Research on the Microbiological Diversity of the Chernozem Soil of Dobroudja, in the VALU TRAIAN Region

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Abstract. The most poignant global crisis is the one triggered by the conflict between the environment, the agriculture and the society. The continuous loss of humus and of the biodiversity combined with the decrease of the soil’s fertility, with losing its capacity to produce nourishment, energy and raw stocks lead to the impoverishment of the population. Dobroudja is the most barren region, the draught being an endemic phenomenon, desertification also being present, currently in the soil there is a maximum 2.65 t/ha of active biological substance and 109 t/ha of humus, which represents a 56% decrease compared to 80 years ago. We have set off to create an adequate plan to ecologically and biologically reconstruct the soil based on the research done in the Valu Traian area of Dobroudja. Firstly, we have stock-listed the current biological state of the soil in the Dobroudja ecosystem and then, by using laystall and Biovin bioactivators, we will try to feed into the soil as much micro-organisms and specific organisms as possible that are necessary to restore and biocatalytically reconstruct the organic substance into heteropolycondensated humus.

Keywords: humus, micro-organisms, bioactivators, fertility, biodiversity.

INTRODUCTION

80 years ago Dobrogea’s soils comprised a quantity of humus that surpassed 6%, which means approx. 235 t/ha (specific weight=1.3) expanded on a 30 cm depth. During that period Dobroudja’s soil contained approx. 15 t/ha micro-organisms and organisms that were undergoing an intense activity in order to disaggregate, degrade and decompose the organic substance, as well as to restore and biocatalytically reconstruct it into heteropolycondensated humus.

Currently, according to the data published by Berca M. (2008) [1], there are maximum 2.65 t/ha of active biological substance in Dobroudja’s soil. The result is that the gravimeter mass of the humus from the soil has been reduced from 235 t/ha (80 years ago) to its current 109 t/ha. This loss totals a 56%. Therefore, at the two Berlin inter-ministry conferences from January 2009 and 2010, the EU specialists have set an objective in restoring the biological activity in the soil by using different technologies, organic substance and biological activators. In order to start and create an adequate plan for the biological reconstruction of the soil we had to research the Valu Traian area of Dobroudja to assess the current biological state of the soil in the Dobroudja agroecosystem, followed by using new techniques of soilwork, using laystall and Biovin bioactivators, we will try to feed into the soil as much micro-organisms and specific organisms as possible that are necessary to restore and biocatalytically reconstruct the organic substance into heteropolycondensated humus.
MATERIALS AND METHODS

The experiments have taken place on a 7.5 ha lot situated in the external area of Valu Traian, in Constanța district. Josef wheat was cultivated on the entire area, which was divided in 7 variants, each variant being administered a different type of fertilizer in different quantities and periods, as follows:

- **V1** – only chemical fertilizers-100kg/ha N15P25K15 in autumn, 150kg/ha NH₄NO₃ at the beginning of spring;
- **V2** – Biovin organic fertilizer 400kg/ha and Biovin 30 of l/ha, ½ at herbicide stage and ½ at flour stage;
- **V3** – garden soil - 15t/ha in autumn;
- **V4** – l/ha of Biovin 30, ½ at herbicide stage and ½ at flour stage;
- **V5** – Biovin 150kg/ha administered during sowing, 150kg/ha NH₄NO₃, 40kg/ha at the beginning of spring, 50kg/ha at herbicide stage and 60kg/ha at flour stage;
- **V6** – Biovin 375kg/ha, liquid Biovin 30 of l/ha, ½ at herbicide stage and ½ at flour stage, 1mc Green Mycos, 1l Bactofil Professional;
- **V7** – March – were not applied amendments.

Biovin Fertilizers are being administered for the first time in Dobroudja:

- **Biovin** is being produced through a technological process from grape kernels. 12 years of western research proved the following: it aerates the soil, it improves it (it contains up to 70% humus makers) it purveys all plants with nutritive elements and biostimulators, it enriches the soil with micro-organisms that create humus, it strengthens the roots and it multiplies the percentage of smooth roots and radicular winthergr;
- **Bactofil Professional** is a product used for the ecologizing of the soil and it contains: *Azospirillum brasiliense, Azotobacter vinelandi* (azote fixators in associative system up to 100 – 110 kg/ha), *Bacillus megaterium, Bacillus polymyxa, Pseudomonas fluorescens, Streptomyces albus* and other versions of micro-organisms which speed up the decomposing of extinct organic substance and increase the agrod disponibility of phosphorus and potassium;
- **Green Mycos** contains a mixture of *Penicillium sp* dominant micro-organisms, algates, polysaccharides, enzymes, hormones and inoculation of arbuscular mycorrhizal and it activates the soil for up to 20 years.

