

Thesis of Doctoral (PhD) Dissertation

**ARTIFICIAL FERTILIZER AND BIO PREPARATIONS AFFECTING
THE MICROBIOLOGICAL ACTIVITY AND FERTILITY OF SOIL**

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1. INTRODUCTION

Using the environmentally friendly farming system in recent decades, the environmental loads decreased in the soil, the quality of product, the stability of yield, and sustainable production was aimed with the cropping system. Reduction in the pressure on the environment by using energy-efficient materials and technologies to the alleviation of the economic harms can be achieved due to global climate change. The sustainability is ensured with the suitable proportion of the ecological, biological and agricultural engineering factors (VÁRALLYAY, 1997; RUZSÁNYI-PEPÓ, 1999; PEPÓ, 2002).

Under the sustainable nutrient management the coordination of production and environmental demands, the minimal environmental impacts and adaptation to the ecological terms must be primarily considered. In the case of site-specific fertilization, the application of the optimal dose and the correct selection of applied fertilizers can help to prevent the undesirable over-fertilization. Additionally the rational fertilizing adapted to the lack of nutrient of plants can not be neglected. Soil degradation resulting from increasing environmental pressure draws attention to the efficient use of fertilizers (NÉMETH T., 1977; KÁDÁR I., 1992; NÉMETH T., 1995; ÁRENDÁS et al., 1999).

When nutrient supply is not reasonable, a lot of negative effects e.g. reduction in crop yield, deterioration of nutritional value, worsening environmental pollution (water and soil contamination) are resulted. The use of suitable quantity and quality of artificial fertilizers can increased the nutrient stocks of soil what can achieve higher average of yield. Professionally executed fertilization preserving on soil fertility were maintained healthier life in soil (KÁDÁR, 2011).

The mentioned negative effects caused by unilateral, insufficient nutrient ratio and quantity and quality of fertilizers, as well as the increasingly emphasis on economical aspects, the application of different microbial products has been spreading in agricultural cultivation nowadays. The bio-preparations containing microorganisms which have different activity but in this view, there are many false known. Comparative examination is necessary to compare the effects of some commercially available preparations and artificial fertilizers on the yield growth and soil parameters.

AIM AND SCOPE

In recent years an increasing use of microbial inoculants has been applied in agricultural production to improve the soil fertility, to stimulate the soil life and increase crop yields. Short scientific data were evolved about the complex effects of bio preparations in soil-plant system. However, there are few scientific data about the complex effects of these microbial preparations in the soil-plant system.

Our aims were the following:

- To examine in pot experiment the effects of bacterial preparations (BactoFil A10, EM-1 and Microbion UNC) alone and compared the effects to control, NPK fertilizer and wheat straw basic treatments, as well as combined these preparations with the basic treatments. We established the effectiveness of the combinations of the preparations and their impacts on the soil parameters and on the biomass of test plant (*Lolium perenne*, L.).

- To study in pot experiment the application of different doses (simple and double) of artificial fertilizer and root vitalizing (Amykor) preparation and the doses combined with NPK fertilizer. Our objective was to establish the most effective treatments, as well as finding the optimal dose of root vitalizing and combinations on the bases of examined soil parameters and the biomass of test plant (*Allium cepa*, L.).

- To establish in small plot experiment the effect of different doses (simple and triple dose) and the various application methods of Amykor (with sown simultaneously, and worked in to the 0-20cm of soil layer), and the favourite impacts on the growth of maize (*Zea mays*, L.).

- To investigate and get complex results about the effect of different microbial preparations on the chemical, microbiological parameters of soil and on the biological parameters of test pants.

- To examine with laboratory methods some soil chemical parameters so the soil pH, the NO_3^- N, the AL-soluble phosphorus and potassium content of soil.

- In order to tracking the population dynamic of living microorganisms of soil, the total number of bacteria, the number of aerobic cellulose decomposing and nitrifying bacteria and the amount of microscopic fungi were determined.

- In order to follow the microbiological activity of soils, the soil respiration, the amount of microbial biomass carbon, the rate of nitrate-exploration, and urease, saccharase, catalase, phosphatase and dehydrogenase enzymes' activities were determined.

- To measure the biomass of ryegrass and onion in pots experiment and, in field experiment the LAI index, and yield parameters of maize (the length of cobs, grain/cob%,

weight of thousand kernels, the amount of yield, the moisture content of yield and hectolitre weight of yield at the end of the experiment) were examined.

