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# Effect of using sludge from waste water treatment plants as an organic reserve in agriculture

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## Abstract

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Large amounts of sludge are produced in the biological treatment of wastewater. They accumulate in the areas off treatment plants and can pollute the natural environment. Many studies carried out to characterize the sludge found that they are biomass and represent an organic reserve in connection with the lack of organic fertilizers in our country and disturbed balance of organic matter in the Bulgarian soils. It is necessary to establish the effect of sludges as a fertilizer and their influence on the yield and quality of plant production and the soil. The results were compared with those obtained using the manure.

The purpose of the study is to determine the effect of using WWTP sludge and the impact on the yield and quality of plant production and soil. To compare with the effect of using manure.

A vegetation experiment was conducted on two soil types – leached cinnamon soil and smolnitza. Two soil improvers were used – sludge from WWTP and manure from a cow farm. An indicator crop is silage corn. Experiments were carried out at a percentage of sludge and manure (5%, 15%, 25%) by weight of the soil, including controls, without added improvers. 3 kg soil conteiners were used in 4 repetitions. The experiment was performed out in 2020. Standardized laboratory methods have been applied. When collecting the indicator crop, the yield was reported and a mathematical-statistical analysis was made. Maize was harvested, according to variants in the 7-8 leaf phase, being drawn from each pot -container separately, after which the results were averaged from the individual replicates. The plant production and the soil from the different variants were analyzed according to chemical, agrochemical and microbiological parameters. A comparison was made with the effect of using manure.

It was established that the sludge does not contain heavy metals above the MPC and other pollutants that may limit its use in agricultural practice. The yield of maize grown on smolnitza and cinnamon forest soil increases as the fertilization rate increases. Higher yields are obtained, when using the sludge on the cinnamon forest soil, while the smolnitza responds better to manure fertilization. The statistical analysis shows that the main factor influencing the yield is the rate of fertilization, followed by the type of improver applied and the soil difference. The results were compared with those obtained using the manure. No negative impact on plant production was observed when using the sludge. The sludge have a positive impact on the soils, as they are rich in macro- and microelements and the values of heavy metals are below the MPC indicated in the legislation. The sludge did not burden the two types of soils used in the vegetation experiments with faecal bacterial flora, both in terms of indicator microorganisms and pathogens.

The obtained results build on the conducted research and establish that sludge from WWTP is not inferior to organic fertilizers and can be used as an organic reserve, in connection with the shortage of organic sources in our country and disturbed balance of organic matter in Bulgarian soils, according to the requirements of the legislation.

*Keywords:* WWTP; sludge, organic reserve; soil improver; soil fertility
*Abbreviations:* WWTP – Waste water treatment plant; MPC – Maximum permissible concentration; MPN – Most probable number

## Introduction

The production of sludge in biological wastewater treatment is increasing every year, which is a result of the increase of the population in larger settlements and the accelerated development of industry. They accumulate in the area of treatment plants, hinder their functioning and can pollute the environment. With proper management of sludge, and in accordance with current legislation, they can be used for fertilizing agricultural practice

In connection with the shortage of organic fertilizers in our country and disturbed balance of organic matter in the Bulgarian soils, it is necessary to search for reserves. A number of studies of the chemical, agrochemical and microbiological characteristics of the sludge, have established that they are biomass rich of nutritional macro and microelements, organic mass, biologically active substances and represent an organic reserve (Marinova, 2002; 2008; Pallavi et al., 2015; Colón et al.,2017; Georgieva et al.,2017; Stankova et al., 2021).

According to a European Commission report, published in 2010, 39% of the sludge produced in the European Union, is recycled in agriculture (Lamastra et al., 2018). It is expected that in the coming years, stabilized sludge will be used in large quantities in agriculture in many EU member countries, such as France, Germany, Italy, Spain, and also in Great Britain (Colón et al., 2017). In the Mediterranean areas, e.g. in France, about 30% of the produced sludge is used for agricultural purposes (Marinova et al., 2015).

The main document specifying the procedure and method for the utilization of sludge in Bulgaria, is the Ordinance on the procedure and method for the utilization of sludge from waste water treatment, through their use in agriculture, adopted by Resolution No. 201 of 04.08.2016, State Gazette. no. 63 of 2016. The Ordinance also defines the requirements that the sludge must meet in order to ensure that they do not have a harmful impact on human health and the environment.

The purpose of the research is to determine the effect of using WWTP sludge and its influence on the yield and quality of plant produce and soil, to compare with the effect of using manure.

## Material and Methods

A vegetation experiment was carried out on two soil types – Leached cinnamon soil from the area of the village of Tsalapitsa and Smolnitza from the area of the village of Bozhurishte.

Two soil improvers were used – sludge from WWTP and manure from a cow farm. An indicator crop is silage corn.

The experiment was carried out at different percentages of sludge and manure (5%,15%, 25%) by weight of the two types of soil, and controls, without added improvers were also included. 3 kg soil conteiners were used in 4 replicates. The experiment was carried out in 2020.

The chemical and agrochemical parameters were analyzed in accredited laboratories by applying verified international standardized methods (ISO) and validated laboratory methods. Samples were analyzed from:

- the starting products – manure, sludge and both types of soil – before starting the experiment;
- the plant production of the experimental crop after its harvest
- soil samples of all variants after completion of vegetation experiments

When carrying out the microbiological tests of the sludge and the soil samples from the experimental variants with sludge, a validated laboratory test method LMI:ICZ 02.01: „Soils, biowaste and sludge from wastewater treatment. Determination of microbiological indicators“ was applied.

During the vegetation period, the sprouting and development of the crops was followed, which was documented with photographic material.

## Results and Discussion

***Sludge and manure characterization***

The results of the studies of the original organic materials – sludge and manure – are presented in Table 1.

From the chemical and agrochemical analyzes carried out, it was found that both products have a neutral to slightly alkaline reaction. The total amounts of biogenic elements show that the manure contains higher values of total ni-

**Table 1. Agrochemical analysis of the WWTP sludge and manure, used for the vegetation experiments**

| Analyzes | Sludge from WWTP Sofia | Manure from a cow farm |
|---|---|---|
| pH –$H_2O$ | 6.85 | 7.51 |
| Dry residue, % | 75.60 | 42.00 |
| Organic C, % | 22.53 | 16.92 |
| Total N% | 1.13 | 2.06 |
| Total P, % | 0.87 | 1.39 |
| Total K, % | 0.16 | 0.40 |
| Ammonium N, % | 0.0069 | 0.007 |
| Nitrate N, % | 0.48 | 0.70 |
| Mobile P, % | 0.068 | 0.08 |
| Mobile K, % | 0.059 | 0.03 |
| Ca, % | 1.51 | 0.17 |
| Mg, % | 0.45 | 0.08 |
| As, mg/kg | <10 | – |
| Pb, mg/kg | 106.18 | 88.00 |
| Cd, mg/kg | 1.87 | 0.89 |
| Cu, mg/kg | 39.40 | 6.60 |
| Ni, mg/kg | 31.00 | 28.90 |
| Cr, mg/kg | 36.90 | 29.00 |
| Hg, mg/kg | <10 | – |
| Zn, mg/kg | 843.50 | 652.00 |

trogen, phosphorus and potassium. The content of organic carbon in both improvers is enough to reduce the deficit of organic matter in the soil and satisfy the needs of plants for nutrients.

