Soils. The site's soil is a typical chernozem. The irrigation effect was studied by comparing irrigated chernozem with un-irrigated and fallow (natural standard) chernozems. Sampling was carried out in three soil profiles to a depth of 170 cm. Samples of the experimental plots were collected from 0-30 cm and 30-50 cm layers twice during the growing season.

Microbiological properties. Microbial biomass (MB) was measured by the rehydration method based on the difference between C extracted with 0.5 M K₂SO₄ from dried soil at 65-70⁰C for 24 h and fresh soil samples by K_c coefficient of 0.25. K₂SO₄ - extractable organic C concentrations in the dried and fresh soil samples were simultaneously measured by dichromate oxidation. The quantity of K₂SO₄ - extractable C was determined at 590 nm with "Specol-221" spectrophotometer (Germany). Stocks of the MB have been calculated taking into account the carbon content of the microbial cell and the bulk density of soils.

Counts of culturable microorganisms (heterotrophic bacteria, humus-mineralizing microorganisms, bacteria from *Azotobacter* genus and fungi) were obtained on agar plates.

Enzymatic activity. The dehydrogenase activity (potential) was determined by the colorimetric technique on the basis of occurring triphenylformazan (TPF) from

TTC (2,3,5-triphenyltetrazolium chloride) added to air-dry basis of soil. The polyphenoloxidase activity (potential) was determined by the colorimetric technique on the basis of the oxidation of phenolic compounds to quinones.

Soil chemical properties. Organic C was analyzed by the dichromate oxidation method. Available P and K was extracted with 1% $(\text{NH}_4)_2\text{CO}_3$ by the Olsen method. P_2O_5 was determined by colorimetric technique, K_2O - by flame photometric method. Exchangeable cations were extracted with 1M NH_4Cl in the 60% $\text{C}_2\text{H}_5\text{OH}$ at pH=8.5; Ca and Mg in the extracts were analyzed by the complexometric method, Na - by the flame photometric technique. Soil pH was measured in a 1:2.5 soil : water slurry using a glass electrode. The microbiological and enzymatic indices were evaluated statistically by analysis of variance.

RESULTS AND DISCUSSION

The influence of irrigation. The long-term irrigated typical chernozem showed a significant decrease in the microbiological and enzymological indices as compared to un-irrigated and fallow soils (Table 1, Figure 1). The highest level of the microbial biomass and organic carbon content as well as enzymatic activities have been determined in the A_1 horizon of the fallow chernozem and whereas the lowest - in the BC and C horizons of all profiles. The stock of microbial biomass in the 0-160 cm layer of the arable irrigated chernozem decreased by 14.9% as compared with the un-irrigated chernozem and was 1.8 times lower than in the fallow soil.

Irrigation affected the structure of soil microbial communities. There was a 3.6 - fold increase in the number of humus-mineralizing microorganisms. Irrigation led to decreases in the number and diversity of soil fungi. Shannon index went down from 2.53 in the fallow chernozem to 2.28 in the irrigated chernozem. In the irrigated soil there were fungi of *Penicillium*, *Fusarium* families and *Alternaria alternata* species. A negative impact on soil's enzymatic properties was observed as a result of the irrigation with poor water quality (Figure 1). Dehydrogenase and polyphenoloxidase activities in the soil profiles increased with its depth. The dehydrogenase activity in the 0-50 cm of the irrigated chernozem was 1.2 - 5.2 times lower than in un-irrigated and fallow soils. The profile distribution of the polyphenoloxidase activity was more complex. However, the inhibitory effect of irrigation on the activity of this enzyme is obvious.

Modifications of the microbiological and enzymatic properties in the degraded chernozem are reported as by changes in the chemical and physical properties of the soil. Negative shifts in the state of microorganisms and enzymes occurred because of the soil alkalization, changes in the composition of the organic matter and the deterioration of the soil structure. The number of exchangeable Na^+ in the irrigated chernozem reached 12%, which corresponds to the moderate degree of alkalization.

