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MICROBIOLOGICAL ACTIVITY AND PRODUCTIVITY OF SOIL IN PLUM ORCHARD

M. PESAKOVIC[a]*, D. DJUKIC[b], L. MANDIC[b], R. MILETIC[a]

[a]*Fruit Research Institute, 1/9 Kralja Petra Street, 32 000 Cacak, Serbia*
[b]*Faculty of Agronomy, University of Kragujevac, 34 Cara Dusana Street, 32 000 Cacak, Serbia*
E-mail: *marijanap@tfc.kg.ac.rs*

Abstract. Microorganisms population, as major soil component, is very significant indicator of soil fertility, i.e. the process of soil degradation. Microbial population density and enzyme activity of microorganisms may be highly reliable practical indicators which can suggest the answer to the question concerning conditions that favour plant production. The influence of different NPK fertiliser rates on microbiological activity of the alluvial soil planted with plum culture has been studied over a 3-year period. The effect of the mineral fertiliser used was determined 3 times over the growing season and was monitored through determinations of the number of different groups of microorganisms and proteinase activity in the soil. The 3-year results suggest that the effect of the applied fertilisers was induced by numerous factors, which primarily refers to climate effects and its specific elements, the activity of the plant itself and its root metabolism, as well as the effect of soil specificities, i.e. its structural, chemical and agro-chemical properties. Taking into account biological properties of soil, plum production, economic and environmental indicators of the results of the stated treatments, the rate of 600 kg ha^{-1} can be recommended for growing plum under the stated environmental conditions.

Keywords: microorganisms, proteinase activity, mineral fertilisers, soil.

AIMS AND BACKGROUND

Nitrogen fertiliser is the most important and widely used chemical in farming[1]. Nitrogen is required for normal growth of a great number of plants. Even in soils richest in organic matter its content does not exceed 4% (Ref. 2). Regardless of its major role in tree productivity and soil fertility, the application of nitrogen fertilisers may induce a series of negative consequences from the microbiological, economic and ecological aspects. The intensity of the stated processes is governed by the type and rate of applied fertilisers. Therefore, a basic task of science is to enable intensive agricultural production without causing any environmental and human health damages, and placing special focus on the proper use of different fertilisation systems.

* For correspondence.

Biological productivity of different soil types is determined by the composition, the structure and the activity of the soil biological component. Microorganisms population, as major soil component, is very significant indicator of soil fertility, i.e. the process of soil degradation. Soil microbiological activity should be studied for deeper knowledge of the substance of soil fertility and development of the measures that will contribute to the most efficient mineral fertiliser utilisation. Microbial population density and enzyme activity of microorganisms may be highly reliable practical indicators which can suggest the answer to the question concerning conditions that favour plant production. It was with respect to this that the aim of these investigations – to examine the effect of different mineral fertiliser rates on the number of different groups of microorganisms and productivity of alluvium under plum – was determined.

EXPERIMENTAL

The investigations were carried out in the 2003–2005 period in a trial orchard of Fruit Research Institute in Cacak and at the Microbiology Department of the Faculty of Agronomy in Cacak.

Mineral fertiliser NPK 8:16:24 + 3% MgO, product of 'Zorka' Sabac, was applied in autumn 2002 at the following rates (kg ha^{-1}): 400 (variant N_1); 600 (variant N_2); 800 (variant N_3) and 1000 (variant N_4). The soil untreated with the stated nutritives served as the control. In the ensuing 2 years, residual effects of applied fertilisation were tested, and the plum orchard was not subsequently fertilised.

The trial field was 68 m^2 large, planted with plum cultivar Stanlay in random block design, in 3 replications. The soil was alluvial, of slightly acid reaction (pH value in 1n KCl = 5.9), moderately supplied with organic matter (2.65% humus) and well supplied with plant-available P and K (AL-method: mg 100 g^{-1} of soil = 15.0 P_2O_5 and 20.4 K_2O).

Common cropping practices were used during the growing season. The soil samples were taken in the terms as follows: May 12, September 17 and November 11 of each testing year, for the 1st, the 2nd and the 3rd sampling, respectively.

The number of microorganisms was determined by the indirect dilution method on adequate substrata: total number of microorganisms on total number of microorganisms medium – 7 days (28°C); the number of ammonifiers on MPA medium – 3 days (28°C); the number of fungi on Chapek medium – 5 days (28°C).