The content of total nitrogen in the sludge was established to be 1.13%, total phosphorus 0.87% and total potassium 0.16%. Only a small part of the nitrogen is found in a plant-absorbable form. The ratio between ammonium and nitrate forms indicates that the mineralization process is not fully completed. In the the overstayed sludge , the ratio between nitrogen, phosphorus and potassium is not balanced – it is in favor of nitrogen. This should be taken into account and when using the sludge in agriculture, balancing fertilization with mineral fertilizers should be carried out, depending on the requirements of the crops.

According to literature sources, the sludge are characterized by a variable chemical composition: the nitrogen content varies from 1 to 7% (to absolute dry matter), phosphorus from 1 to 4%, potassium – from 2 to 3% (Marinova et al. , 2000; Tsolova & Marinova; 2005). During the different treatments of the sludge, the nitrogen content changes. For example, drying reduces the nitrogen content by 40-50%. Composting also changes the nutrient content. Some specialists value, such compost more as a soil conditioner, than as a fertilizer (Marinova, 2000).

The amount of phosphorus found in wastewater is different and reaches over 10% of the dry matter. On average it is 1-2%. Phosphorus plays a major role in tissue growth and development. Phosphorus content is subject to significant fluctuations and with varying availability to plants. The sludge satisfies the total phosphorus requirement of the corn. It can be said that by dosing the sludge, to meet the nitrogen needs of a given plantation, the phosphorus needs for 2-3 harvests are also met. Dewatered sludge is the main source of phosphorus for plants. Applied to the soil, at a rate of 1.5 t/da to dry matter, it is sufficient to satisfy the need of cereals and pasture grasses for a period of 2-3 years (Marinova, 2008).

The potassium content in the sludge is extremely low. The noted values are of the order of 0 – 0.2%. (Marinova, 2008).

The amount of dry matter is 75.60% and the sludge can be transported by mobile means, such as manure. pH of the medium is 6.85. Organic carbon is 22.53%. All investigated heavy metals in the sludge have values below the MPC. (Tytła, 2020; Zhang et al., 2017).

Regarding the content of heavy metals in WWTP sludge, there are many studies.

The results of a study of sludge from five municipal WWTPs located in the largest industrial area of Poland, show that the concentrations of selected heavy metals (Cd, Cr, Cu, Ni, Pb, Zn and Hg) do not exceed the permissible levels in terms of requirements for the agricultural use of sludge (Tytła, 2020). Similar results were reached in a study of sludge from five WWTPs in southern Spain (Alonso et al., 2009)

When a health-ecological study was carried out on the content of Pb, Cd, As, Cu, Zn, Cr, Ni and Hg in 400 sludge samples from 43 WWTPs in Bulgaria, it was found that in most samples the level of toxic elements was below the MPC, defined in the Ordinance on the utilization of sludge in agriculture (Sidjimov et al., 2013).

According to studies of sludge obtained during the biological treatment of wastewater (Marinova, 2002; 2008; Stankova et al., 2021), before their introducing into the soil, analyses for the content of the main macro-, microelements and heavy metals should be made. Fertilizing sludges are recommended that have more than 30% dry matter and heavy metal content below the maximum permissible concentrations, according to the requirements of the legislation. The rate is determined by the nitrogen content.

The manure used in the vegetation experiment contained organic carbon 16.92%. It is well stocked with total nitrogen. The values of the main nutrients were, respectively, 2.06% for total nitrogen, 1.39% for total phosphorus and 0.40% for

**Table 2. Microbiological results of tests carried out on WWTP sludge, used in a vegetation experiment as a soil improver**

| Investigated microbiological parameters | *E.coli* MPN / g* | *Coliforms* MPN / g* | *Enterococci* MPN / g* | *Clostridium perfringens* MPN /g* | *Salmonella* spp. presence/absence in 20 g |
|---|---|---|---|---|---|
| Norms according to the Regulation | up to 100 /g (up to 1. $10^2$ /g) | absent from the regulation | absent from the regulation | up to 300 /g (up to 3. $10^2$ /g) | absence (not allowed) in 20 g wet weight |
| Sludge from WWTP Kubratovo, with which the experiment was conducted | <0,30 | >1,1. $10^5$ (> 110 000) | <0,30 | >1,1. $10^3$ (>1 100) | absence in 20 g |

* MPN/g – most probable number – most probable number of the microorganism of interest in 1 g

total potassium. Trace elements and heavy metals are in acceptable concentrations and do not pose a risk, when using manure for fertilization.

***Microbiological studies of WWTP sludge***

The sludges was examined for the presence of indicator and pathogenic microorganisms – *E. coli, Clostridium perfringens, Salmonella* spp., according to the requirements of the Ordinance on the use of sludges. Non-normative groups of microorganisms – coliforms and enterococci – were also analyzed. In this way, more complete information was obtained about the hygienic condition in terms of the faecal load of the sediment.

As can be seen from Table 2, the sludge did not contain *E. coli, Enterococci and Salmonella* spp. *Coliforms* are in high quantities, as well as clostridia – *Clostridium perfringens* exceeds the norm up to 300/g (ULF/ g). This means that the death processes of these two groups of microorganisms require a certain period of sediment residence time.

The qualities of WWTP sludge in terms of microbiological parameters have been the subject of various studies.

In a Lebanese study on the quality assessment of sewage sludge, no helminth eggs and *Salmonella* bacteria were found in 36 sludge samples. Deviations are related to the amount of *E. coli, Staphylococcus aureus* and *Acinetobacter* spp., which are elevated in some of the samples (> 100 CFU/ 100 g) and do not meet the requirements of local legislation (Romanos et al., 2019).

In a Bulgarian study of sludges from sewage treatment plants for the purpose of using them as soil improvers, no eggs of helminths and pathogenic bacteria of the genus *Salmonella* were found. In case of insufficient treatment or storage, bacteriological findings of representatives of the family *Enterobacteriaceae – E. coli, coliforms* from the genera *Klebsiella, Citrobacter* and *Enterobacter* are found. The most resistant and in higher quantities are sulfite-reducing clostridia, to which *Clostridium perfringens* belongs (Georgieva et al., 2017).

A Brazilian study characterized the diversity and microbial population structure of 19 wastewater sludges from São Paulo, Brazil. We are looking for a connection with the sources of sludges – domestic and mixed wastewater (domestic + industrial), biological treatment and chemical properties, through the methods of molecular biology. It is concluded that sludge has a high microbial diversity, which can affect the efficiency of the WWTP and the quality of the soil, regardless of whether it is used as fertilizer (Nascimento et al., 2018).

