

# THE DISTRIBUTION OF MACRO- AND MICRONUTRIENTS IN MAIZE WITHIN SEPARATED DIGESTATE FERTILIZING (DIGESTATE FIBRE AND DIGESTATE LIQUOR): FIELD TRIAL

Jana KODYMOVÁ<sup>1</sup>, Hana ŠVEHLÁKOVÁ<sup>2</sup>, Miroslav KYNCL<sup>3</sup>, Magdaléna BÁRTKOVÁ<sup>4</sup>

<sup>1</sup> Ing. Ph.D., Department of Environmental Engineering, Faculty of Mining and Geology,  
VŠB – Technical University Ostrava  
17. listopadu 15, Ostrava, tel. (+420) 59 732 5756  
e-mail jana.kodymova@vsb.cz

<sup>2</sup> Ing., Department of Environmental Engineering, Faculty of Mining and Geology,  
VŠB – Technical University Ostrava  
17. listopadu 15, Ostrava, tel. (+420) 59 732 3887  
e-mail hana.svehlakova@vsb.cz

<sup>3</sup> prof. Dr. Ing., Department of Environmental Engineering, Faculty of Mining and Geology,  
VŠB – Technical University Ostrava  
17. listopadu 15, Ostrava, tel. (+420) 59 732 3556  
e-mail miroslav.kync1@vsb.cz

<sup>4</sup> Ing., Department of Environmental Engineering, Faculty of Mining and Geology,  
VŠB – Technical University Ostrava  
17. listopadu 15, Ostrava, tel. (+420) 59 732 3106  
e-mail magdalena.bartkova.st@vsb.cz

## Abstract

Along with the current increase in the number of biogas plants, huge amounts of digestate, i.e. waste products, are being generated. The common practice in the Czech Republic is to plough the digestate into the land. In our field trial, we compared the fertilizing effects of standard fertilizers applied in the Czech Republic with digestate as the only fertilizer (e.g. digestate fibre and digestate liquor) in real agricultural conditions to find out whether a comparable amount of nitrogen is used in accordance with European legislation. The impact of separated digestate fibre and separated liquor on the soil fertility and quality was observed on the basis of the distribution of macro- and micronutrients in maize. The evaluation of growth increments in maize suggests that the fertilizing effects of digestate liquor or digestate fibre do not match the standard fertilizer in agriculture, but especially digestate liquor is comparable with other mineral fertilizers. Our field trial also shows that digestate liquor is a better fertilizer than digestate fibre, which may be explained by more appropriate ratios of available nutrients in digestate liquor. Digestate fibre may be recommended mainly for the aeration of clayey soil, but is not sustainable as fertilizer.

**Key words:** biogas plant, Czech Republic, digestate, maize, micronutrients, soil

## 1 INTRODUCTION

Anaerobic digestion is an effective method of biomass processing, by which the biogas plants, the organic matter, decomposes in the absence of oxygen from air to give two valuable products, which are biogas and digestate. Biogas is a very useful source of renewable energy, while digestate is considered a valuable bio-fertilizer. Digestate can be considered a stabilized material containing undigested biomass and dead microorganisms that have entered into the process of anaerobic digestion [1].

Digestate from each biogas plant is unique, its composition is affected by many factors, particularly the type of processed input materials, pretreatment method, process conditions (operating temperature in fermentors, solids content), and the method and time of storage [2]. In the Czech Republic, there are more than 500 of biogas plants in operation. One biogas plant with an installed capacity of 1 MW produces an average of 100 m<sup>3</sup> of digestate per day [3].

There are several methods for the separation of digestate which are used to separate the solid component from the liquid using different techniques. These include, in particular, sedimentation, filtration, centrifugation,

and pressing. Mechanical separation using a belt or screw presses, drum screens, decanting centrifuges, etc., or their combination, is the most widely used method. In some cases, prior to the fermentation flocculants or coagulants were applied [4] for better separation of the solid fraction of the digestate. Such chemical separation increases efficiency and allows the separation of certain nutrients, together with a solid fraction (e.g., phosphorus). The resulting solid fraction is more or less dried and after subsequent stabilization becomes an ideal medium with a higher content of phosphorus and stable organic matter for improving soil properties. The liquid fraction contains most of potassium and inorganic nitrogen from the original digestate. This fraction may be further adjusted by ammoniacal stripping or by membrane filtration [5].

