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Enzymatic activity and microbiological characteristics of luvic and pseudogley soils in western Slavonia

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Abstract

Background and Purpose: *Soil microorganisms participate in major processes associated with sustainable soil management and ensure cycling of nutrients required for plant production. Their role is of special importance for the dynamics of generation and decomposition of soil organic matter, and they also take part in other processes that provide soil fertility and health. Different anthropogenic influences as well as mechanical and chemical properties of soil have a significant influence on the presence of functional groups of microorganisms and on enzymatic activities in the plough-layer. The main research aim was to study the microbial characteristics and enzymatic activity in pseudogley and luvic soil types of western Slavonia.*

Materials and Methods: *Soil samples collected from soil profiles differently affected by anthropogenic action, from four localities in western Slavonia were analyzed.*

Results: *Average total number of micoorganisms was 13.90 × 10^6 CFU/g soil in pseudogley and 21.33 × 10^6 CFU/g soil in luvic soil. In pseudogley the proportion of functional groups was poor to medium and proteolytic activity was 6.21 (gelatine units/g soil). In luvic soil very good proportion of aerobic asymbiotic nitrogen fixators and cellulolytic fungi was determined. The average proteolytic activity was 5.08 gelatine units/g soil. Research results for cellulolytic activities in luvic soils and pseudogley show that higher values were determined for endogluconase than for cellobiohydrolase.*

INTRODUCTION

In the last decades, agricultural production has been faced with an ever increasing demand for food to feed the growing world population. On the other hand, it is burdened by continuing reduction of agricultural land caused by different types of land damage or its permanent reallocation. A major land damage process is soil depletion of humus and decrease in the number and diversity of microbial populations in soil, that is, reduced soil biogenity (*9*). Soil microorganisms participate in major processes associated with sustainable soil management and ensure cycling of nutrients required for plant production (*5, 12*). Their role is of special importance for the dynamics of generation and decomposition of soil organic matter, and they also take part in other processes that provide soil fertility and health. Intensive field crop production has so far mostly ignored the importance and function of the biological component of soil. This was the reason for the numerous negative processes reflected in deterioration of physical and chemical soil properties (*4*). Studies of microbial populations in agricultural soil and assessment of their enzymatic activity provide information on soil quality, namely its property essential for agricultural production – soil fertility (*7, 9, 10*)

As soil quality is also reduced by erosion, contamination and loss of soil organic matter, modern agricultural production is no more focused on high yields but requires from agronomists to apply new findings to contribute to controlled production of all crops and to conserve soil as an irreplaceable resource. Luvic soil is the most widespread soil type in western Slavonia and agricultural production areas are mainly developed on luvic soils. According to the effective soil classification in Croatia, luvic soil belongs to the class of eluvial-illuvial soils within the group of automorphic soils. Luvic soil is characterized by the presence of eluvial and illuvial horizons, or A-E-Bt-C (humus accumulative horizon; E-eluvial horizon; Bt argiluvic horizon; C-parent material) profile structure. However, due to deep tillage, the upper horizons are mixed in agricultural soil, so the profiles studied in this study are structured P-Bt-C (P refers to the anthropogenic plough horizon). Along with luvic soils, agricultural production in western Slavonia is also carried out on pseudogley soils, which mostly have unfavorable physical and chemical properties, making this soil type marginally suitable for agriculture. However, pseudogley soils is widely cultivated due to its spread on as much as 558,000 ha (*1*), favorable climate and suitable relief. It is a soil type that belongs to the pseudogley class of the division of hydromorphic soils (*13*). It mainly occurs on mildly sloping terrains and mesoelevations in Pannonian Croatia. Pseudogley is characterized by the exchange of dry and wet periods in which processes of reduction and oxidation go on, respectively. Profile structure of this soil is A-Eg-Btg-C (A-humus accumulative horizon; Eg-eluvial pseudogleyed horizon; Btg-argiluvic pseudogleyed horizon, C-parent material). On agricultural land, at a depth of ca. 30–70 cm, pseudogley has an impermeable or slowly permeable gley (g) horizon on which water stagnates. Owing to the presence of the slowly permeable horizon, the wet soil phase prevails in the spring-winter-autumn part of the year and wet and dry soil phases in summer (*3*). These periodical changes cause redox processes, which result in typical marmorized appearance of the diagnostic g horizon.