The proteinase activity (number of gelatinolytic units per 1.0 g air-dry soil) was determined by the method described by Romeiko[3].The data obtained were processed by the analysis of variance and the significance of differences between the variants examined was analysed by the LSD-test.

RESULTS AND DISCUSSION

An analysis of the results obtained determined that all variants with higher fertiliser rates used had an inhibitory effect on the growth of all examined groups of microorganisms except soil fungi (Table 1).

Table 1. Average number of fungi (10^5/1.0 g of absolutely dry soil) in soil as influenced by applied fertiliser rates (A), periods of sampling (B) and years of research (C)

Fertiliser (A)			N_1	N_2	N_3	N_4	Ø	$\overline{X}$
Year (C)	2003 period (B)	I	4.500	5.335	8.165	9.670	3.835	6.301
		II	13.500	15.835	22.335	23.330	8.831	16.766
		III	6.170	7.835	11.170	23.555	3.500	10.446
	$\overline{X}$		8.057	9.668	13.890	18.852	5.389	11.171
	2004 period (B)	I	2.830	3.670	5.335	6.165	2.335	4.067
		II	8.500	11.665	13.000	14.500	7.000	10.933
		III	5.333	7.665	9.330	12.830	3.665	7.765
	$\overline{X}$		5.554	7.667	9.222	11.165	4.333	7.588
	2005 period (B)	I	2.670	4.335	4.665	5.000	2.330	3.800
		II	9.330	12.500	14.335	16.330	6.835	11.866
		III	4.000	4.165	5.000	6.335	3.335	4.567
	$\overline{X}$		5.333	7.000	8.000	9.222	4.167	6.744
$\overline{X}$		I	3.333	4.447	6.055	6.945	2.833	4.723
		II	10.443	13.333	16.557	18.053	7.555	13.188
		III	5.168	6.555	8.500	14.240	3.500	7.593
	$\overline{X}$		6.315	8.112	10.371	13.079	4.630	8.501

LSD

LSD	A	B	C	A×B	A×C	B×C	A×B×C
0.05	1.831	1.418	1.418	3.171	3.171	2.456	5.492
0.01	2.425	1.878	1.878	4.200	4.200	3.254	7.275

Generally speaking, all fertiliser rates (factor A) had a stimulating effect on the development of soil fungi, which was particularly evidenced in high-rate variants over all phases of the growing season in this fruit species. This trend was notaby observed in the N_4 variant applied in mid- and final phases of the growing season, the growing of which was favoured by higher temperatures and moisture rates alike (Table 2). The incorporation of higher rates of mineral fertilisers into soil, acid ones in particular, and the long-term usage thereof is depressing for the majority of microorganisms[4,5]. However, soil fungi which exhibit a steady enzyme system

which enables them to inactivate even heavily degradable chemical compounds fare well even under such conditions[6], and this stimulating effect of higher mineral fertiliser rates is therefore anticipated. With regard to the predominance of fungi in acid soils, it is also suggested that their population number rises with a more intensive application of the stated fertilisers. A large number of authors addressing this issue account for this rise in population density and activity of the majority of microorganisms in soil by limiting of the C:N relation and the intensification of the mineralising processes therein, as well as by the re-distribution within the complex of microbial cenoses in favour of soil fungi[7–10].

Table 2. Weather characteristics (Cacak Weather bureau) and long-terms means (LTM)

Period	Precipitation and mean air-temperatures in Cacak*							Total precip-itaion	Mean air-temper-ature
	unit	May	June	July	Aug.	Sept.	Oct.	Nov.		
2003	mm	62	51	69	6	34	77	27	326	
	°C	19.8	25.1	24.2	26.4	17.3	10.2	8.9		18.8
2004	mm	66	121	82	58	35	27	98	487	
	°C	16	21.7	23.5	22.8	18.1	14.3	6.2		17.5
2005	mm	72	84	100	66	91	23	83	519	
	°C	17.2	21	23.7	20.3	18.2	11.8	5.2		16.8
LTM	mm	89	98	76	60	56	48	59	486	
(1965–1994)	°C	16.2	19.5	20.9	20.5	16.9	11.8	5.8		15.9

* Air-distance from the experiment about 5 km in E direction.

The using of the highest NPK-rate had produced the highest inhibitory effect on total number of microorganisms and the number of ammonifiers (Tables 3 and 4). The tendency resulted from a change in the established soil biological equilibrium following the incorporation of high mineral fertiliser rates in the soil and activation of non-specific toxinogenic groups of microorganisms.