2. MATERIALS AND METHODS

2.1. Examination of the effects of artificial and bacterial fertilizers in pot experiment

2.1.1. Setting up experiments

At the greenhouse of the Institute of Agricultural Chemistry and Soil Sciences (University of Debrecen) pot experiments were carried out to investigate the effects of three different bacterial fertilizers between 2010 and 2013. The experiment was set on calcareous chernozem soil every year, from the surrounding area of Debrecen (Látókép). The applied test plant in experiment was ryegrass (*Lolium perenne*, L.) every year.

2.1.2. Description of the treatments applied

The treatments used in examination of bacterial fertilizer pot experiment are illustrated in *Table 1*.

Table 1. The applied treatments in the artificial and bacterial fertilizer experiment (2010-2013)

Treatment	Basic treatments	Bacterial fertilizer
1	Control (Untreated)	0
2	NPK fertilizer*	0
3	Wheat straw	0
4	0	BactoFil A10
5	NPK fertilizer*	BactoFil A10
6	Wheat straw	BactoFil A10
7	0	EM-1
8	NPK fertilizer*	EM-1
9	Wheat straw	EM-1
10	0	Microbion UNC
11	NPK fertilizer*	Microbion UNC
12	Wheat straw	Microbion UNC

*NPK: N=NH₄NO₃, P=KH₂PO₄, K=K₂SO₄ + KH₂PO₄

In the experiment control, NPK fertilizer and wheat straw treatments were applied, which were supplemented with three different microbial preparations (BactoFil A10, EM-1 and Microbion UNC). Weight supplement irrigation of the vessels was performed every day for 60% of field water capacity. The experiment was eliminated from the start of germination eight week. The 12 treatments in the experiment applied were arranged in a random block design with three replications, gave totally 36 pots.

In fertilization was dole out nitrogen as NH_4NO_3 ($0.2857 \text{ g pot}^{-1}$), phosphorus as KH_2PO_4 ($0.1915 \text{ g pot}^{-1}$), potassium as KH_2PO_4 ($0.1915 \text{ g pot}^{-1}$) and K_2SO_4 20 cm^3 solution per pot. In the straw treatment (3 g per pot) 7 t per he wheat straw pot^{-1} were stirred in. The bacterial fertilizers 20 cm^3 from BactoFil A10 and 15 cm^3 from EM-1 per pot were dole out as solutions. The applied amount of solid bacterial fertilizer Microbion UNC was 0.01g per pot. The applied quantity of bacterial fertilizers was the double of the recommended doses for field application.

2.1.3. Description of the preparations applied

In the experiment microbiological preparations BactoFil A10, EM-1 and Microbion UNC were applied, which most important characteristics are described *in Table 2*.

Table 2. The most important microbes of the applied bacterial preparations of the artificial-bacterial fertilizer experiment

Microbe	BactoFil A10	EM-1	Microbion UNC
<i>Azospirillum sp.</i>	+	d.n.a.	+
<i>Azotobacter vinelandii</i>	+	d.n.a.	+
<i>Bacillus megatherium</i>	+	d.n.a.	+
<i>Bacillus sp.</i>	+	d.n.a.	+
<i>Streptomyces albus</i>	+	+	+
<i>Saccharomyces cerevisiae</i>	-	+	+
Other microbe	<i>Pseudomonas fluorescens</i>	<i>Rhodospseudomonas sp.</i> , <i>Lactobacillus sp.</i> , <i>Propinebacterium sp.</i> , <i>Streptococcus sp.</i> , <i>Aspergillus sp.</i> , <i>Mucor sp.</i> , <i>Candida sp.</i>	<i>Clostridium sp.</i> , <i>Rhodobacter sp.</i> , <i>Lactobacillus sp.</i> , <i>Trichoderma sp.</i> ,
d.n.a.- data no available		<i>Stb.</i>	

- Bacterial fertilizer **BactoFil® A10** used in production of monocotyledons. The recommended dose of BactoFil A10 for monocotyledon crops in field and horticultural systems is $1-1.5 \text{ litre he}^{-1}$ with $200-400 \text{ litre he}^{-1}$ irrigation water and above 5°C soil temperature.

- The **EM-1** microbiological preparation contains more than 80 strains and the total number of cells more than $5 \cdot 10^8 \text{ piece cm}^{-3}$. The recommended dose of the soluble preparation is $30-50 \text{ litre per hectare}$ applied in field and horticultural crop systems (worked before and in time of sowing).