The principal goal of the investigations was to study the functional groups of microorganisms, total number of microorganisms and enzymatic activities in the plough-layer of the two most widely spread soil types of western Slavonia.

MATERIAL AND METHODS

Soil samples for pedological and microbiological analyses were taken from twenty soil profiles in the plough layers of four locations in western Slavonia (Višnjica, Čađavica, Senkovac). Pedological analyses were performed by standard methods as described in the soil investigation manual (*13*). Microbiological analyses included

TABLE 1

Some physical and chemical soil properties of anthropogenic luvisol soil type.

Location	Systematic unit	Depth cm	Clay %	Soil texture*	Gv* g/cm^3	Kz* %	Soil pH		Humus %	V* %
							H_2O	M KCl		
Višnjica	Luvisol on loess typical, on pad of send	0–40	11.0	PrI	1.51	13.0	5.4	4.5	1.7	66.1
	Luvisol on loess pseudogleyic, on pad of send	0–36	11.2	PrI	1.55	8.2	5.9	5.1	1.9	63.5
	Luvisol on loess, pseudogleyic	0–35	12.4	PrI	1.37	17.2	5.4	4.6	1.2	63.0
Čađavica	Luvisol on loess typical, on pad of send	0–30	9.0	PrI	1.19	16.7	4.8	4.0	1.4	17.8
	Luvisol on loess typical, on pad of send	0–35	8.8	PI			5.4	4,7	1.3	55.6
	Luvisol on loess pseudogleyic, on pad of send	0–30	12.8	PrI	1.50	7.8	5.4	4.2	1.7	39.7
	Luvisol on loess pseudogleyic, on pad of send	0–40	11.2	PrI	1.52	8.9	4.6	4.0	1.1	37.2
Senkovac	Luvisol on loess typical, on pad of send	0–38	11.6	PrI	1.45	18.5	5.1	4.1	1.5	41.5
	Luvisol on loess pseudogleyic, on pad of send	0–40	12.6	I	1.46	17.4	5.1	4.2	1.2	35.8
	Luvisol on loess pseudogleyic, on pad of send	0–35	9.6	I	1.61	14.8	4.8	4.0	1.2	33.6

* Gv – Volume density; Kz – soil capacity for air; V – base saturation; PI – sandy-loam; PrI – silty-loam; I-loam

determination of the total number of microorganisms, the presence of functional groups of aerobic and anaerobic nitrogen fixators, cellulolytic bacteria and fungi as well as nitrificators in soils, and also determination of proteolytic and cellulolytic enzymatic activities. Total number of microorganisms was determined by the dilution method on agarized soil extract. Aerobic asymbiotic nitrogen fixators, cellulolytic microorganisms and nitrificators were determined by the silica gel plating method using appropriate selective nutrient media. Anaerobic nitrogen fixators were determined by the dilution method on selective nutrient media under anaerobic culture conditions. The obtained values represent the mean values of three replicates on 1 gram dry soil basis. Aerobic nitrogen fixators, cellulolytic microorganisms and nitrificators were determined on the basis of the percent of »fertile« soil grains (*6*). Soil proteolytic activity was determined by the titration method after Romejko (1969); activities of the enzymes endogluconase and cellobiohydrolase were measured by the method of Kong, Balandreau, Dommergues (1971).