Digestate can be applied directly as fertilizer on agricultural land or can be used to produce enriched compost, which can be used either directly as a fertilizer, or further processed to manufacture various substrates [6-7].

Digestate is often considered as an organic fertilizer, but it contains stable organic matter, so it is rather a mineral fertilizer [8] and Babička [9] also confirmed this conclusion. The use of digestate as fertilizer is limited primarily by hygiene requirements, the presence of hazardous elements and salinity. As for a number of fermentation residues, higher concentrations of Cu and Zn were found, which did not comply with legislative requirements [10]. The export of digestate as fertilizer to agricultural land is governed by the Council Directive 91/676/EEC [11].

## 2 MATERIALS AND METHODS

The test site was a field located in Stonava, the Czech Republic. The farm is situated in a climatic region MW7 (short summers which are moderately cold and moderately dry; mild spring, mild autumn) [12]. It has an area of 120 ha and is commonly used for agricultural purposes. The field was divided into 3 subplots of 50 m<sup>2</sup>. One of the subplots was a reference field, where a standard fertilizer was used in accordance with the agronomic practice of the Stonava farm.

The soil characteristics before the field assay (April 2014) are given in Table 1. All the analyses herein were carried out in a Czech accredited laboratory.

**Table 1: Characteristics of the subplots and reference field (including methods used)**

Element	Unit	Two subplot for experiment (DFF and DLF)	Reference field	Used method
total dry matter	%	82.84	81.57	gravimetry
pH (CaCl <sub>2</sub> )		6.08	6.77	potentiometric
available Ca		1510	2028	flame atomic absorption spectrometry - Mehlich III (the soil is leached by Mehlich III solution, which is composed of: 0.2 mol.l <sup>-1</sup> CH <sub>3</sub> COOH, 0.015 mol.l <sup>-1</sup> NH <sub>4</sub> F, 0.013 mol.l <sup>-1</sup> HNO <sub>3</sub> , 0.25mol.l <sup>-1</sup> NH <sub>4</sub> NO <sub>3</sub> and 0.001 mol.l <sup>-1</sup> EDTA.)
available K	mg/kg of dry matter	319	267	flame emission spectrometry - Mehlich III
available Mg		190	201	flame atomic absorption spectrometry - Mehlich III
available P		60	268	photometrically - Mehlich III
N-NH <sub>4</sub> <sup>+</sup>		2.92	12	photometrically (resp. by titration)
NO <sub>3</sub> <sup>-</sup>		23.8	62.1	potentiometry using ion-selective electrodes
mineral nitrogen		26.7	74.1	calculation

In the reference field (RF), standard fertilization was applied in the preparation (urea (46% N) and Polidap (18% N, 46% P)), leave fertilization – LAV 27% N, Mg 3%, 4% CaO, herbicide protection (Bolton Duo and Predict), and herbicides (Adengo and Akris). In the subplots, two different fertilization scenarios were tested: DF as the only fertilizer and DL as the only fertilizer.

Mechanical dewatering by centrifugation is the simplest separation method of digestate solid and liquid substances. Therefore, we centrifuged the whole digestate in an industrial centrifuge BEHO at 1200 rpm (the content of dry matter in DL and DF see in Table 2).

The composition of digestate is very closely related to the composition of digester feedstock. In our experiment, the digestate was from biogas plants which process their own waste products (manure, silage, and Corn Cob Mix). In summer, the feedstock may include hay silage, fresh sorghum, fresh grass, and the whole digestate, while in autumn beet pulps and fresh corn are added. Table 2 below gives the composition of DF and DL.