- The **Microbion UNC** is a solid microbiological preparation. The recommended dose of the preparation is 1.5-3 kg per hectare applied in field and horticultural crop systems.

2.1.4. Sampling: soil and plant sampling

The plant samples were cut off above 2 cm of the soil surface and collected for each pot after 4 and 8 weeks. After 8 weeks were collected soil samples for each pot, too. After cutting plant samples the vessels were vacated and carefully homogenised the soil samples after removal the root system, dried, and finally sieved through in 2 mm sieve. The contents of each vessel were transported to the laboratory. To the soil microbiological examinations the samples were stored in refrigerator on 5°C. After soil microbiological investigations the soil samples were dried to air-dry condition, than sieved anew and finally homogenized. The main soil parameters are illustrated in *Table 3*.

Table 3. The main parameters of soil applied in artificial and bacterial fertilizer experiments (2010-2013)

Parameters	Years of the experiment			
	2010	2011	2012	2013
K_A	38	37,5	40	40
Li%	51	50	45	50
Hu%	2,4	2,8	2,8	3,0
pH _{H2O}	6,6	6,6	7,2	6,9
pH _{KCl}	5,5	5,5	6,2	6,1
AL-P ₂ O ₅ (mg kg ⁻¹)	140	312	150	106
AL-K ₂ O (mg kg ⁻¹)	316	360	240	200

2.1.5. Measurements of the laboratory examinations of the soil samples

2.1.5.1 Soil physical examinations

Of the soil physical examinations was measured the *soil moisture content* expressed in percentage by mass at the end of the experiment (RAJKAI, 1993). The amount of *clay and silt content (Li%)*, as well as *the plasticity index according to Arany (K_A)* was determined, concluded the soil physical condition and texture (VÁRALLYAY, 1993).

2.1.5.2. Soil chemical examinations

The measured soil chemical examinations of the air-dried soil samples were measured the soil:water and soil:1M KCl *pH* of suspension in dilution 1:2.5 (BUZÁS, 1988). The *humus content of soil (Hu%)* was measured by colorimetric method of BUZÁS (1988). The NO₃⁻-N content of soil was established by FELFÖLDY's (1987) sodium-

salicylate method, and in maturation laboratory experiment the nitrate-exploration of soil was measured in thermostat at 37°C. The *ammonium-lactate (AL) soluble phosphorus and potassium content of soil* was determined by MSZ 20135:1999.

2.1.5.3. Soil microbiological examinations

The *total number of bacteria* (on Bouillon soup medium) and *the amount of microscopic fungi* (on peptone-glucose medium) of the soil microscopic parameters were determined by the plate dilution method of SZEGI (1979). According to POCHON-TARDIEUX (1962) were established *the amount of aerobe cellulose decomposing and nitrifying bacteria*. The *rate of soil respiration* with NaOH trapping was measured after 10 days in incubation examination (WITKAMP, 1966). The *microbial biomass carbon content of soil* (CFI) was determined by the chloroform fumigation incubation method of JENKINSON-POWLSON (1976).

2.1.5.4. Enzyme activity examinations

Characterizing the biological processes can also help to investigate the enzyme activities of soil. The urease and saccharase enzyme activities of soil samples were determined by the method of SZEGI (1979), the catalase enzyme activity of soil was measured by KUPREVICS-SCSERBAKOVA (1956), by the method of KRÁMER-ERDEINÉ (1958) was established the phosphatase enzyme activity of soil, the dehydrogenase enzyme activity of soil was measured by MERSI-SCHINNER (1991).

2.1.6. Measuring plant biomass

2.1.6.1. Green weight and dry mass of plant examinations

The green mass of ryegrass was determined. The biomass of ryegrass was dried on air and on 50°C in drying cabinet to the mass stability. Knowing the average values of dried biomass were calculated the ratio (%) of dry matter and moisture content (LOCH et al., 2003).

2.2. Examination of the effects of artificial fertilizer and mycorrhizal preparation

2.2.1. Assessment in pot experiment

The pot experiment was performed at the greenhouse of the Institute of Agricultural Chemistry and Soil Sciences (University of Debrecen) applied sandy soil from the

surrounding area of Debrecen (Pallag) and test plant of onion (*Allium cepa*, L.). The applied soil was slightly acidic, sandy soil with low supply of nitrogen and phosphorus and medium supply of potassium. The applied treatments are illustrated in *Table 4*.