The experiments started in autumn of 2010 by reaping soil samples as follows:

- on a 0-20 cm depth in order to determine certain biological parameters (bacteria, actinomycetes, fungus) the following has been carried out:
  - quantitative testing of bacteria and fungus mycroflora, [3], [4], - the method of cultivating decimal dilutions of soil on solidifying nutritive environments (pulvis yeast extract 2.5g; Peptona 0.2g; Agar 17-20g. It was sterilized for 20 minutes at 120°C), and mycromyces, Czapec-Dox environment (NaNO₃ 3g; K₂HPO₄ 1g; MgSO₄ 0.5g; KCl 0.5g; FeSO₄ samples; Saccharine 30g; Agar 17-20g; pH 5.5; it was sterilized for 30 min at 115°C) [3], [4], [7].
- testing the fixing bacteria clear of atmospheric azote (Azobacter), the method of cultivating decimal dilutions of soil on solidifying nutritive Ashby environment [5], (Manitol 15g; K₂HPO₄ 0.2g; MgSO₄ · 7H₂O 0.5g; NaCl 0.2g; CaSO₄ · 7H₂O 0.1g; CaCO₃ 5g; Agar 17-20g. it was sterilized for 30 min at 115°C);

The total number of bacteria per gram of soil was calculated using the formula: \( \text{no. bacteria, actinomycetes, microfungi} = X \text{ colonies} \times \text{dilution} \times 10 \times 100/100-U \) where \( X = \)
average of colonies grown on culture medium, $10 = $ balancing coefficient of 0.1 ml of inoculum in the reporting of dilution soil U% = soil moisture. [8]

RESULTS AND DISCUSSION

Regarding the biological parameters, heterotrophic bacteria, nitrogen-fixing bacteria, mycromyces, useful in the microbiological degradation process and in the humus reconstruction process were found in the following quantity (NTBx10^5 ufc/g arid soil – total number of X bacteria dilution gradient, colony making units per soil gram):

![Graph](image)

Fig. 1. Changes of abundance of nitrogen-fixing bacteria (October 2010)

At the beginning of the experiment, density of free nitrogen-fixing bacteria ranging from 39.58x10^5 CFU/g dry soil (Fig. 1), to 115.8x10^5 CFU/g dry soil (Fig. 1).

In case of variants II, III, and V values are very close to those recorded for control (Fig. 1).

The highest number recorded for variants II, III, V and VII ranging between 95.52x10^5 CFU/g dry soil (Fig. 1), and 115.8x10^5 CFU/g dry soil (Fig. 1), rely on local trophic conditions.

Thus, the lowest abundance was detected in variant VI, nitrogen-fixing bacteria having a mean abundance of 39.58x10^5 CFU/g dry soil (Fig. 1).

The abundance was highest instead on variant VII, martor, in which case the total number of nitrogen-fixing bacteria reached 115.8x10^6 CFU / g dry soil (Fig. 1).
Fig. 2. Distribution of heterotrophic bacterial density in the initial stage of the experiment (October 2010)

Thus, the lowest abundance was detected in variant VII, heterotrophic bacteria having a mean abundance of $19.30 \times 10^6$ CFU/g dry soil (Fig. 2).

The abundance was highest instead on variant VI, in which case the total number of heterotrophic bacteria reached $204.89 \times 10^6$ CFU/g dry soil (Fig. 2).

In case of variants III, and V values are very close to those recorded for control (Fig. 2).

Fig. 3. Distribution of microfungi density in the initial stage of the experiment (October 2010)

Thus, the lowest abundance was detected in variant II, distribution of microfungi having a mean abundance of $9.99 \times 10^5$ CFU/g dry soil (Fig. 3).

The abundance was highest instead on variant I, in which case the total number of microfungi reached $23.02 \times 10^5$ CFU/g dry soil (Fig. 3).

In any case, there was a certain uniformity of abundance of microfungi beginning of the experiment.
CONCLUSIONS

The initial estimations have revealed a relatively low abundance variability between different experimental variants.

Changes in microbial abundance in experimental and control reflect the heterogeneity of normal physicochemical and trophic conditions of the soil, the values recorded can be considered normal for chernozem soil type.

The results of the microbiological analysis of the soil show a weak contribution of the micro-organisms in improving the fertility and the biodiversity. In order to improve this state an increase in the quantity of the micro-organisms by adding bacteria into the soil is needed, leading to the existence of valuable nourishment for the micro-organisms (straws, organic leftovers, mulcs) as well as adding Biovin bioactivators.

Practicing an agriculture that harmonizes the biochemical laws with the ecological ones must be enforced; as well as imposing an agriculture that bioecologically reconstructs the soil due to the fact that degradation has reached a high speed in the agricultural ecosystems because of the excessive chemicalization and also because of the unfit soil works. These works predominantly favour the mineralizing process releasing high values of CO$_2$ which directly contributes to the process of climatic changes.

REFERENCES