***Chemical and agrochemical characteristics of the soils with which the study was conducted***

**– Smolnitza**

This type of soil does not different in the individual areas of distribution, which shows that the decisive factor for their formation is the plain relief conditions, the specific montmorillonite character of the soil-forming materials and the nature of the plant relations in their initial formation. They are characterized by an intense gray-black coloring of the soil profile, the thickness of which reaches 60-80 cm. Despite the intense black color, these soils are not distinguished by a particularly high content of humus – usually ranges from 2.5 – 3.5% and can reach up to 5 – 6%.

The data from the agrochemical analysis of the leached smolnitza from the area of Bozhurishte village (Table 3) show that the soil is well stocked with mobile phosphorus – 23.8 mg $P_2O_5$/100 g soil and with mobile potassium – 27.7 mg $K_2O$/100 g soil. The total nitrogen is approximately 0.1%, and the sum of the mineral nitrogen fractions is 9.2 mg/kg. The content of microelements compared to other soils in the country is higher, which is due to the location and related accumulation processes. The pH of the leached smolnitsa is slightly acidic to neutral. The content of heavy metals is below the maximum permissible concentration.

**– Leached cinnamon meadow soil**

The leached cinnamon-meadow soil from the Tsalapitsa region is slightly acidic with a pH of 6.1 in water, slightly humus and with very poor agrochemical properties. This soil is much poorer in essential nutrients than leached smolnitsa . The

**Table 3. Chemical and agrochemical characteristics of smolnitza and leached cinnamon meadow soil, with which the vegetation experiments were conducted**

| Analyzes | Smolnitza | Leached cinnamon meadow soil |
|---|---|---|
| pH –$H_2O$ | 6.9 | 6.1 |
| pH –KCL | 6.2 | 5.8 |
| Hummus, % | 2.10 | 1.3 |
| Total N, % | 0.11 | 0.03 |
| Mobile $P_2O_5$, mg/100g | 23.8 | 0.84 |
| Mobile $K_2O$, mg/100g | 27.7 | 15.05 |
| Mineral N – ($NH_4$+ $NO_3$), mg/kg | 9.2 | – |
| Cu, mg/kg | 35.5 | 4.3 |
| Zn, mg/kg | 18.5 | 15.1 |
| Mn, mg/kg | 600.0 | 588 |
| Cr, mg/kg | 19.5.0 | 16.3 |
| Ni, mg/kg | 21.5 | 9.4 |
| Co, mg/kg | 18.5 | 13.5 |
| Pb, mg/kg | 27.5 | 25.2 |
| Cd, mg/kg | 0.5 | 0.5 |

soil is very poorly stocked with total nitrogen – about 0.03%, middle stocked with mobile potassium – 15.05 mg $K_2O$/100g and very little, almost not stocked with mobile phosphorus. And in this soil, the content of heavy metals is below the maximum permissible concentration (MPC) (Table 3).

***Physicochemical characteristics of the soils with which the study was conducted***

The results of the physicochemical characteristics of the two soils, used for the vegetation experiments, are shown in Table 4.

Cinnamon – meadow soil is leached, moderately acidic (pH=6.1), moderately colloidal ($T_{8,2}$ is between 20-30 megv/100 g), mineral, deeply built soil. No acidification (exchange acidity AL=0).

According to the prevailing clay mineralogy (Tsa=78.2% of T8.2), it is montmorillonite-illite with evolution to montmorillonite-illite.

Here, a greater basic saturation than the value of the strongly acidic ion exchanger of the soil adsorbent (Bases > Tsa) is noticed, which indicates a contemporary course of clay formation in the leached soils (Table 4).

According to the quantitative classification system, the smolnitza is leached, neutral (pH=7.0), highly colloidal, mineral, totally constructed with a moderate colloidal construction of Achor, with a montmorillonite composition of the clay minerals and a process towards montmorillonite formation. There is no acidification (Vol. AL=0), and due to the action of the hydrolysis-alkaline adsorption salt (bases Tca), the cation exchange on the roots is of the hydrolysis-alkaline type (Table 4). The clayey and rich colloidal mechanical composition determine a number of unfavorable physico-mechanical properties of the resin – high stickiness, cohesiveness and plasticity in the wet state and high hardness, combined with strong cracking upon drying (Table 4).

***Indicator crop development and maize yield in the vegetation experiment***

Maize germinated and developed favorably in all test variants. During the growing season, irrigation was carried out according to the marginal field moisture content.

Photographs were taken several times during the planting of the experiments and during the vegetation period to establish the development of the plants. Differences are observed in the individual variants, depending on the rate of the used components – sludge and manure.

**Table 4. Physicochemical parameters of the soils, with which the vegetation experiment was performed – cation exchange capacity ($T_{8.2}$), strongly acidic ($T_{CA}$), weakly acidic ($T_A$) positions of the soil substrate, general acidic soils ($H_{8.2}$), exchange acidity (Al) and composition of the most important exchange-adsorbed cations of the leached smolnitza and alluvial-meadow cinnamon soils**

| Soil | Pн / $H_2O$ | $T_{8.2}$ | $T_{CA}$ | $T_A$ | Exchanged $H_{8.2}$ | Exchan-ged Al+H | Exchanged Ca | Exchanged Mg |
|---|---|---|---|---|---|---|---|---|
| | | in eqv/100 g | | | | | | |
| Leached smolnitza | 7.0 | 54.9 | 51.9 | 3.0 | 2.2 | 0.0 | 47.0 | 6.0 |
| Leached cinnamon meadow soil | 6.1 | 30.2 | 23.6 | 6.6 | 5.0 | 0.0 | 21.4 | 4.4 |

| $T_{CA}$ In % of $T_{8.2}$ | $T_A$ | Exchanged $H_{8,2}$ | Exchanged Al | Exchanged Ca | Exchanged Mg | Degree of saturation with bases, % |
|---|---|---|---|---|---|---|
| 94.53 | 5.46 | 4.0 | 0.0 | 85.6 | 10.9 | 96.0 |
| 78.2 | 21.8 | 16.6 | 0.0 | 70.9 | 14.6 | 83.4 |