Table 4. The main soil parameters of applied sandy soil

Parameters	Years of the experiment	
	2012	2013
K _A	<30	<30
Li%	9	10
Hu%	1%	1.2%
pH _{H2O}	6.3	6.0
pH _{KCl}	5.8	5.3
AL-P ₂ O ₅ (mg kg ⁻¹)	160	240
AL-K ₂ O (mg kg ⁻¹)	250	200

The experiments were carried out in the first year on 5th June 2012 and in the next year on 2nd May 2013. The experiments were completed in the first year on 10th July 2012 and in the next year on 11th June 2013. Into the perforated pots 1.35 kg air-dried soil was measured in the first year, in the second one the previous year remained soil were reinvested in accordance with the remaining treatments and completed in the specified weight. In the pots 2-2 pieces of seed onion were sown. Weight supplement irrigation of the vessels was performed every day 70% of field water capacity after the germination of seed onion (the duration of the experiment was 4 week from germination time). The applied treatments of the experiment are illustrated in *Table 5*.

Table 5. Applied treatments in the artificial fertilizer and Amykor pot experiment (2012-2013)

Treatments		
	2012	2013
1	Untreated (control)	Untreated (control)
2	NPK fertilizer	NPK fertilizer
3	Amykor 1 (1x dose)	Amykor 1 (1x dose)
4	NPK fertilizer + Amykor 1 (1x)	NPK fertilizer + Amykor 1 (1x)
5	Amykor 1 (2x dose)	Amykor 1 (2x dose)
6	NPK fertilizer + Amykor 1 (2x)	NPK fertilizer + Amykor 1 (2x)
7	-	Amykor 2 (1x dose)
8	-	NPK fertilizer + Amykor 2 (1x)
9	-	Amykor 2 (2x dose)
10	-	NPK fertilizer + Amykor 2 (2x)

The composition of the NPK fertilizer applied was the same in the artificial and bacterial fertilizer experiment. The fertilizer to the soil thorough mixing was added as solution (20 cm³ of pot⁻¹) paying attention to the homogenization. The dose of Amykor 1 root vitalizator were applied in 10 and 20 cm³ pot⁻¹, namely in the amount of 5-5 and 10-10 cm³ onion⁻¹. The dose of Amykor 2 root vitalizator were applied in 5 and 10 cm³ pot⁻¹,

namely in the amount of 2,5-2,5 and 5-5 cm³ onion⁻¹. The pre-weighed quantities of Amykor were poured into the designed holes of the soil, than placed on the onion seeds and soil, too. The applied quantities of Amykor were the recommended doses for field and horticultural application.

Amykor 1 was an expanded clay mineral (perlite) preparation with mycorrhizal fungal strains (AM-27 *Glomus intraradices*). The macronutrient content of the product are the following: N>0,4 m/m%, P₂O₅>0,3 m/m%, K₂O>0,2 m/m%. The composition of **Amykor 2** is a patent-pending fungal composition consisting essentially of Amykor 1 composition same. The difference between the preparations was the applied doses.

At the end of the experiment (10th July 2012, 11th June 2013) were cut off first the *onion leaves* 1 cm above onion bulbs. The *onion bulbs* were collected warily after sieved all the soil of each pot carried to the roots integrity. Sieved soil samples were taken from each pot, which are individually boxed, further laboratory order 5°C were stored in a refrigerator. The moisture content of the onion leaves after mass stability on 50°C was established. The experiments were arranged in a random block design with three replications. At the end of the experiment the *plasticity index according to Arany (K_A)*, *the silt and clay content (%)*, *the pH (H₂O and 1M KCl)*, *NO₃⁻-N*, *AL-P₂O₅ and AL-K₂O*, *Hu%*, *organic-C content*, *the total number of bacteria*, *the number of cellulose decomposing and nitrifying bacteria*, *the amount of microscopic fungi*, and *urease and phosphatase enzyme activities of the soil* were measured.

2.2.2. Assessment in field experiment

The field experiment was performed at the surrounding area of Debrecen (Látókép) on calcareous chernozem soil and test plant of maize (*Zea mays*, L.) in 2011 and 2012. The soil was slightly acidic, loamy soil with medium supply of nitrogen and phosphorus, and good supply of potassium. The maize was sown in the first year on 28th April 2011 and in the next year on 15th May 2012. The size of the plots is the following: 45.6-45.6 m² (10 m*4.56 m). Six line of maize were planted per parcel (~4.56 m parcel⁻¹). The applied treatments are illustrated in *Table 6*.