RESULTS AND DISCUSSION

Results of pedological analyses are given in Table 1 and indicate that luvic soils on loess are mostly of silty-loam texture. Very acid to acid reaction and low base saturation of the soil are serious limitations, which should be eliminated by liming. Humus content in the plough layer varies from 1.1–1.9%, that is, within the limits of humus-poor soils. In intensive agricultural production, adequate agrotechnical management practices should be applied to raise the humus content to at least 3% and then maintain that level. According to many authors, the total number of microorganisms is a major indicator of soil biogenity (*2, 7, 8, 11, 14*). On the basis of the total number of microorganisms in soil, Redžepović (*8*) divides soil biogenity into five categories, ranging from 11 × 10^6 CF U/g soil (low biogenity) to 100 × 10^6 CFU/g soil (very high biogenity). Results of microbiological analyses given in Table 2 show that luvic soils of the studied locations are of medium biogeny, the average values of the total number of microorganisms being 21.33 × 10^6 CFU/g soil. Reasons for medium biogeny lie in the acidity and rather low humus content of luvic soils. Values of the total number of microorganisms range from 7.67–44.3 × 10^6; the differences are most likely due to different supplies of available nutrients. Regarding functional groups, very good proportion of aerobic asymbiotic nitrogen fixators (80.14% on average) and cellulolytic fungi (78.94%) was determined. Proportion of nitrificators was most strongly affected by soil acidity and aeration. Acid reaction of analyzed soil samples is the cause of the lower proportion of nitrificators, notably nitrate bacteria (28.07% on average). Proteolytic activity is low, generally amounting to 5.08 gelatine units/g soil, and is due to the low humus content in analyzed soil samples. Research results for cellulolytic activity in luvic soils show that higher values were determined for endogluconase (0.44 mg red glucose/g soil) than for cellobiohydrolase (0.26 mg red glu-

TABLE 2

Microbiological characteristics and enzymatic activity of luvic soils.

Location	Systematic unit	Total number of microrga-nisms 1 × 10^6	Endgluconase (mg red. glucose/1 g soil)	Cellobiohydrolase (mg red. glucose/g soil)	Proteolytic activities (gelatine units/g soil)	Aerobic free living N fixers (%)	Anaerobic free-living N fixers 1 × 10^4	Cellulolytic mo. (%)		Nitrifying bacteria (%)	
								Fungi	Bacteria	Nitrite-oxidizing bacteria	Ammonia-oxidizing bacteria
Višnjica	Luvisol on loess typical	20.0	0.42	0.23	3.37	88.00	28.00	94.67	10.67	17.33	10.0
	Luvisol on loess pseudogleyic	44.3	0.42	0.41	7.25	90.67	20.67	73.33	21.33	25.33	4.0
	Luvisol on loess pseudogleyic	15.67	0.50	0.23	6.12	56.00	28.34	32.0	98.68	38.68	16.0
Čađavica	Luvisol on loess (P-169,P-233)	7.67	0.45	0.23	3.42	78.67	19.67	100.0	12.0	64.0	44.00
		13.67	0.42	0.20	3.61	92.00	16.67	92.0	2.67	74.67	62.67
	Luvisol on loess pseudogleyic (P-107; P-131)	19.67	0.42	0.41	4.40	86.68	12.67	22.68	18.68	37.32	1.32
		23.00	0.38	0.18	3.08	82.67	22.33	96.0	26.67	86.67	96.0

cose/g soil). Cellulolytic activity in soil depends on the ecological conditions of the agrosystem (aeration, pH, moisture, humus content), the presence of cellulolytic microorganisms, and also on the crop grown. Good proportion of cellulolytic fungi was found in the studied soil samples, but cellulolytic activity was moderate owing to less favorable conditions (pH, humus content) and lower proportion of cellulolytic bacteria as the main cellulose degrading agents in soils.

