EFFECT OF FERTILIZATION ON MICROBIAL ABUNDANCE IN SUGARBEET RHIZOSPHERE

EFEKAT ĐUBRENJA NA BROJNOST MIKROORGANIZAMA U RIZOSFERI ŠEĆERNE REPE

Nastasija MRKOVAČKI, Jelena MARINKOVIĆ, N. ČAČIĆ

Institute of Field and Vegetable Crops, Novi Sad, Serbia
Corresponding author: Nastasija MRKOVAČKI, e-mail nastasija@ifvcns.ns.ac.yu

Abstract: *Microorganisms are a heterogeneous group of organisms whose enzymatic systems take 60-90% of the total metabolic activity in soil. Dominance of certain groups of microorganisms affects the processes of soil synthesis and decomposition and it determines the quality of soil and its applicability for the production of safe food. The nonsymbiotic nitrogen fixer Azotobacter is an obligate aerobe living in the soil, the rhizosphere and the plant root. The ability of various strains to fix atmospheric N positively affects plant growth and yields. In this study we have determined the abundance of azotobacter, fungi, actinomycetes and the total number of microorganizms in sugarbeet rhizosphere. Simultaneously we studied the effect of application of nitrogen fertilizer, manure and harvest residues. Samples of rhizosphere soil were taken three times in the course of growing season (May, July, September) the experiment included two variants (inoculated with azotobacter and noninoculated) in four fertilization levels (nonfertilized control, 50, 100, 150 and 200 kg N/ha), in five replication. The results of the study for all four fertilization types and all four levels of added N showed that azotobacter abundance was higher in inoculated treatments than uninoculated ones. The highest percentage increases of azotobacter numbers, total microbial abundance and number of fungi and actinomycetes were recorded in the inoculated treatments with barnyard manure, NPK fertilizer, and barnyard manure plus harvest residues, respectively. In inoculated and uninoculated treatments alike, the highest azotobacter abundance was found in the treatment with no N added, while the lowest was recorded in the treatment with 200 kg N/ha added. The highest total microbial abundance was obtained with 100 kg N/ha in inoculated treatments and 200 kg N/ha in uninoculated ones.*

Abstract: *Mikroorganizmi su veoma heterogena grupa organizama i sa svojim enzimatskim sistemima učestvuju sa 60-90% u celokupnoj metaboličkoj aktivnosti zemljišta. Dominantnost pojedinih grupa mikroorganizama usmerava procese ka sintezi ili razgradnji i time određuje kvalitet zemljišta za proizvodnju zdravstveno ispravne hrane. Slobodni azotofiksator Azotobacter je obligatni aerob koji živi u zemljištu, rizosferi i na korenu biljke. Sposobnost različitih sojeva da fiksiraju atmosferski azot pozitivno deluje na rast biljke i prinos. U ovom radu odredili smo brojnost azotobaktera, gljiva, aktinomiceta i ukupan broj mikroorganizama u rizosferi šećerne repe. Uporedo smo proučavali i efekat primene azotnog đubriva, stajnjaka i žetvenih ostataka. Uzorci rizosfernog zemlišta uzimani su tri puta u toku vegetacije (maj, juli, septembar) a eksperiment je uključio i dve varijante (inokuliasno sa azotobakterom i neinokulisano) u četiri nivoa đubrenja (neđubrena kontrola, 50, 100, 150 i 200 kg N/ha), u pet ponavljanja. Rezultati ispitivanja, kod sve 4 vrste đubrenja i sva četiri nivoa dodataka azota, pokazali su da je na inokulisanim varijantama broj azotobaktera bio veći nego na neinkulisanim. Najveći procenat povećanja broja azotobaktera na inokulisnim varijantama dobijen je na varijanti sa dodatkom stajnjaka, ukupnog broja mikroorganizama na varijanti sa NPK, dok je najveći procenat povećanja broja gljiva i aktinomiceta dobijen dodatkom stajnjaka i žetvenih ostataka. Najveća brojnost azotobaktera dobijena je na varijanti bez dodatka azota, a najmanji sa 200 kg N/ha, i kod inokulisane i neinokulisane varijante. Najveći ukupan broj mikroorganizama dobijen je sa 100 kg N/ha na inokulisanoj varijanti, a sa 200 kg N/ha na neinokulisanoj*

Key words: *soil, microorganisms, rhizosphere, sugarbeet*
Ključne reči: *zemljište, mikroorganizmi, rizosfera, šećerna repa*

INTRODUCTION

Soil fertility is a combination of physical, chemical and biological soil properties, and the most important role in the development and maintenance of these properties is played by microorganisms (Lee, 1994). Microorganisms are a heterogeneous group of organisms whose enzymatic systems take 60-90% of the total metabolic activity in soil (Taylor, 2001). Dominance of certain groups of microorganisms affects the processes of soil synthesis and decomposition and it determines the quality of soil and its applicability for the production of safe food.

One approach to improving the nitrogen economy of crops has been to apply diazotrophic bacteria to non-leguminous crops in the expectation that they would fix atmospheric nitrogen and so provide combined nitrogen to the plant for enhanced crop production. (Sloger and Van Berkum, 1992; Sturz *et al*., 2000; Mrkovački and Milić, 2001). The nonsymbiotic nitrogen fixer Azotobacter is an obligate aerobe living in the soil, the rhizosphere and the plant root. The ability of various strains to fix atmospheric N positively affects plant growth and yields. Sugar beet requires large amounts of nitrogen fertilizers (Božić *et al*., 1997), so inoculation with *Azotobacter* may help reduce mineral fertilizer rates used for this crop.