**Table 5. Yield of silage maize (in grams) from a vegetation experiment with the participation of manure and WWTP sludge on two soil types**

| Variants | 1 measurement | 2 measurement | 3 measurement | Average | Relative yield, % |
|---|---|---|---|---|---|
| *Fertilizing with sludge – leached cinnamon-meadow soil* | | | | | |
| 1. Control – pure soil | 13.47 | 16.53 | 14.06 | 14.68 | 100.00 |
| 2. 5% sludge by soil weight | 55.09 | 59.33 | 50.77 | 55.06 | 375 |
| 3. 15% sludge by soil weight | 84.80 | 73.53 | 79.20 | 79.17 | 539 |
| 4. 25% sludge by soil weight | 75.18 | 102.76 | 96.92 | 91.62 | 624 |
| *Fertilization with sludge – smolnitza* | | | | | |
| 5. Control – pure soil | 21.70 | 19.91 | 22.12 | 21.24 | 100.00 |
| 6. 5% sludge by soil weight | 56.92 | 58.24 | 59.03 | 58.06 | 273 |
| 7. 15% sludge by soil weight | 77.35 | 55.08 | 63.71 | 65.38 | 307 |
| 8. 25% sludge by soil weight | 82.74 | 80.16 | 78.95 | 80.61 | 379 |
| *Fertilization with manure – leached cinnamon-meadow soil* | | | | | |
| 9. Control – pure soil | 13.95 | 15.93 | 12.74 | 14.20 | 100.00 |
| 10. 5% manure by soil weight | 70.87 | 62.84 | 65.69 | 66.46 | 468 |
| 11. 15% manure by soil weight | 71.60 | 76.75 | 79.23 | 75.87 | 534 |
| 12. 25% manure by soil weight | 75.01 | 76.00 | 76.90 | 75.99 | 535 |
| *Fertilization with manure – smolnitza* | | | | | |
| 13. Control – pure soil | 23.16 | 21.58 | 26.95 | 23.89 | 100.00 |
| 14. 5% manure by soil weight | 74.55 | 78.93 | 79.42 | 76.63 | 320 |
| 15. 15% manure by soil weight | 113.04 | 95.25 | 105.97 | 104.75 | 443 |
| 16. 25% manure by soil weight | 120.18 | 123.60 | 109.00 | 117.59 | 492 |

The results of the yield of silage maize in the different test variants are presented in Table 5. From the data, it can be seen that in all the test variants, using manure from a cow farm and sludge from the WWTP, the corn develops normally. Pronounced differences were observed in the individual variants compared to the controls, both with the participation of the sludge and with the manure.

The results of the experiments with sludge established that at all tested rates of fertilization, an increase in yield was observed. A factor for the higher yields in the cinnamon soil is phosphorus, which is introduced with the sludge.

When fertilizing with manure, a high yield is obtained in the variant with 15% participation of the manure. It is likely that the higher amount of nutrients contained in the smolnitza and the addition of manure has a positive effect on the obtained yield.

In order to assess the degree of influence of the tested factors, a statistical analysis of the obtained results was performed.

The results of the variance analysis of the maize yield data show that the type of soil improver applied has a significant effect on the yields only at the smolnitza. For this soil, when applying manure, the obtained yields of silage maize were statistically proven to be higher than those, when using sludge as a soil improver for each fertilization rate. With both soil improvers, with an increase in the introduced amount of improver, the yield also increases, and the maximum is reached at a norm of 25% of the soil weight. Propably the required rate for obtaining the maximum yield was not reached, especially for the sludge, for which the difference between the yield obtained at a rate of 25% of the soil weight and the yield at 15% was proven to be higher (with a 0.1% difference). It is necessary to conduct an experiment with higher rates of the investigated soil improvers (Tables 6 and 7).

In the case of the leached meadow-cinnamon soil, the yields obtained when fertilizing with 5% manure by weight of the soil were statistically higher, compared to those with the sludge. With an increase in the introduced amounts of improver (15% and 25%), the maize yields are higher when fertilized with sludge. When fertilized with manure, yields increased with increasing fertilization rate, but no significant difference was proven between those obtained at 5%, 15% and 25%. Probably, the optimal rate is already reached, when applying manure 5% of the weight of the soil. When adding sludge, the highest yield is proven to be observed at a rate of 25%. Experiments with higher amounts of sludge are needed.

**Table 6. Smolnitza – statistical analysis**

| Source of variation | Sum of the squares | Sum of the squares, % | Degree of freedom | Average square | F-ratio | Signifi-cance level |
|---|---|---|---|---|---|---|
| Fertilizer type (F) | 3643.763 | 14.31 | 1 | 3643.763 | 102.035 | 0.000*** |
| Norms (R) | 19926.138 | 78.27 | 3 | 6642.046 | 185.995 | 0.000*** |
| F*R | 1317.574 | 5.18 | 3 | 439.191 | 12.299 | 0.000*** |
| Error | 571.374 | 2.24 | 16 | 35.711 | | |
| Corrected Total | 25458.850 | | 23 | | | |

| Fertilizer type | Norms | Average yield g/container |
|---|---|---|
| Manure | 0 | 23.897 |
| | 5 | 77.633 |
| | 15 | 104.753 |
| | 25 | 117.593 |
| Sludge | 0 | 21.243 |
| | 5 | 58.063 |
| | 15 | 65.380 |
| | 25 | 80.617 |

SPD 5% = 10.344
SPD 1% = 14.252
SPD 0.1% = 19.590
SPD – smallest proven differences

**Table 7. Leached cinnamon-meadow soil – statistical analysis**

| Source of variation | Sum of the squares | Sum of the squares, % | Degree of freedom. | Average square | F – ratio | Signifi-cance level |
|---|---|---|---|---|---|---|
| Fertilizer type (F) | 24.261 | 0.13 | 1 | 24.261 | 0.649 | 0.432 - |
| Norms (R) | 17674.599 | 93.75 | 3 | 5891.533 | 157.555 | 0.000*** |
| F*R | 555.023 | 2.94 | 3 | 185.008 | 4.948 | 0.013 * |
| Error | 598.295 | 3.18 | 16 | 37.393 | | |
| Corrected Total | 18852.178 | | 23 | | | |

| Fertilizer type | Norms | Average yield g/container |
|---|---|---|
| Manure | 0 | 14.207 |
| | 5 | 66.467 |
| | 15 | 75.860 |
| | 25 | 75.970 |
| Sludge | 0 | 14.687 |
| | 5 | 55.063 |
| | 15 | 79.177 |
| | 25 | 91.620 |

SPD 5% = 10.585
SPD 1% = 14.584
SPD 0.1% = 20.046
SPD – smallest proven differences

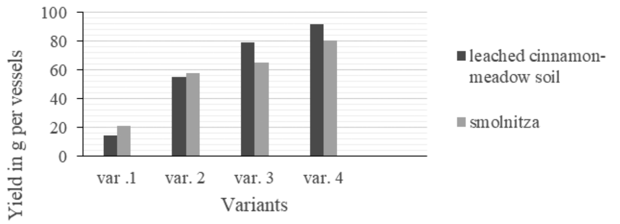


**Fig. 1. Effect of the sludge on the maize yield at two soil types**

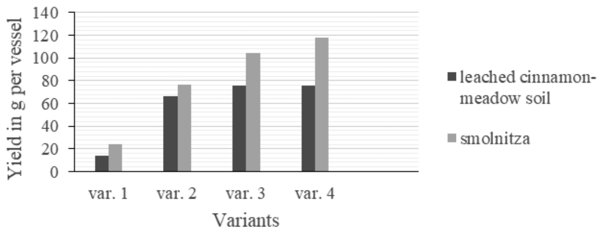


**Fig. 2. Effect of the manure on the maize at two soil types**

Graphically presented, maize yields by soil differences and by variants look like this:

From Figure 1 it can be seen that the sludge had a more favorable effect on the cinnamon forest soil, regardless of the worse chemical and agrochemical properties of this soil. Due to the lack of nutrients, cinnamon forest soil reacts faster.