Results of pedological analyses given in Table 3 indicate that pseudogley soils are characterized by an increased content of clay particles in the sub-plough horizon, with textural designation varying from silty clay loam to silty loam. Values of volume density and soil air capacity in the sub-plough layer indicate high compaction, dense particle packing, slow water permeability and sporadic stagnation of precipitation water, pointing to the need of soil loosening. Very acid to acid soil reaction and low to medium soil saturation with bases are also serious limitations, which should be eliminated by liming. As the humus content in the plough-layer is about 2.0 % (range from 1.7 to 3.1 %) i.e., within the limits of humus-poor soils, agrotechnical measures should be applied in intensive agricultural production to raise the humus content to at least 3 %, and maintain this level in agricultural practice.

The chemical characteristics of soil, such as low pH value, proportion of available nutrients and organic matter content in soil, influenced also the number and activity of microbial populations in the studied soil type (Table 4). Total number of microorganisms in soil

samples ranged from 1.33×10^6 CFU/g soil (Bukovica location) to 30.00×10^6 CFU/g soil (Čađavica location); thus, the soils are of low to medium biogenity (*8*). The average total number of microorganisms in pseudogley samples is 13.90×10^6 CFU/g soil. Regarding functional groups, there is a very good proportion of aerobic nitrogen fixators (71.62, on average) and high proportion of cellulolytic fungi at all locations (89.67 %, on average). The number of nitrificators in soil is mainly influenced by soil acidity and aeration (*2, 11*). Acid reaction of studied soil samples is the cause of the smaller amount of nitrificators, particularly nitrate bacteria (36.11 %, on average). Proteolytic activity was 6.21 (gelatinous units/g soil) and was due to the low humus content in soil samples. Average cellulolytic activity of pseudogley indicates a higher proportion of endogluconaze (0.49 mg reduced glucose/g soil) compared to lower proportion of cellobiohydrolase (0.27 mg red. glucose/g soil). Fair pro-

TABLE 3

Some physical and chemical properties of pseudogley soil type.

Location	Pedosystematic units	Soil depth cm	Clay %	Soil texture*	Gv* g/cm^3	Kz* %	pH in H_2O	pH in M KCl	Humus %	V* %
Čađavica	Pseudogley of level terrains, semigleyic, medium deep, with short to medium long wet phase	0–35	16.4	PrI	1.54	5.5	5.0	3.8	2.3	38.8
		35–85	28.6	PrGI	1.61	4.8	5.8	4.5	0.8	91.6
	Pseudogley of level terrains, semigleyic, medium deep, with short to medium long wet phase	0–35	19.2	PrI	1.37	5.1	6.4	5.3	2.5	69.7
		35–70	25.8	PrI	1.60	4.2	7.0	6.2	0.9	89.2
Bukovica	Pseudogley of level terrains, semigleyic, medium deep, with long wet phase	0–43	27.2	PrI	1.42	9.4	5.9	4.6	3.1	55.8
		43–80	37.8	PrGI	1.51	5.0	6.7	5.7	1.3	76.9
	Pseudogley of level terrains, semigleyic, medium deep, with long wet phase	0–40	9.8	PrI	1.38	5.5	5.5	4.1	2.4	53.5
		40–78	34.4	PrGI	1.52	4.0	6.0	4.5	1.1	67.6
Senkovac	Pseudogley of level terrains, medium deep, with medium long to long wet phase	0–30	12.8	PrI	1.49	13.7	4.6	3.8	2.0	24.2
		30–72	23.6	PrI	1.60	5.7	5.7	4.4	0.6	63.6
	Pseudogley of level terrains, medium deep, with medium long to long wet phase	0–32	13.6	PrI	1.33	11.7	5.5	4.5	1.7	
		32–80	20.4	PrI	1.47	7.2	6.7	5.2	0.6	
Višnjica	Pseudogley of level terrains, semigleyic, medium deep, with long wet phase	0–35	9.0	PrI	1.40	13.0	7.2	7.0	2.1	
		35–73	21.4	PrI	1.57	6.2	6.4	6.1	0.7	
	Pseudogley of level terrains, semigleyic, medium deep, with long wet phase	0–40	9.9	PrI	1.21	12.8	5.1	4.2	2.0	37.6
		40–73	21.2	PrI	1.49	7.7	5.8	4.9	1.0	78.1

* Legend: Gv – soil bulk density; Kz – soil capacity for air; V – soil adsorption complex base saturation; PrI – silty loam; PrGI – silty clay loam

TABLICA 4

Microbiological characteristics and enzymatic activity of pseudogley soils.