**Table 2: Composition of DL and DF (including the used methods)**

Element	Unit	DF	DL	Used method
Al	mg·kg <sup>-1</sup> of dry matter	349	378	flame atomic absorption spectrometry
Cu		24.1	125	
Fe		1140	1940	
Mn		135	283	
Pb		<2.50	<2.50	
Zn		167	1050	
Ca	g·kg <sup>-1</sup> of dry matter	11.7	30.1	photometrically
Mg		7.54	7.18	
K		20.7	129	
P		12.1	10.4	
total dry matter	%	12.01	2.09	gravimetry
pH (CaCl <sub>2</sub> )		7.21	9.38	potentiometric
available Ca	mg·kg <sup>-1</sup> of dry matter	4329	5736	flame atomic absorption spectrometry – Mehlich III
available K		19130	109100	flame emission spectrometry – Mehlich III
available Mg		3992	2967	flame atomic absorption spectrometry – Mehlich III
available P		10260	5730	photometrically – Mehlich III
available-Fe		119	135	flame atomic absorption spectrometry – Mehlich III
available Al		<10	<10	
available Mn		53	130	
combustible substances	% in dry matter	86.2	69.3	gravimetry
C:N ratio		13	2	calculation
N-NH <sub>4</sub> <sup>+</sup>	mg·kg <sup>-1</sup> of dry matter	6110	102000	photometrically (resp. by titration)
NO <sub>3</sub> <sup>-</sup>		2170	<2,50	potentiometry using ion-selective electrodes
mineral N <sub>2</sub>		8280	102000	calculation
Total N <sub>2</sub>	% of dry matter	3.3	14.6	calculation

Hybrid maize (Hybrid Kws Agrovitallo) was grown on the test field as well as the RF. We applied eight fertilizing doses of DL and DF from April to September 2014. Within each dose, machinery applied 160 l of DL on digestate liquor field (DLF) and 40 l of DF on digestate fibre field (DFF) into the soil. These amounts correspond to having centrifuged 200 l of the whole digestate.

The DL and DF doses, which were applied to the research area, comply with the Council Directive 91/676/EEC (eutrophication risk) and fertilization limits for each crop (in the case of maize there is a determined value of 230 kg N/ha per vegetation period).

We discussed the application of the fertilizer with an agronomist from the Stonava farm and proceeded according to his experience. Each fertilization was carried out every 14 days between 29/6 and 31/8 with respect

to current weather conditions. The DL was poured onto the surface, and then was incorporated into the soil with a hoe; the DF was incorporated directly to the roots of plants with a hoe. We also eliminated weeding regularly, because herbicide products were not applied in the subplots.

After finishing the field trial, we sampled the soil again from each subplot and RF. We randomly collected 20 plants including the root system from each of the three fields. We randomly selected again two plants from each set and analysed them. Micro- and macronutrients were determined in the roots, stems and leaves, grains, and in the cobs. We also measured the height of plants and the length and weight of the cobs.

### 3 RESULTS AND DISCUSSION

We observed and measured soil micronutrients twice per season (before seeding in April 2014 and before the harvest in September 2014).

The texture of the soil is clay-loam with optimal soil acidity (optimum for maize is 5.5 to 6.8), see Table 3 below showing the exchangeable acidity of soil ( $\text{CaCl}_2$ ). Table 5 shows agrochemical characteristics of the subplots and reference field at the beginning of our experiment.