Figure 2 shows the more favorable effect of manure at the smolnitza. It is a better stocked soil with nutritional ele-

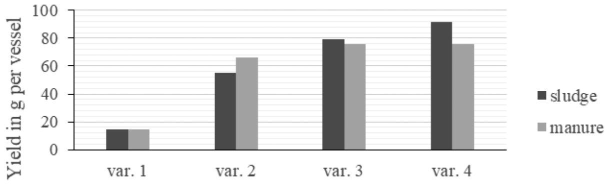


**Fig. 3. Effect of the sludge and the manure on the maize yield at a leached cinnamon-meadow soil**

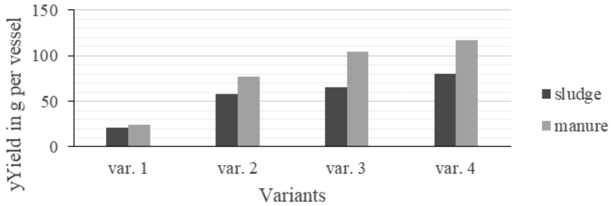


**Fig. 4. Effect of the sludge and the manure on the maize yield at a smolnitza**

ments. The introduction of manure, which is richer in nitrogen, leads to a greater stocking of the soil with this element, and from there to a greater yield.

Figure 3 also present the positive influence of sludge, compared to manure on cinnamon soil. Obviously, the soil type does not have a significant influence on the corn yield, but the type of soil improver is decisive

In Figure 4, weaker results of the influence of the sludge on the smolnitza are observed compared to the manure.

The figures clearly show the positive influence of the sludge, obtained in the biological treatment of wastewater, on the yield of maize on both soil types. The study found that the sludge represents an organic reserve and can be used to maintain and increase off soil fertility.

***Photographic material representing the development of maize during the vegetation period***

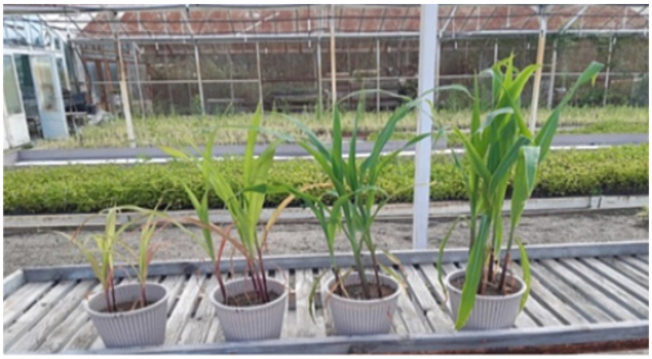

**Photo 1. Development of plants, fertilized with sludge from WWTP on a cinnamon-meadow soil**

**Photo 2. Development of plants, fertilized with sludge from WWTP on a smolnitza**

**Photo 3. Development of plants, fertilized with manure on a cinnamon-meadow soil**

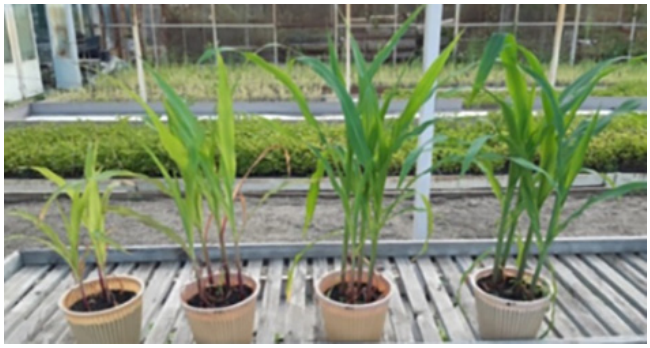

**Photo 4. Development of plants, fertilized with manure on a smolnitza**

Research by Turkish scientists also reached similar conclusions regarding the positive impact of WWTP sludge on plant production. Experiments have been conducted to determine the effect of sludge application on plant yield. The results showed that all yield components of wheat, cabbage and tomato increased significantly with increasing levels of sewage sludge compared to the control (Özyazıcı, 2013).

As we mentioned in the methodological part, when harvesting the maize, plant and soil samples were taken to determine the changes that occurred in the chemical and agrochemical parameters when the sludge and manure were used.

***Characteristics of plant production from silage corn***

Table 8 presents the results of the chemical and agrochemical characteristics of plant samples of maize, harvested at the end of the experiment.

The plant analysis found that with the introduction of sludge, the nitrogen content increases with the increase of the rate in the cinnamon soil, and decreases in the smolnitza. This is probably due to the reduced content of nutrients

**Table 8. Chemical and agrochemical characteristics of the plant production of maize from a vegetation experiment with the participation of WWTP sludge and manure**

| Variants | N, % | P, % | K, % | Ca, % | Mg, % |
|---|---|---|---|---|---|
| Fertilizing with sludge – leached cinnamon-meadow soil | | | | | |
| 1. Control – pure soil | 0.47 | 0.43 | 4.09 | 0.39 | 0.20 |
| 2. 5% sludge by soil weight | 0.58 | 0.46 | 3.95 | 0.45 | 0.42 |
| 3. 15% sludge by soil weight | 0.95 | 0.32 | 3.45 | 0.78 | 0.53 |
| 4. 25% sludge by soil weight | 0.92 | 0.34 | 3.40 | 0.72 | 0.56 |
| Fertilization with sludge – smolnitza | | | | | |
| 5. Control – pure soil | 0.46 | 0.26 | 3.59 | 0.50 | 0.21 |
| 6. 5% sludge by soil weight | 0.30 | 0.27 | 3.60 | 0.52 | 0.22 |
| 7. 15% sludge by soil weight | 0.30 | 0.21 | 2.21 | 0.46 | 0.26 |
| 8. 25% sludge by soil weight | 0.20 | 0.22 | 2.22 | 0.47 | 0.26 |
| Fertilization with manure – leached cinnamon-meadow soil | | | | | |
| 9. Control – pure soil | 0.03 | 0.42 | 3.95 | 0.47 | 0.20 |
| 10. 5% manure by soil weight | 0.02 | 0.43 | 3.92 | 0.46 | 0.22 |
| 11. 15% manure by soil weight | 0.03 | 0.45 | 6.90 | 0.43 | 0.40 |
| 12. 25% manure by soil weight | 0.03 | 0.45 | 6.93 | 0.44 | 0.42 |
| Fertilization with manure – smolnitza | | | | | |
| 13. Control – pure soil | 0.03 | 0.31 | 4.25 | 0.48 | 0.24 |
| 14. 5% manure by soil weight | 0.03 | 0.32 | 4.26 | 0.49 | 0.24 |
| 15. 15% manure by soil weight | 0.03 | 0.34 | 4.78 | 0.37 | 0.26 |
| 16. 25% manure by soil weight | 0.02 | 0.34 | 4.72 | 0.37 | 0.26 |