As concerns the sampling period (factor B), the greatest number of studied group of microorganisms was reported over the 2nd sampling. As the character of fertiliser activity on the development of microorganisms is directly dependent on cumulative effect of different ecological factors (moisture and temperature of soil, level of cultivation of soil, and grown plant species[11]), the results arising from the experiment had been anticipated. The 2nd sampling (September 17) was characterised by more regular distribution of rainfall and temperatures (Table 2), which resulted in higher number of the microorganisms over this period. The variants with lower fertiliser rates had a stimulatory effect on proportionally equal level on these groups of microorganisms in all 3 research periods. Increased number of the examined groups of microorganisms triggered by the effect of mineral fertilisers

could be attributed to soil enrichment with mineral elements required for the development of these microorganisms group[12]. These fertilisers affected the mobility of soil organic matter which was subjected to hydrolysis immediately after their incorporation into the soil, prior to activation of biological processes in the soil.

Table 3. Average number of the total number of microorganisms (10^6/1.0 g of absolutely dry soil) in soil as influenced by applied fertiliser rates (A), periods of sampling (B) and years of research (C)

Fertiliser (A)			N_1	N_2	N_3	N_4	Ø	$\overline{X}$
Year (C)	2003 period (B)	I	115.665	149.170	61.830	32.830	82.330	88.365
		II	110.835	292.500	53.835	36.335	107.500	120.201
		III	76.665	94.500	32.500	21.165	53.665	55.699
	$\overline{X}$		101.055	178.723	49.388	30.110	81.165	88.088
	2004 period (B)	I	28.833	27.665	12.665	7.165	19.335	19.133
		II	24.500	28.000	21.835	19.500	22.335	23.234
		III	20.835	25.830	15.500	13.330	16.000	18.299
	$\overline{X}$		24.723	27.165	16.667	13.332	19.223	20.222
	2005 period (B)	I	28.835	49.165	17.665	10.835	19.330	25.166
		II	22.333	35.170	20.500	13.000	18.835	21.968
		III	28.165	35.330	19.335	8.665	19.830	22.265
	$\overline{X}$		26.444	39.888	19.167	10.833	19.332	23.133
$\overline{X}$		I	57.778	75.333	30.720	16.943	40.332	44.221
		II	52.556	118.557	32.057	22.945	49.557	55.134
		III	41.888	51.887	22.445	14.387	29.832	32.088
$\overline{X}$			50.741	81.926	28.407	18.092	39.907	43.814

LSD							
LSD	A	B	C	A×B	A×C	B×C	A×B×C
0.05	4.936	3.824	3.824	8.550	8.550	6.623	14.809

Table 4. Average number of ammonifiers (10^6/1.0 g of absolutely dry soil) in soil as influenced by applied fertiliser rates (A), periods of sampling (B) and years of research (C)

Fertiliser (A)			N_1	N_2	N_3	N_4	Ø	$\overline{X}$
Year	2003 period	I	66.500	68.665	16.165	6.665	40.665	39.732
(C)	(B)	II	67.830	140.835	43.335	39.993	53.835	69.166
		III	25.053	47.000	10.835	8.335	28.835	24.012
	$\overline{X}$		53.128	85.500	23.445	18.331	41.112	44.303
	2004 period	I	16.830	18.000	15.830	14.500	16.335	16.299
	(B)	II	14.830	24.665	7.335	6.330	8.835	12.399
		III	13.335	14.500	11.000	8.330	12.335	11.900
	$\overline{X}$		14.998	19.055	11.388	9.720	12.502	13.533
	2005 period	I	21.315	33.330	12.000	14.833	17.170	19.730
	(B)	II	24.330	39.330	11.835	5.830	14.330	19.131
		III	17.835	25.335	9.830	7.170	12.665	14.567
	$\overline{X}$		21.160	32.665	11.222	9.278	14.722	17.809
	$\overline{X}$	I	34.882	39.998	14.665	11.999	24.723	25.254
		II	35.663	68.277	20.835	17.384	25.667	33.565
		III	18.741	28.945	10.555	7.945	17.945	16.826
	$\overline{X}$		29.762	45.740	15.352	12.443	22.778	25.215