Bacterization introduces into the soil live cells of nitrogen-fixing bacteria (both symbiotic and free-living ones), which, besides fixing nitrogen, also produce various polysaccharides, vitamins and growth substances and thus create favourable living conditions for other microorganisms (Holbs, 1985).

Therefore, the objective of this study was to determine the abundance of azotobacter, fungi, actinomycetes and the total number of microorganizms in inoculated and noninoculated sugarbeet rhizosphere.

MATERIAL AND METHOD

A field trail was established on a chernozem soil at experiment field of Institute of Field and Vegetable Crops at Rimski Šančevi. The experimental object was the sugarbeet hybrid variety Sara developed at the Institute. The trail included two variants, inoculated and non-inoculated. Inoculation was performed with a mixture of *Azotbacter* strains, with the concentration of 10^9 per ml, incorporated into the soil before planting. The trail was established in a block design with five replications, four levels of N fertilization (50, 100, 150 and 200 kg N/ha) and the nonfertilized control. Simultaneously we studied the effect of application of nitrogen fertilizer, manure and harvest residues. Samples of rhizosphere soil were taken three times in the course of growing season (May, July, and September). Besides the effects of different levels of mineral N, we also examined the effect of application of manure, harvest residues and their combinations.

Total number of microorganisms was determined in soil agar (at the dilution of 10^6). Fungi were determined on Chapek agar, actinomycetes on a synthetic agar (at the dilution of 10^4), and azotobacters on Fiodor substrate (at the dilution of 10^2).

RESULTS AND DISCUSSION

The results of the study for all four fertilization types and all four levels of added N showed that azotobacter abundance was higher in inoculated treatments than uninoculated ones. Inoculation of soil in sugarbeet field with sufficient inoculum consisting of selected, highly effective *Azotobacter chroococcum* strains increased the population size of azotobacter in the rhizosphere as a result of bacterial adaptation to the environment and ecological conditions (Arte and Shende, 1981; Mrkovački and Mezei, 2003).

The highest azotobacter abundance was obtained in the variant without N and the smallest with 200 kg N/ha (tab. 1). These results are in agreement with our previous results (Mrkovački *et al*., 2003; Mrkovački *et al*., 2006).

The highest number of fungi and total microbial abundance were obtained with 100 kg N/ha in inoculated treatments. In same treatments, the smallest number of fungi was recorded with 150 kg N/ha, and the lowest total microbial abundance was in the variant without N, which is not in agreement with previous results (Mrkovački *et al*., 2006). The smallest number of these two microbial groups were obtained in the variant without N in uninoculated treatments, and the highest with 200 kg N/ha (tab. 1).

The results of this experiment showed that the largest number of actinomycetes, in both variants (inoculated and noninoculated), were obtained with 50 kg N/ha, and the lowest in the variant with 100 kg N/ha, which is contrary to results of Sarić (1972) in which the highest number of actinomycetes was in variants with 100 kg N/ha (tab. 1).

Table 1

Abundance of microbes in sugarbeet rhizosphere depending on inoculation and N fertilization

Microbial group	Inoculation with *Azotobacter*	Ø	kg N/ha				
			50	100	150	200	average
Azotobacter	- A	95,24	90,09	63,95	41,70	37,11	65,62
	+ A	170,17	123,96	153,89	70,80	64,04	116,57
Fungi	- A	11,25	19,64	16,87	18,78	22,69	17,85
	+ A	22,23	24,39	24,84	13,66	17,16	20,46
Actinomycetes	- A	21,14	25,00	17,02	22,23	21,67	21,41
	+ A	26,26	30,32	21,57	25,23	27,09	26,09
Total number	- A	126,65	140,82	155,83	160,62	187,24	154,23
	+ A	201,57	211,11	241,78	218,85	217,62	218,19

In this study, inoculation tended to increase the abundance of all groups of microorganisms in sugarbeet rhizosphere. The average increases of azotobacters were 77.64% and total microbial abundance 41.47 %.

Table 2 shows that in the noninoculated variant the largest number of fungi and total microbial abundance were registered in the variant with NPK plus manure, azotobacter in the variant with NPK plus harvest residues and actinomycetes in the variant with NPK fertilization alone.

Table 2

Abundance of microbes in sugarbeet rhizosphere depending on inoculation and kind of fertilization

Microbial group	Inoculation with *Azotobacter*	NPK	NPK+ manure	NPK+ harvest residues	NPK+ h.residues+ manure	average
Azotobacter	- A	65.62	60.99	111.33	90.81	82.19
	+ A	116.57	110.44	150.65	126.75	126.10
Fungi	- A	17.85	24.70	20.70	17.31	20.14
	+ A	20.46	21.21	22.63	22.57	21.72
Actinomycetes	- A	21.41	15.78	20.34	18.51	19.01
	+ A	26.09	17.73	21.76	22.89	22.12
Total number	- A	154.23	173.10	159.04	160.67	161.76
	+ A	218.19	218.66	177.64	193.53	202.01

In the inoculated variant, largest number of azotobacters and fungi were found with NPK plus harvest residues. The highest number of actinomycetes was in the variant with NPK fertilization alone, while the largest number of total microbial abundance was with NPK plus manure and NPK alone (tab. 2). That is in agreement with our previous results (Mrkovački *et al*., 2006).

On average for the four kinds of fertilization, inoculation increased the number of azotobacter and total number of microorganisms by 53.42 % and 24.88% respectively.

CONCLUSIONS

The inoculation with *Azotobacter* increased the biological value of sugarbeet rhizosphere.

The largest number of azotobacter was obtained in the variant without N, the smallest with 200 kg N/ha.

The highest total microbial abundance was obtained with 100 kg N/ha in inoculated treatments and 200 kg N/ha in uninoculated ones.

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