Table 1

Profile distribution of the organic C and microorganisms in the irrigated typical chernozem in comparison with un-irrigated and fallow soils

Genetic horizon and depth, cm	Organic C, %	MB		Heterotrophic bacteria, CFU g ⁻¹ soil*10 ⁶	Humus-mineralizing microorganisms, CFU g ⁻¹ soil*10 ⁶	Stock of MB, kg ha ⁻¹ (0-160 cm)
		μg C g ⁻¹ soil	kg ha ⁻¹			
Typical chernozem (40-year-old fallow land)						
A ₁ 0-3	3.53	979.5	640.6	10.5±1.29	5.7±0.12	8073.5
A 3-38	2.64	519.2	4070.5	6.1±0.64	4.9±0.07	
B ₁ 38-79	2.16	298.3	2959.7	1.3±0.06	0.6±0.03	
B ₂ 79-101	0.71	66.8	402.7	0.6±0.03	0.2±0.01	
BC101-123	0.59	0	0	0.07	0.2±0.01	
C 123-170	0.62	0	0	0	0	
Un-irrigated arable typical chernozem						
Ap 0-25	2.45	237.5	1460.6	5.2±0.38	15.9±0.28	5274.2
A 25-41	2.14	206.9	1387.9	4.1±0.21	9.8±0.18	
B ₁ 41-64	1.47	176.6	1121.1	1.8±0.10	2.8±0.10	
B ₂ 64-89	0.85	107.5	763.3	0.3	0.6±0.03	
BC 89-114	0.51	22.4	160.2	0.1	0.2	
C 114-160	0.60	27.8	381.1	0	0.4	
Arable typical chernozem irrigated with water from the Reut river						
Ap ₁ 0-21	2.45	214.8	1127.7	5.4±0.36	19.5±0.25	4486.9
A ₂ 21-35	2.23	213.9	778.6	5.3±0.11	15.3±0.09	
A 35-49	2.02	193.3	774.0	4.5±0.05	12.8±0.02	
B ₁ 49-69	1.10	164.0	944.6	2.6±0.09	4.7±0.10	
B ₂ 69-91	0.87	92.9	605.0	1.3±0.09	2.3±0.04	
BC ₁ 91-110	0.67	45.7	257.0	0.8±0.02	0.3±0.01	
C 110-150	0.56	0	0	0	0	

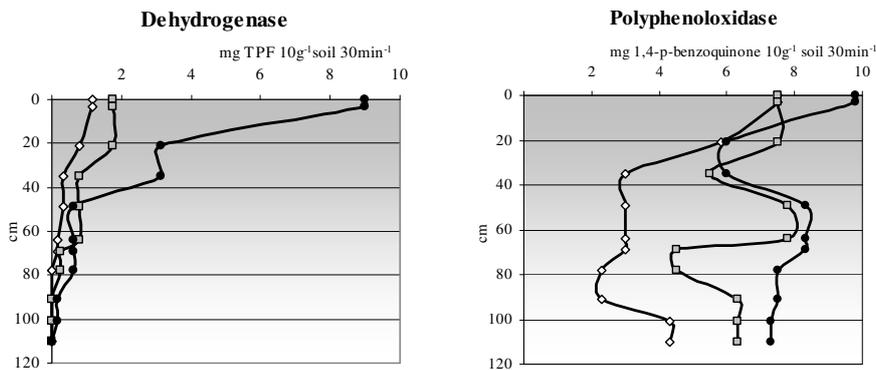


Fig. 1. Effects of long-term irrigation on the profile distribution of enzyme activity of typical chernozem (—●— fallow; - -□- - un-irrigated; ···◇··· irrigated)

The soil became slightly alkaline, pH increased from 7.1 to 7.6. The data presented in these studies demonstrate that the long-term irrigation of typical chernozems with water from the Reut river has negatively affected the microbial communities and the soil in general. The values of microbiological and enzymatic indices were typical for degraded chernozems. These soils have a low ecological stability and need the amelioration of the soil's root layer.

Effects of amelioration technology. The application of wastes to improve the ameliorative status of irrigated soils changes the habitat of microorganisms. The ameliorants improve the microbiological and enzymatic properties of the irrigated chernozem. The modifications that occurred were higher in the 0-30 cm layer than in the 30-50 cm layer. However, there were significant differences in the enzymatic activities between the values of the control (un-irrigated) plot and the amended plots to a depth of 50 cm. Positive effects of wastes on soil chemical and physical properties and organic matter content promote microbial proliferation in the soil's root layer. The use of wastes (3 and 5 years reserve) together with 60 t ha⁻¹ of manure led to an increase of the microbial biomass content from 170.3-217.7 µC g⁻¹ soil to 245.0-302.3 µC g⁻¹ soil in the 0-30 cm layer. This fact has been interpreted as the response of soil microbial communities to the favorable environmental conditions.