LSD

LSD	A	B	C	A×B	A×C	B×C	A×B×C
0.05	4.784	3.706	3.706	8.287	8.287	6.419	14.353
0.01	6.338	4.909	4.909	10.977	10.977	8.503	18.013

NPK-fertilisation (the factor A) significantly influenced proteinase activity in the soil (Table 5). Regarding the term of soil sampling influences (the factor B), the highest proteinase activity was found in the 2nd term of sampling (for 28% higher compared to the control), while in the 3rd term of sampling it was on the same level as the control (non-significant difference). The year of testing (factor C) is also an important factor of proteinase activity because in the 3rd year of study (2005) it was with 28% (2003) and 26% (2004) lower, while differences between 2003 and 2004 were non-significant. Proteinase activity differences among terms of soil sampling were non-significant for the control, but for the applied fertilisation it was significantly higher in the 2nd sampling compared both with the 1st and 3rd terms of the treatments 600, 800 and 1000 kg ha^{-1} of fertiliser. A decline in the activity in final vegetative development stages of plum resulted from decreased soil moisture, which significantly induced a reduction in the number of

microorganisms accountable for the production of many enzymes including the proteolytic ones[13].

Table 5. Average of proteinase activity (number of gelatinolytic units/1.0 g air-dry soil) in soil as influenced by applied fertiliser rates (A), periods of sampling (B) and years of research (C)

Fertiliser (A)			N_1	N_2	N_3	N_4	Ø	$\overline{X}$
Year (C)	2003	period (B) I	18.959	19.792	8.763	4.167	16.263	13.589
		II	21.250	24.584	16.042	9.375	19.167	18.083
		III	21.665	22.708	7.917	6.667	12.292	14.250
		$\overline{X}$	20.625	22.361	10.907	6.736	15.907	15.307
	2004	period (B) I	13.750	19.375	7.292	6.458	10.834	11.542
		II	23.959	32.500	13.125	8.125	18.124	19.166
		III	20.000	21.457	8.125	7.292	13.333	14.041
		$\overline{X}$	19.236	24.444	9.514	7.292	14.097	14.917
	2005	period (B) I	15.000	16.667	8.750	5.625	12.917	11.792
		II	19.833	25.084	12.750	6.958	14.209	15.767
		III	10.834	13.957	7.706	4.167	8.959	9.124
		$\overline{X}$	15.222	18.569	9.735	5.583	12.028	12.228
	$\overline{X}$	I	15.903	18.611	8.268	5.416	13.338	12.307
		II	21.681	27.389	13.972	8.153	17.166	17.672
		III	17.500	19.374	7.916	6.042	11.528	12.472
	$\overline{X}$		18.361	21.791	10.052	6.537	14.011	14.150

LSD							
LSD	A	B	C	A×B	A×C	B×C	A×B×C
0.05	1.298	1.005	1.005	2.248	2.248	1.741	3.893
0.01	1.719	1.331	1.331	2.977	2.977	2.306	5.157

The plum yield results indicate a pronounced stimulatory effect of the N_2 rate, too (Table 6) (Ref. 14), the reported yield being 24.172 t ha^{-1}.

Table 6. Effect of fertilisers applied on the plum yield

Variants	Plum yield (kg ha^{-1})
N_1	18980
N_2	24172
N_3	20833
N_4	20125
Ø	23852
$\overline{X}$	21592

CONCLUSIONS

Based on the research results on the effect of mineral fertilisers on the number of different groups of microorganisms and on proteinase activity of soil under plum orchard the following can be concluded:

- The number of the studied group of microorganisms and proteinase activity depended on the fertiliser rate, period of sampling and year of study;
- High fertiliser rates caused decline in total number of microorganisms, the number of ammonifiers and on the proteinase activity;
- The most inhibitory effect was reported in the variant with highest nitrogen application;
- The application of lower and moderate fertiliser rates gave rise to the greater number of the studied groups of microorganisms;
- The most stimulating effect was reported in the variant with medium nitrogen application;
- The applied fertilisers brought about increase in the number of fungi, particularly in the variants that included highest nitrogen content;
- The effect of fertiliser applicaton was most pronounced over the 2nd period of sampling and in the 1st year of study (2003);
- The most significant rise in plum yield was obtained with the application of 600 kg ha^{-1} mineral fertiliser.

Taking into account biological properties of soil, plum production, economic and environmental indicators of the results of the stated treatments, the rate of 600 kg ha^{-1} can be recommended for growing plum under the stated environmental conditions.

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