Table 6. The applied treatments in the fertilizer and Amykor field experiment (2011, 2012)

<i>Treatments</i>		
	<i>2011*</i>	<i>2012*</i>
1	Untreated/Control	Untreated/Control
2	200 kg he ⁻¹ NPK fertilizer	200 kg he ⁻¹ NPK fertilizer
3	Amykor 200 kg he ⁻¹ (simple dose, 0-20cm-es incorporated to the fertile layer of soil)	-
4	Amykor 600 kg he ⁻¹ (triple dose, 0-20cm-es incorporated to the fertile layer of soil)	-
5	Amykor 150 kg he ⁻¹ (simple dose, with sown simultaneously)	Amykor 150 kg he ⁻¹
6	Amykor 450 kg he ⁻¹ (triple dose, with sown simultaneously)	Amykor 450 kg he ⁻¹

* The plots were delivered uniformly to 200 kg he⁻¹ NH₄NO₃ artificial fertilizer.

In the small pot experiment were applied 6 different treatments in 2011 and 4 treatment in the next year. The experiments were arranged in a random block design with four replications, totally gave 24 (2011) and 16 (2012) parcels. The examined areas (all parcel) were fertilized with 200 kg he⁻¹ fertilizer (68 kg N as NH₄NO₃) before sowing. In the treatments the effects of NPK fertilizer and different doses of Amykor root vitalizer (applied in pot experiment, early demonstrated fungal preparation) were investigated. The applied maize hybrid in both years was *MV Tarján*, a FAO 380, with early maturation time, dent corn.

2-2 times both of year soil samples was collected in the experiment. The first soil sampling was conducted upon the blooming period (65 days after germination) and the second at the end of the experiment (black layer formation time). The collection of soil samples per parcel point-like were made as random selection. 8-8 soil samples were collected per parcels, which were placed in sterile plastic bags and to the laboratory examinations stored in refrigerant at 5°C. The sampling of the first and third series parcel were occurred. *The moisture content (%)*, *the pH (H₂O and KCl)*, *the nitrate-nitrogen content* and *nitrate exploration* of the soil were measured. *The AL-soluble phosphorus and potassium content*, *the total number of bacteria*, *the amount of microscopic fungi*, *the soil respiration*, and *urease and phosphatase enzyme activities* were established.

Every third line of the parcel the sixth to tenth of the plant was examined. The *Leaf Area Index* of maize (LAI m² m⁻²) was determined, in time of blooming. The experiments at “the black layer formation time” were ended. At the end of the experiment *the number of pipes (piece)*, *the number barren pipes (piece)*, *the length of active pipe (cm)*, *the number seed rows (piece)*, *the mass of cob (g)*, *the mass of core*

(g), seed/core and seed/pipe percentage (%), the thousand kernel weight (g), and the mass of maize (g) were established. The harvest weight of maize (kg), the moisture content of corn (%) and hectolitre weight (kg hl⁻¹) were measured from the four middle lines of the parcels (2*2 lines) at the end of the experiment.

2.4. Statistical analysis methods

For the examination of the statistically justifiable differences between the average values of the results was applied AYDINAPL et al. (2010) *factor analysis of variance* on statistical data, which showed the 5% significant difference (LSD_{5%}) values average values and *relative deviation* (CV%). Regarding the results of the artificial and bacterium fertilizer experiment the artificial fertilizer, wheat straw and bacterium fertilizer treatments to the control, the average values of combined NPK+Bacterial fertilizer to the NPK fertilizer, and the values of straw+bacterial fertilizer treatments to the wheat straw treatments were compared. The average values of the artificial fertilizer and Amykor pot and field experiments in all cases to the average values of the control (untreated) were compared.

The correlation and regression analysis was made with MS Office Excel 2003 to determine the relations between values, the values of correlation coefficient (r) and regression coefficient (r²) (SVÁB, 1967).

3. RESULTS AND DISCUSSION

3.1. Results from experiments of the breeding house

3.1.1. Results of the artificial and bacterial fertilizer pot experiment

In this experiment the effects of different treatments on the soil chemical and microbiological parameters are shown, our assessment was performed by yearly (2010-2013). Along the evaluation of results of the fertilizer, straw and bacterial fertilizer were compared to control, the combined NPK+bacterial fertilizer treatments were compared to results of NPK, and the straw+bacterial fertilizer treatment was compared to treatment of wheat straw.