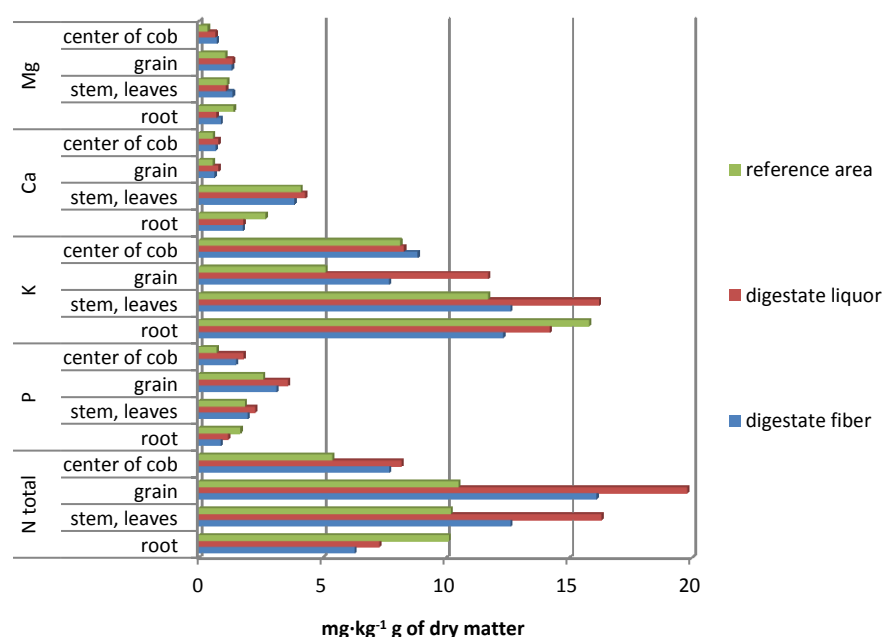
**Table 3: Exchangeable soil reactions in the digestate fibre field (DFF), digestate liquor field (DLF), and the reference field (RF)**

month/year	DFF	DLF	RF
4/2014	6.08		6.77
9/2014	6.17	6.52	6.22

In general, a slightly acidic reaction supports an uptake of many important macro and micronutrients. The differences in pH at the beginning of the experiment between the experimental fields (DFF and DLF) and the RF were caused by the fertilization of RF, which adjusts the pH value to an optimum value for maize. The analyzed soil elements may occur in several chemical forms in the soil. The molecules change forms, achieving dynamic equilibrium that shifts according to certain soil conditions, including pH, texture, soil aeration, the presence of other ions etc.

#### Distribution of nutrients in maize

The nutrients which a plant receives from the soil and subsequently distributes into various plant tissues are dependent on their availability in the soil, actual needs of the plant, its age and physiological condition. Vaněk et al. [13] identified that the nutrient uptake during a growing season is not linear, and before creating its panicles, maize receives generally 75% of nutrients. Figure 1 summarizes the distribution of macronutrients in the maize body. The following values are the results of a mixed sample of 20 plants from each of the three fields (DFF, DLF, and RF).



**Figure 1: Distribution of macronutrients in maize**

Nitrogen N-NH<sub>4</sub> is stored in roots and in such form rarely passes into shoots; it is usually transformed into amino acids already in the roots and thus transported. On the contrary, N- NO<sub>3</sub> is very mobile in roots and through the xylem is transported into aboveground biomass quite intensively [14].

Regarding the distribution of **total nitrogen**, in all fields, the highest relative concentration is in the grains (29 - 38%), followed by its concentrations in the leaves and stems (28 - 35%), the centres of the cobs (16 - 18%), and the roots (14 - 15%). Plants on RF exhibit the same relative concentrations in the roots, and stems and leaves, and the concentration in the centres of the cobs is 15%. The highest absolute concentration in the roots was measured in RF (10.11 g·kg<sup>-1</sup>) in the leaves and stems, grains, and the centres of the cobs in DFF.

Plants absorb **phosphorus** in the form of H<sub>2</sub>PO<sub>4</sub><sup>-</sup> or HPO<sub>4</sub><sup>2-</sup> as phosphates have structural and energetic functions in plants. According to [15] differences between inorganic phosphorus and total phosphorus are the greatest in young leaves, which contain a relatively large amount of organic P in the form of nucleic acids. Skowronska and Filipek [16] monitored the phosphorus content in maize tissues fertilized with NPK fertilizers. They found the highest content of phosphorus in the grains (5.4 g·kg<sup>-1</sup>), followed by leaves (1.3 g·kg<sup>-1</sup>), stems (0.6 g·kg<sup>-1</sup>), cobs (0.5 g·kg<sup>-1</sup>), and roots (0.4 g·kg<sup>-1</sup>).