**Table 9. Content of heavy metals in the plant production of maize from a vegetation experiment with the participation of WWTP sludge and manure, mg/kg**

| Variants | Pb | Cd | Ni | Cr | Cu | Zn | Mn | Fe | As |
|---|---|---|---|---|---|---|---|---|---|
| Fertilizing with sludge – leached cinnamon-meadow soil | | | | | | | | | |
| 1. Control – pure soil | 0.15 | 0.082 | 0.92 | 1.69 | 4.94 | 23.50 | 72.60 | 44.00 | 0.06 |
| 2. 5% sludge by soil weight | 0.30 | 0.064 | 0.79 | 2.06 | 4.70 | 22.60 | 41.50 | 38.00 | 0.05 |
| 3. 15% sludge by soil weight | 0.20 | 0.083 | 0.55 | 1.52 | 5.69 | 45.8 | 46.40 | 69.50 | 0.10 |
| 4. 25% sludge by soil weight | 0.15 | 0.12 | 0.49 | 1.37 | 6.62 | 55.30 | 46.80 | 79.40 | 0.12 |
| Fertilization with sludge – smolnitza | | | | | | | | | |
| 5. Control – pure soil | 0.05 | 0.020 | 0.47 | 0.87 | 2.89 | 12.90 | 38.40 | 59.40 | 0.05 |
| 6. 5% sludge by soil weight | 0.05 | 0.016 | 0.60 | 1.32 | 3.05 | 15.90 | 26.30 | 57.80 | 0.03 |
| 7. 15% sludge by soil weight | 0.04 | 0.035 | 0.64 | 1.34 | 3.36 | 29.50 | 35.30 | 50.50 | 0.03 |
| 8. 25% sludge by soil weight | 0.03 | 0.053 | 0.59 | 1.08 | 4.72 | 22.20 | 19.20 | 80.30 | 0.07 |
| Fertilization with manure – leached cinnamon-meadow soil | | | | | | | | | |
| 9. Control – pure soil | 0.05 | 0.049 | 0.61 | 0.92 | 5.91 | 14.90 | 66.40 | 88.00 | 0.11 |
| 10. 5% manure by soil weight | 0.04 | 0.080 | 0.41 | 0.87 | 4.46 | 18.00 | 42.50 | 42.50 | 0.08 |
| 11. 15% manure by soil weight | 0.04 | 0.205 | 0.40 | 0.81 | 6.37 | 33.50 | 48.60 | 64.80 | 0.11 |
| 12. 25% manure by soil weight | 0.04 | 0.244 | 0.39 | 0.68 | 6.65 | 38.20 | 41.40 | 39.80 | 0.08 |
| Fertilization with manure – smolnizsa | | | | | | | | | |
| 13. Control – pure soil | 0.16 | 0.038 | 0.49 | 0.73 | 2.04 | 14.60 | 31.90 | 42.80 | 0.04 |
| 14. 5% manure by soil weight | 0.23 | 0.051 | 0.56 | 0.92 | 2.62 | 25.10 | 22.30 | 49.20 | 0.07 |
| 15. 15% manure by soil weight | 0.20 | 0.054 | 0.41 | 0.69 | 3.20 | 22.50 | 21.60 | 41.70 | 0.05 |
| 16. 25% manure by soil weight | 0.13 | 0.90 | 0.46 | 0.68 | 4.86 | 39.20 | 35.60 | 62.20 | 0.06 |

**Table 10. Agrochemical characteristics of soil samples after harvesting maize from a vegetation experiment with the participation of sewage sludge and manure**

| Variants | N, % | N ammonium, mg/kg | N nitrate, mg/kg | P, mg/kg | K, % | Ca, % |
|---|---|---|---|---|---|---|
| Fertilizing with sludge – leached cinnamon-meadow soil | | | | | | |
| 1. Control – pure soil | 0.22 | 10.00 | 10.20 | 343 | 0.22 | 0.46 |
| 2. 5% sludge by soil weight | 0.18 | 11.50 | 10.80 | 1080 | 0.31 | 0.42 |
| 3. 15% sludge by soil weight | 0.14 | 10.80 | 11.20 | 1706 | 0.48 | 0.38 |
| 4. 25% sludge by soil weight | 0.12 | 10.00 | 10.80 | 2157 | 0.44 | 0.38 |
| Fertilization with sludge – smolnitza | | | | | | |
| 5. Control – pure soil | 0.17 | 8.00 | 9.20 | 683 | 0.38 | 0.50 |
| 6. 5% sludge by soil weight | 0.20 | 8.00 | 11.20 | 1243 | 0.39 | 0.46 |
| 7. 15% sludge by soil weight | 0.21 | 10.00 | 10.00 | 2142 | 0.46 | 0.49 |
| 8. 25% sludge by soil weight | 0.27 | 10.50 | 11.40 | 3237 | 0.52 | 0.40 |
| Fertilization with manure – leached cinnamon-meadow soil | | | | | | |
| 9. Control – pure soil | 0.21 | 9.95 | 10.60 | 304 | 0.19 | 0.39 |
| 10. 5% manure by soil weight | 0.18 | 10.00 | 20.80 | 760 | 0.25 | 0.35 |
| 11. 15% manure by soil weight | 0.17 | 11.60 | 23.80 | 1552 | 0.33 | 0.35 |
| 12. 25% manure by soil weight | 0.29 | 12.50 | 18.00 | 2386 | 0.37 | 0.33 |
| Fertilization with manure – smolnitza | | | | | | |
| 13. Control – pure soil | 0.15 | 8.20 | 9.20 | 701 | 0.24 | 0.48 |
| 14. 5% manure by soil weight | 0.14 | 9.40 | 10.00 | 1069 | 0.28 | 0.47 |
| 15. 15% manure by soil weight | 0.14 | 10.00 | 10.80 | 2134 | 0.29 | 0.37 |
| 16. 25% manure by soil weight | 0.29 | 13.90 | 11.20 | 2781 | 0.40 | 0.35 |

in the leached cinnamon forest soil and absorption of larger amounts of nitrogen. Phosphorus content in the plant mass decreases very slightly by variants in both types of soil.

When fertilizing with manure, the values of nitrogen and phosphorus do not change with an increase in the rate of fertilization, and potassium increases only with cinnamon soil.

The potassium content is higher with the participation of sludge. Calcium values in plants decreased in the last trial options with manure. About magnesium, a slight increase was observed in plants in both soil types.

The plant analysis of maize from a vegetation experiments with the participation of WWTP sludge and manure found, that no significant changes in the values of the main nutritional elements in the plant production were observed regardless of the different soils (Table 8).

The concentration of heavy metals in the plant production from corn, when fertilized with sludge and manure, in the two soil types do not differ significantly and there is no danger when using thees plants for animal feed (Table 9).

***Soil amendments after completion of vegetation experiment***

The soil tests for the content of the main nutritional elements found that no significant changes were observed when using the two improvers on the two soil types (Table 10).