Among the soil **physical parameters** the Arany-type plasticity index (K_A), the moisture content and the silt & clay content was measured in every year. On the bases of results, the texture of soil was loam. On the bases of chemical properties, the soils were neutral or slightly acidic, containing moderate nitrogen and phosphorus, and well supplied in potassium. Type of soil was calcareous chernozem.

Among the **soil chemical properties** the effect of treatments were evaluated on pH_{KCl} , and the quantity of NO_3^- -N, AL- P_2O_5 and AL- K_2O (mg kg^{-1}). On the bases of average values *in the fertilized treatments* every measured parameter decreased significantly compare to control, the pH decreased and due to the plant nutrient uptake the quantity of nutriments also decreased at the end of experiment. *In the straw treatment* both the pH and the quantity of uptakeable nutrients increased. The use of *bacterial fertilizer* slightly increased the pH, and the amount of certain nutrients (especially the nitrate), at the end of experiment compare to control. When the BactoFil A10 and EM-1 *were combined with NPK* the nitrate-nitrogen content of soil increased. The treatments of straw and NPK+Microbion UNC influenced positively the AL-soluble phosphorus and the *straw+bacterial fertilizer* improved the AL-soluble potassium content of soil increased.

In the *treatments of fertilizer* the **soil microbiological properties** significantly increased almost every year. Especially the number of total bacteria was increased by the treatment. The amount of cellulose decomposing bacteria increased in the *straw treatment*, and also in some cases the number of microscopic fungi also had positive changes. The NPK and Microbion UNC treatments increased the total number of bacteria. The EM-1 straw+EM-1 and Microbion UNC treatments influenced positively the number of cellulose decomposing bacteria while the amount of nitrifying bacteria increased at BactoFil A10 and NPK+bacterial treatments. The straw+BactoFil A10, EM-1 and NPK+EM-1 treatments stimulated the amount of microscopic fungi. The combinations of NPK+Microbion UNC and *straw+Microbion UNC* positively influenced **the nitrate-exploration**. When the *bacterial fertilizer was combined with NPK* the increase in **soil respiration** was observed.

Regarding the **soil enzyme activities** the *treatment of fertilizers* enhanced the phosphatase enzyme activity, but the activities the other examined enzymes decreased significantly. In the *straw treatment* the urease and some cases the phosphatase activity increased by the end of experiment. In the combined treatments of *bacteria fertilizer* the urease phosphatase (straw+Microbion UNC) and dehydrogenase activity (NPK+EM-1) increased. The treatments of NPK+Bacterial fertilizer the catalase activity were stimulated. The combination of NPK+bacterial fertilizer and straw+bacterial fertilizer the phosphatase activity increased the most. The NPK+EM-1 combination stimulated the dehydrogenase enzyme activity of soil.

Concerning the **examined plant parameters** totally can be found that the *dry weight of ryegrass, the dry matter and moisture content of plant* were statistically influenced in most years by the treatment combinations used. Among the treatments the *NPK fertilizer* was mostly the best with resulting in a significant increase in yield in every

year. In the pots getting *straw treatment*, the decrease of biomass was observed. The *bacterial fertilizer* alone was not so effective, than with *combined with NPK*, where the negative influences were mitigated in several cases.

Among the bacteria preparations the Bactofil A10 together with the NPK, the EM-1 alone, while the Microbion UNC together with straw combination resulted in a statistically justified change in the examined properties. The combination of *straw + bacterial fertilizer* generally caused negative effects.

3.2. The results of the fertilizer and mycorrhizal preparation experiments

3.2.1. Results of the pot experiment

Along the evaluation of the pot experiment setting with artificial fertilizer and root vitalizing Amykor, the results from Amykor treatment was compared to control, while the combined treatments of NPK fertilizer+Amykor were compared to the results of control and NPK-treatments.

Among the **soil physical parameters** the Arany-type plasticity index (K_A), the moisture content and the silt & clay content was measured from the *control* soil. On the bases of results, the texture of soil was sand. On the bases of chemical properties, this soil were slightly acidic, containing low nitrogen and phosphorus content, and medium supplied with potassium. Type of soil was humus sandy soil.