Location	Systematic unit	Total number of microorganisms 1×10^6	Endgluconase (mg red. glucose/1g soil)	Cellobiohydrolase (mg red. glucose/g soil)	Proteotlytic activities (gelatine units/g soil)	Aerobic free-living N fixers (%)	Anaerobic free-living N fixers 1×10^4	Cellulolytic mo. (%)		Nitrifying bacteria (%)	
								Fungi	Bacteria	Ammonia-oxidizing bacteria	Nitrite-oxidizing bacteria
Bukovica	Pseudogley of level terrains	15.33	0.46	0.32	3.20	13.33	15.33	98.68	0.0	25.32	36.0
	Pseudogley	1.33	0.48	0.26	5.31	62.68	6.33	100.0	4.0	9.32	4.00
	Pseudogley of level terrains	7.67	0.53	0.25	7.24	74.68	15.67	92.0	10.68	44.00	56.0
	Pseudogley of level terrains	6.67	0.55	0.25	6.20	60.00	15.00	100.0	10.68	61.32	36.0
Višnjica	Pseudogley of level terrains	22.33	0.61	0.38	4.97	100.0	8.33	66.67	6.67	38.67	18.67
	Pseudogley of level terrains	8.5	0.63	0.51	7.44	8.0	18.67	89.33	9.33	19.0	9.33
Čađavica	Pseudogley of level terrains, semigleyic P-81; P-189)	30.00	0.42	0.18	5.14	98.68	15.67	98.68	40.0	58.68	42.68
		10.67	0.48	0.25	4.97	100.0	24.67	80.0	28.0	48.0	28.0
	Pseudogley of level terrains (P-125; P-227)	20.33	0.51	0.28	3.32	86.67	22.33	100.0	44.0	58.67	88.0
		11.00	0.37	0.12	2.97	90.67	8.67	50.67	0.0	58.67	90.67
Senkovac	Pseudogley of level terrains	19.67	0.48	0.24	10.84	95.33	26.33	100.0	6.67	14.68	12.00
	Pseudogley of level terrains	13.34	0.36	0.23	12.96	69.36	8.67	100.00	16.00	44.00	12.00
Average number		13.90	0.49	0.27	6.21	71.62	15.47	89.67	14.67	40.03	36.11

portion of cellulolytic fungi was determined in the studied soil samples, but not so good cellulolytic activity due to less favorable conditions (pH, humus content). There was also a considerable number of cellulolytic bacteria, significant cellulose degraders in soils (*2, 9*).

CONCLUSIONS

The proportion of functional groups of microorganisms and their enzymatic activity in soils are dependent on soil mechanical and chemical properties as well as on different anthropogenic influences. The up-to-date approach focusing on sustainability, biological diversity and soil conservation influences investigations, which can contribute to the knowledge of microorganisms and enzymes responsible for all the major biochemical processes in soil.

Low biogenity of luvic and pseudogley soils of western Slavonia is due to the agricultural management in which high yields of field crops were the main criterion of efficacy. To achieve this aim, mineral fertilization was abundant, harvest residues were more often burnt than ploughed-in, stable manure was hardly used and no liming was applied. Modern agricultural production is based on sustainability, preservation of natural resources and soil protection, so monitoring of the microbiological component and its activity in soil can provide the insights necessary for a rational, sustainable and environmentally acceptable agricultural production. Besides, only integral agroameliorative and agrotechnical practices, primarily deep loosening of soil, liming and ploughing-in of harvest residues as well as application of organic fertilizers can reduce further soil degradation in western Slavonia.

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