The measured values for the content of heavy metals in all experimental variants are below the MPC and do not pose a danger, when using the sludge and manure as soil improvers (Table 11).

**Microbiological studies of soil samples from the experimental variants with WWTP sludge**

After the experiments were completed, the soils from the variants with sludge were examined, according to the microbiological parameters – *E. coli,* coliforms, enterococci, *Clostridium perfringens, Salmonella* spp.

In the experiments with the leached meadow cinnamon soil and in the three variants with added sludge (5%, 15%, 25%), all normalized microbiological indicators do not exceed the permissible limits in the regulation. They are found in rather low quantities, including *Clostridium perfringens*. Enterococci maintain low counts. Coliforms are in higher levels, but no correlation is established between the amount of introduced sludge and their number. Coliforms also prevail in the soil itself, as can be seen from the control – variant 1. The highest presence of coliforms is found at 5% sludge. In the variant with 15% sludge by soil weight, these bacteria are found in a lower quantity. In the variant with 25% – they even die (Table 12). The only logical explanation is that mi-

**Table. 11. Content of heavy metals (in mg/kg) in soil samples after harvesting maize from a vegetation experiment with the participation of WWTP sludge and manure**

| Variants | Pb | Cd | Ni | Cr | Cu | Zn | Mn | Fe | As | pH |
|---|---|---|---|---|---|---|---|---|---|---|
| Fertilizing with sludge – leached cinnamon-meadow soil | | | | | | | | | | |
| 1. Control – pure soil | 20.02 | 0.02 | 10.72 | 7.08 | 115.3 | 87.20 | 714.8 | 24138 | 3.11 | 7.87 |
| 2. 5% sludge by soil weight | 24.47 | 0.15 | 11.47 | 7.11 | 121.8 | 111.7 | 723.7 | 24487 | 3.18 | 7.94 |
| 3. 15% sludge by soil weight | 29.50 | 0.28 | 12.10 | 8.02 | 136.3 | 151.3 | 744.4 | 25425 | 3.70 | 7.79 |
| 4. 25% sludge by soil weight | 32.93 | 0.34 | 12.74 | 9.17 | 145.7 | 172.0 | 752.0 | 25348 | 3.56 | 7.77 |
| Fertilization with sludge – smolnitza | | | | | | | | | | |
| 5. Control – pure soil | 39.38 | 0.08 | 25.59 | 16.60 | 49.9 | 90.3 | 1414.6 | 36905 | 9.13 | 7.94 |
| 6. 5% sludge by soil weight | 43.57 | 0.15 | 25.30 | 18.34 | 62.7 | 123.6 | 1283.0 | 36275 | 8.33 | 7.90 |
| 7. 15% sludge by soil weight | 45.07 | 0.29 | 25.02 | 20.01 | 79.5 | 178.9 | 1232.8 | 39750 | 8.57 | 7.82 |
| 8. 25% sludge by soil weight | 49.32 | 0.40 | 26.60 | 20.44 | 105.4 | 234.7 | 1174.4 | 38200 | 10.67 | 7.57 |
| Fertilization with manure – leached cinnamon-meadow soil | | | | | | | | | | |
| 9. Control – pure soil | 20.55 | 0.04 | 11.00 | 8.25 | 120.8 | 83.2 | 755.0 | 2620 | 4.88 | 7.99 |
| 10. 5% manure by soil weight | 18.80 | 0.02 | 10.87 | 8.13 | 114.8 | 73.7 | 743.9 | 24516 | 3.48 | 8.08 |
| 11. 15% manure by soil weight | 19.55 | 0.05 | 9.42 | 9.02 | 111.4 | 86.1 | 716.2 | 22793 | 3.17 | 8.12 |
| 12. 25% manure by soil weight | 22.28 | 0.06 | 10.48 | 7.22 | 109.8 | 96.6 | 750.5 | 24535 | 3.86 | 7.92 |
| Fertilization with manure – smolnitza | | | | | | | | | | |
| 13. Control – pure soil | 35.85 | 0.09 | 26.00 | 19.68 | 52.00 | 100.1 | 1420.1 | 35488 | 8.26 | 8.55 |
| 14. 5% manure by soil weight | 34.94 | 0.07 | 25.42 | 19.56 | 51.30 | 99.40 | 1327.4 | 34387 | 7.98 | 8.15 |
| 15. 15% manure by soil weight | 33.02 | 0.02 | 24.79 | 17.72 | 52.80 | 166.2 | 1233.3 | 36661 | 8.47 | 8.03 |
| 16. 25% manure by soil weight | 32.12 | 0.04 | 24.63 | 19.39 | 57.80 | 143.3 | 1286.3 | 35611 | 8.23 | 8.04 |
| Norms according to the Regulation* | 120.0 | 3.0 | 110.0 | 200.0 | 200.0 | 300.0 | – | – | 25.0 | – |

*Ordinance on the procedure and method for the utilization of sludge from waste water treatment by using it in agriculture, adopted by Decree No. 201 of 04.08.2016, SG No. no. 63 of 2016

**Table 12. Microbiological results of tests carried out on soil samples with different added amounts of sludge as a soil improver**

| Investigated microbiological parameters | *E. coli,* MPN/ g | *Coliforms,* MPN/ g | *Enterococci,* MPN/ g* | *Clostridium perfringens,* MPN/ g | *Salmonella* spp. presence/ absence in 20 g |
|---|---|---|---|---|---|
| Norms according to the Regulation* | up to 100 /g (up to 1. $10^2$ / g) | absent from the regulation | absent from the regulation | up to 300/g (up to 3. $10^2$ / g) | absence (not allowed) in 20 g wet weight |
| Fertilizing with sludge – leached cinnamon-meadow soil – variants | | | | | |
| 1. Control – pure soil | <0.30 | > 110 | 1.6 | 2.3 | absence in 20 g |
| 2. 5% sludge by soil weight | <0.30 | >1.1. $10^4$ (> 11000) | <0.30 | 9.3 | absence in 20 g |
| 3. 15% sludge by soil weight | <0.30 | > 110 | 24 | 24 | absence in 20 .g |
| 4. 25% sludge by soil weight | <0.30 | <0.30 | <0.30 | 9.3 | absence in 20 g |
| Fertilization with sludge – smolnitza – variants | | | | | |
| 5. Control – pure soil | <0.30 | 24 | <0.30 | 110 | absence in 20 g |
| 6. 5% sludge by soil weight | <0.30 | >1.1. $10^3$ (> 1100) | <0.30 | > 110 | absence in 20 g |
| 7. 15% sludge by soil weight | <0.30 | > 110 | <0.30 | >1.1. $10^3$ (> 1100) | absence in 20 g |
| 8. 25% sludge by soil weight | <0.30 | >1.1. $10^4$ (> 11000) | <0.30 | > 110 | absence in 20 g |

*Ordinance on the procedure and method for the utilization of sludge from waste water treatment by using it in agriculture, adopted by Decree No. 201 of 04.08.2016, SG No. no. 63 of 2016 croorganisms are unevenly distributed in this type of matrices, which affects the final results of laboratory analyses.