Among the **soil chemical properties** significant increase was measured in the examined parameters *in the fertilized pots* compare to the control treatment at the end of the experiment, however, the pH was decreased due to the acidifying effect of the fertilizer. The two doses of Amykor (1x and 2x) most of the time increased the pH of the soil and increased the uptakeable quantity of nutrients as well. In the treatments of the *Amykor 1 and NPK combination* (1x and 2x doses) decreased the pH in slightly range compare to control, but the quantity uptakeable nutrients (especially the potassium) increased at the end of the experiment. In the *treatments of Amykor 2* (1x and 2x doses) the soil pH decreased in slightly, but the quantity of certain nutrients (especially the nitrate and potassium) increased. In the treatments of the *Amykor 2 and NPK combination* (1x and 2x doses) the soil pH increased, and also increased the quantity of soil nutrients. Among the treatments at the Amykor 2 doses and combination the nitrate content, at NPK+Amykor 1 simple and double dose the AL-soluble potassium and at the double dose of Amykor 1 and 2 preparations the AL-soluble potassium content increased.

Regarding the **soil microbiological properties** *in the fertilized treatments* significant increase was experienced in the quantity of nitrifying bacteria on the bases of

average results of pot experiment, while the quantity of aerobic cellulose decomposing bacteria and microscopic fungi decreased. In the *treatments of Amykor 1* (1x and 2x doses) the total number of bacteria and cellulose decomposing bacteria increased significantly proved by statistical methods. *Amykor 1 and NPK combination* (1x and 2x doses) treatments positively influenced the quantity of nitrifying bacteria. The *treatments of Amykor 2* (1x and 2x doses) and the *Amykor 2 and NPK combination* (1x and 2x doses) stimulated positively mainly the total number of bacteria.

The fertilization increased the urease enzyme activity while the different doses and combinations of Amykor stimulated the phosphatase enzyme activity.

Among the mycorrhizal preparations *the Amykor 2 doses and their different combination* proved to be the most effective regarding to the examined soil parameters. Among the positive effect of this mycorrhizal product may be the applied quantity of this preparation, which was higher than the recommended doses of *Amykor 2*.

Regarding to the results of the properties in the *overland parts of the onion crop*, the NPK, and the *Amykor 1* doses as well as their treatment combinations resulted in significant changes. The wet weight of the *underground parts of the onion* was increased significantly by the 2x dose of *Amykor 1* and its combination with fertilizer. The cumulative average values of onion wet biomass the NPK+*Amykor 1* and the *Amykor 2 1x* dose seemed to be more effective. Concerning the grown of shoots the *Amykor 1 preparation*, regarding to underground pieces of plant the *Amykor 2* can be proposed for using.

3.2.2. Results of the field experiment

The results of the field experiment setting up with NPK and Amykor plant vitalizing preparation are shown on the soil parameters and plant biomass (Debrecen-Látókép, 2011-2012).

Among the **soil physical parameters** the Arany-type plasticity index (K_A), the moisture content and the silt & clay content was measured from the control soil. On the bases of results, the texture of soil loam, the soil was with slightly leached. On the bases of chemical properties, this soil was weakly acidic, containing medium nitrogen and phosphorus content, and well supplied with potassium. Type of soil was chernozem soil.

Among the **soil chemical properties** the nitrate content of soil was mainly influenced by the effect of the *fertilization and Amykor* mixing them to the 0-20 cm soil layer, while the quantity of AL-soluble phosphorus, and potassium was most effected by

Amykor applying in time of sowing. Concerning the nutrient content of soil, the Amykor 1x dose can be recommended (getting to soil together with the seed).

Regarding to the **microbiological parameters of soil** the total amount of bacteria and microscopic fungi were stimulated by the different doses of *Amykor* (0-20 cm). In the **nitrate exploration** the *NPK-fertilization* resulted in a significant decrease. Among the **enzyme activities**, inhibitor effect of *Amykor* (0-20 cm) was detected on urease activity, while the *Amykor getting to soil with the seed* stimulated the phosphatase activity.

The leaf area index of corn increased significantly in the treatments of *NPK and Amykor* getting to the soil with the seeds. The **number of corncob** was influenced positively by the *doses of Amykor* (0-20 cm), **the active length of corncob** was increased slightly by the *Amykor 1 (applying with seeds)*. The **seed/cob (%)** decreased in the *NPK* treatments. The **thousand-seed weight (g)** increased in the treatment of *Amykor 1x* (0-20cm). The **quantity of harvesting yield (kg plot⁻¹)** was significantly increased by the *Amykor* (0-20 cm) treatments, the results were statistically proved, at the same time the **moisture content of the yield (%)** was decreased. The **hectolitre mass** is an important parameter (kg hl⁻¹), the *Amykor getting to soil with the seeds* increased this parameter.