Table 2

Scale of microbiological and enzymatic parameters (confidence intervals) of the irrigated typical chernozem (0-30 cm)

Degree of stability	MB, µg C g ⁻¹ soil	Viable counts of microorganisms		Enzymatic activity	
		Humus miner.micr., CFU g ⁻¹ soil*10 ⁶	<i>Azotobacter</i> , CFU g ⁻¹ soil	Dehydrogenase, mg TPF 10g ⁻¹ soil 24h ⁻¹	Polyphenoloxidase, mg 1,4-p-benzoquinone 10g ⁻¹ soil 30 min ⁻¹
Control (Na ⁺ , 12 % from the total exchangeable cations)					
Low	170-218	18 – 19	111 – 115	1.2 – 1.9	5.3 – 6.9
Sugar lime applied annually (Na ⁺ , 10 % from the total exchangeable cations)					
Low	219-313	16 – 17	151 – 185	1.7 – 3.2	8.1 – 13.7
Sugar lime, 3 years reserve (Na ⁺ , 8 % from the total exchangeable cations)					
Moderate	249-302	13 – 16	195 – 231	1.9 – 4.0	9.8 – 14.0
Sugar lime, 5 years reserve (Na ⁺ , 7 % from the total exchangeable cations)					
Moderate	245-291	11 – 14	232 – 254	2.5 – 4.7	11.3 – 16.5

Wastes amended soils showed a significant increase in the counts of culturable microorganisms. The number of heterotrophic bacteria increased on average by 18-37%. There was a 1.5-2.2-fold increase in the counts of bacteria from *Azotobacter*

genus compared to the un-irrigated control plot. Counts and diversity of fungi were higher under application of wastes. Shannon index raised from 2.28 to 2.45. As a result of the application of waste with in reserve for 5 years the counts of humus - mineralizing microorganisms decreased by 30%.

The amelioration technology significantly improves the enzymatic status of irrigated chernozem. According to the statistical parameters, the dehydrogenase activity in the 0-30 cm layer increases by 1.6-2.3 times and the polyphenoloxidase activity - by 1.8-2.3 times depending on the ameliorants doses. A similar trend has been noticed in enzymatic activities in the 30-50 cm layer.

The microbiological and enzymatic parameters of soil depended on the doses of sugar lime material and the content of Na^+ in the absorbing complex. A scale to estimate the irrigated chernozem reclamation was elaborated (Table 2). The use of sugar industry waste (the reserve for five years) with manure 60 t ha^{-1} has been recommended for the regeneration of microbial communities, immobilization of enzymes and improvement of soil quality.

CONCLUSIONS

1. The long-term irrigation of the typical chernozem with saline water leads to its degradation. This is reflected in the deterioration of soil microbiological and enzymatic properties. The stocks of microbial biomass decreased by 1.8 times in comparison with the standard. The enzymatic activity went down 3.2 times. The structure of microbial community changes. In the irrigated soil humus destroying microorganisms dominate. The values of the microbiological and enzymatic indicators, the organic carbon content decrease in the following sequence: typical chernozem (40-year-old fallow land) → typical un-irrigated arable chernozem → typical irrigated arable chernozem. The soil's root layer is covered by degradation processes.
2. The soil management with the application of wastes (sugar industry sugar lime, manure) created conditions for the improvement of the microorganism's vital activity in the soil which degraded as a result of irrigation. The microbial system is stabilized, increasing the microbial biomass content in average by 1.4 times, improving biodiversity, reducing the content of humus-mineralizing microorganisms by 30% and activating enzymes. A microbiological method to estimate the ameliorative status of irrigated soils has been elaborated. A scale of microbiological and enzymatic parameters for the national soil quality standards has been proposed.

REFERENCES

1. xxx, 2004. *Programul complex de valorificare a terenurilor degradate și sporirea fertilității solurilor*. Partea a II-a, Red. resp. S. Andrieș. Chișinău, Pontos (pp. 38).