Similar trends were found in the experiments on the smolnitza. *E. coli*, enterococci and *Salmonella* spp. are not proven. The amounts of *Clostridium perfringens* are normal, with the exception of variant 7, where they are elevated. Coliforms are also in higher quantities, and here too parallelism cannot be proven between the introduced sludge percentage and their amount.

Coliforms, in addition to being found in large quantities in the feces of humans and warm-blooded animals, they can be found in aquatic environments, in soil and vegetation – so soils are their natural biotope.

## Conclusions

As a result of the vegetation experiments conducted to establish the effect of using WWTP sludge as a soil improver, the following conclusions can be drawn:

The analyzed WWTP sludge is rich in nutritional macro- and microelements and can be used as an organic reserve to maintain and increase soil fertility and yields from agricultural crops. It does not contain heavy metals above the MPC and other pollutants that can limit the use of the sludge in agricultural practice.

From the yield data, it can be seen that in the case of corn grown on smolnitza and cinnamon forest soil, an increase in yield is observed with an increase in the rate of fertilization. Higher yields are obtained, when using the sludge on the cinnamon forest soil, while the smolnitza responds better to manure fertilization. The main factor affecting the yield is the rate of fertilization, followed by the type of improver applied and the soil difference.

Agrochemical and chemical studies of plant production show positive changes, when using sludge as a soil improver. The results were compared with those, obtained using the manure. No negative impact on plant production was observed, when using WWTP sludge as a soil conditioner.

The soil research after the completion of the vegetation experiment also found that the sludge can be successfully used as an improver, since they are rich in nutritional macro- and microelements and the values of heavy metals are below the MPC specified in the legislation.

Microbiological studies show that the sludge does not burden the two types of soil, used in the vegetation experiments with fecal bacterial flora, both in terms of indicator microorganisms (*E. coli,* enterococci*, Clostridium perfringens*) and in terms of pathogens – *Salmonella* spp. was not isolated from any sample. For coliforms, soils are their natural biotope.

In summary, the use of sludge in agriculture is recommended due to the disturbed balance of organic matter in the Bulgarian soils and the shortage of organic sources in the country. The obtained results support the thesis that WWTP sludge can be successfully used as soil improver of the most common soil types in Bulgaria, with the aim of increasing soil fertility and agricultural crop yields. With proper management of sewage sludge, after conducting chemical, agrochemical, physico-mechanical and biological studies, according to the requirements of the legislation, they can be used as a suitable soil improver, without damaging plant production and without contaminating the soil. Applied to the soil, sludge become a source of plant-absorbable nutrients

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## References

**Alonso, E., Aparicio, I., Santos, J., Villar, P. & Santos, A**. (2009). Sequential extraction of metals from mixed and digested sludge from aerobic WWTPs sited in the south of Spain. *Waste Management, 29* (1), 418-424.

**Colón, J., Alarcón, M., Healy, M., Namli, A., Ponsá, S., Dilek Sanin F. & Taya, C.** (2017). Chapter 14: Producing sludge for agricultural applications. In: Innovative Wastewater Treatment & Resource Recovery Technologies. https://doi.org/10.2166/9781780407876_0296 – on line eBooks from Iwa Publishing

**Georgieva, V., Marinova, S. & Sidzhimov, M.** (2017). Microbiological studies and assessment of sludge from wastewater treatment plants, in order to use them as soil improvers. *Bulgarian Journal of Public Health, 9* (1), 16-24.

**Lamastra, L., Suciu, N. A. & Trevisan, M.** (2018). Sewage sludge for sustainable agriculture: contaminants' contents and potential use as fertilizer. *Chem. Biol. Technol. Agric. 5* (10); https://doi.org/10.1186/s40538-018-0122-3

**Marinova, S.** (2000). Determination of the effect of the sludge from waste water treatment station near Sofia-city as Fertilizer. Int. Symposium on Desertification. Konja, Turkey, 333-338.

**Marinova, S**. (2002). Agroecological assessment of sludge from wastewater treatment plants and basic approaches to their use in agriculture. Habilitation thesis for "Professor". Sofia, Bulgaria (Bg).

**Marinova, S.** (2008). Sludges from wastewater treatment plants and rules for their utilization. *Publisher PublishSySet-Eco,* Sofia (Bg).

**Marinova, S. , Petrova, l., Chipilsla, L., Chatalbasheva, M. & Bratinova, S.** (2000). Agro-ecological assessment of the sludge from the Sofia WWTP with a view to its use in agriculture. Agreement with Sofia Municipality, (Bg).

**Nascimento, A. L., Souza, A. J., Andrade, P. A. M., Andreote, F. D., Coscione, A. R., Oliveira, F. C. & Regitano, J. B.** (2018). Sewage Sludge Microbial Structures and Relations to Their

Sources, Treatments, and Chemical Attributes. Frontiers in Microbiology. 9: 1462.doi: 10.3389/fmicb.2018.01462.

**Özyazıcı, M. A.** (2013). Effects of sewage sludge on the yield of plants in the rotation system of wheat-white head cabbage-tomato. *Eurasian Journal of Soil Science, 2,* 35–44.

**Pallavi, B., Nupur, M., Anuradha, S. & Pradeep, B.** (2015). Recycled sewage sludge: a step to sustainable agriculture. *Int. J. Chem. Sci., 13*(4), 1611-1620.

**Romanos, D., Nemer, N., Khairallah, Y. & Abi Saab, M. T. (2019).** Assessing the quality of sewage sludge as an agricultural soil amendment in Mediterranean habitats. *International Journal of Recycling of Organic Waste in Agriculture*, *8*, 377-383.

**Sidjimov, M., Metodiev, V., Metchkueva, L., Stankova, D., Paunova, G. & Shanova, L.** (2013). Heavy Metals in Sludge from Urban Waste Water Treatment Plants. *Journal of Balkan Ecology, 16* (2), 179-189.

**Stankova, D., Paunova, G., Georgieva, V. & Georgieva, R.** (2021). Study of sludge from wastewater treatment plants – agrochemical parameters and content of heavy metals and arsenic. *Bulgarian Journal of Public Health, 13* (3), 69 – 77.

**Tsolova, V. & Marinova, S.** (2005). Soluble heavy metal compounds contained in sewage treatment plant sludges. *Soil Science, Agrochemistry and Ecology, XL* (3), 22-24 (Bg).

**Tytła, M.** (2020). Identification of the Chemical Forms of Heavy Metals in Municipal Sewage Sludge as a Critical Element of Ecological Risk Assessment in Terms of Its Agricultural or Natural Use. *International Journal of Environmental Research and Public Health, 17,* 4640.

**Zhang, X., Wang, X. & Wang, D.** (2017). Immobilization of Heavy Metals in Sewage Sludge during Land Application Process in China: A Review, *Sustainability, 9(11).*

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