With the results applying these bacteria preparations we proved the mainly positive and sometimes negative affects of the treatments on the soil parameters. Among the preparations the *Amykor* (0-20 cm) doses can be recommended for use in similar soil, and good or lower nutrient supplying capacity of soils.

The effect mechanism of microbial preparations is largely depending on the temperature and moisture content of soil. In the experiment the only way for water supply came from the precipitation in both the growing seasons. So it is important to show certain **meteorological** dates regarding to the experimental areas.

- **In 2011** the average monthly values of temperature was higher in the growing season compared to the average of the 30-years. In July nearly 180 mm rainfall was measured showing the uneven distribution of precipitation. Compare to the 30-year average, a small increase was in temperature, while the precipitation was about the same amount.

- **In 2012**, during the first half of the growing season more precipitation and higher monthly mean temperature was detected compared to the 30-year average. In the period of August-September, there was a minimal monthly rainfall and a higher temperature, so a significant shortage of water was experienced in the growing season.

4. NEW AND NEW-TYPE SCIENTIFIC RESULTS

Based on our examination results the new scientific results and conclusion are summarised:

1. The results demonstrated that the soil chemical and microbiological parameters were positively influenced by the **bacteria products** (BactoFil A10, EM-1 and Microbion UNC) and **root vitalizing** (Amykor) product. The larger parts of the positive impact were not significant effects under the given soil conditions; so the need of a certain type of monitoring system would be necessary in the application of these products.
2. In pot experiment the specific effects of tested bacterial preparations were manifested which were occurred of the microbial composition the products. Using the **BactoFil A10** preparation statistically increased the number of nitrifying bacteria. Applying the **EM-1** preparation increased the number of aerobic cellulose decomposing bacteria and microscopic fungi. The **Microbion UNC** application increased the total number of bacteria and the number of aerobic cellulose decomposing bacteria, intensified the activity of phosphatase enzyme. The specific impacts of preparations should take into account along the application of products.
3. The **application of bacterial fertilizers together with fertilizers** increased the microbiological activity of soil through indirect way, as improved the plant nutrition. **Applying with wheat straw** supported the increasing of the soluble plant nutrients and other physical soil parameters due to the changes of the nutrient-rate and other indirect impacts of straw.
4. Along **the recommended and increased dose of** micorrhyza fungi containing **Amykor 1** application increased the bacteria population of soil and several enzyme activities. Indirectly, the mass of root and bulbs of onion and the biomass of onion improved at these treatments. The positive effect of this product on root vitalizing and the plant growth with onion test plant was confirmed in pot experiment.
5. In field experiment the effect of **Amykor mycorrhiza containing** product was **influenced by the application method**. **The simple and increased dose of the treatment worked into 0-20 cm soil layer** not influenced, while **the treatment with sown simultaneously of maize** had significant positive effects. The **simple dose of Amykor** (applying at time of sowing) treatment increased the easily available nitrate and phosphorus, as well as the number of microscopic fungi. Concerning **the triple dose of Amykor** increased the phosphatase enzyme activity and the Leaf Area Index of maize, too. As a result of the preparation of fungi-partner of preparation may act as a strong influence on the properties which have been regarded in the application process.

5. SCIENTIFIC RESULTS UTILIZABLE IN THE PRACTICE

Based on our examination results the new scientific results utilizable in the practice are summarised:

1. The bacterial preparations need to be applied according to their nature. They are not considered as a general all-extending “Fertilizers”. The microbiological composition of preparations defined the method of the application. Among the bacteria preparations the **BactoFil A10** product was effective on soil properties when it was combined with NPK fertilizer. The **EM-1** product alone and with NPK, while the **Microbion UNC** combined with straw caused statistically proved positive effect on the different parameters of soils and plants.
2. As a result of the Amykor root vitalizing product the development of connection of plant-microbes is an influencing factor of the symbiotic nature. In pot experiments **the Amykor 1 root-vitalizing product** was the most effective on the examined parameters with onion test plant. It can be recommended in different soils, so in sandy soils weakly supplied in nutrients with slightly acidic pH too. As this product contains clay mineral, it can serve a soil conditioner.
3. Regarding to the results of field experiment **the Amykor treatment with sown simultaneously of maize (simple and triple dose)** seemed to be the most effective on the examined soil and plant parameters. The application of mycorrhiza fungi preparations are influenced by the method and time of application.

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