The relative concentrations in DFF and DLF (grains – 41-42%, stems and leaves – 26%, centres of cobs – 20%, roots – 12-13%) correspond to the conclusions of authors Richter [15] (1994) and Skowronska and Filipek [16]. In RF there were slightly different concentrations (grains 38%, stems and leaves 27%, roots 24%, centres of cobs 11%). The highest absolute concentration was measured in the grains from DLF (3.61 g·kg<sup>-1</sup>).

The intake of **potassium** is influenced by interactions of an antagonistic character. Increasing concentrations of potassium decrease Mg<sup>2+</sup>, Ca<sup>2+</sup>, NH<sub>4</sub><sup>+</sup>, Zn<sup>2+</sup>, Mn<sup>2+</sup> and stimulate the intake of NO<sub>3</sub><sup>-</sup>, H<sub>2</sub>PO<sub>4</sub><sup>-</sup>, Cl<sup>-</sup>, SO<sub>4</sub><sup>2-</sup>. White [17] found that potassium is taken up in large quantities by plants, is highly mobile within plant vascular systems and plays an essential role in a number of metabolic functions.

The highest relative concentration was measured in the stems and leaves in DLF (32%) and DFF (30%), followed by the concentration in the roots (28% - 30%). In DFF, the concentrations were: centres of cobs – 21% and grains 19%; and in DLF: grains – 23% and centres of cobs 17%. The highest relative concentrations in the RF were: roots – 39%, stems and leaves – 29%, centres of cobs – 20%, and grains – 12%. The highest absolute concentration was measured in the stems and leaves in DLF (16.2 g·kg<sup>-1</sup>). A similar potassium concentration was measured in the roots in RF (15.8 g·kg<sup>-1</sup>).

**Calcium** intake is affected by anions. NO<sub>3</sub><sup>-</sup> has the biggest impact, followed by Cl<sup>-</sup> and tailed by SO<sub>4</sub><sup>2-</sup>. On the contrary, increased cation contents restrict the intake of Ca. Calcium moves in the plant through its xylem. Plants require its supply for the whole growth duration.

Calcium has an essential signalling, physiological, and regulatory role for plant fertility. It is present in three forms: (1) covalently bound calcium, (2) loosely bound calcium – typically associated with fixed and mobile anions (ionic bonding); and (3) cytosolic free calcium – an important secondary messenger in cell signalling [18]. Richter states that a maize grain has 3.4% and other parts 96.6% (mainly in vegetative plant organs).

If we evaluate our experiment, the highest relative concentration was observed in the stems and leaves of the plants of all study areas (52 - 57%); the tissues of root and cob centres (7 - 10%) have approximately the same proportion in biomass. The highest absolute concentration was measured in the stems and leaves in DLF (4.3 g·kg<sup>-1</sup>).

**Magnesium** is an essential element of chlorophyll combined as the central atom in the porphyrin core. Richter and Hlušek state that higher levels of magnesium can be found mainly in older leaves. The reception of Mg<sup>2+</sup> antagonistically relate to K<sup>+</sup>, NH<sub>4</sub><sup>+</sup>, Ca<sup>2+</sup>, Mn<sup>2+</sup>, H<sup>+</sup> [14].

According to the data presented in Figure 1 it is clear that most of Mg is in the aboveground biomass, in contrast to Ca, the concentration of Mg increases in generative tissues and decreases in root tissues. The highest relative concentration was observed in the grains in DFF and DLF (31% - 37%) and in the roots in RF (35%). The root tissues in DLF and DFF contain lower concentrations than in RF. The lowest relative concentration is found in the cob centres. The highest absolute concentration was measured in the roots in RF (1.4 g·kg<sup>-1</sup>).

We can say that the distribution of relative concentrations of macronutrients P, Fe, and Mn of monitored plant tissues is identical in all three types of the fertilizer management. DFF and DLF also have the same relative content of total N, K, and Al, but quite different in RF. DLF and RF also have an identical relative concentrations of Cu, and RF conforms to DFF in the relative concentration of Pb. All monitored macronutrients show higher concentrations in the aboveground biomass. In vegetative organs of aboveground biomass, K, Ca, and Mg were significant. The plants from the RF area showed higher concentrations of K and Mg also in the roots.

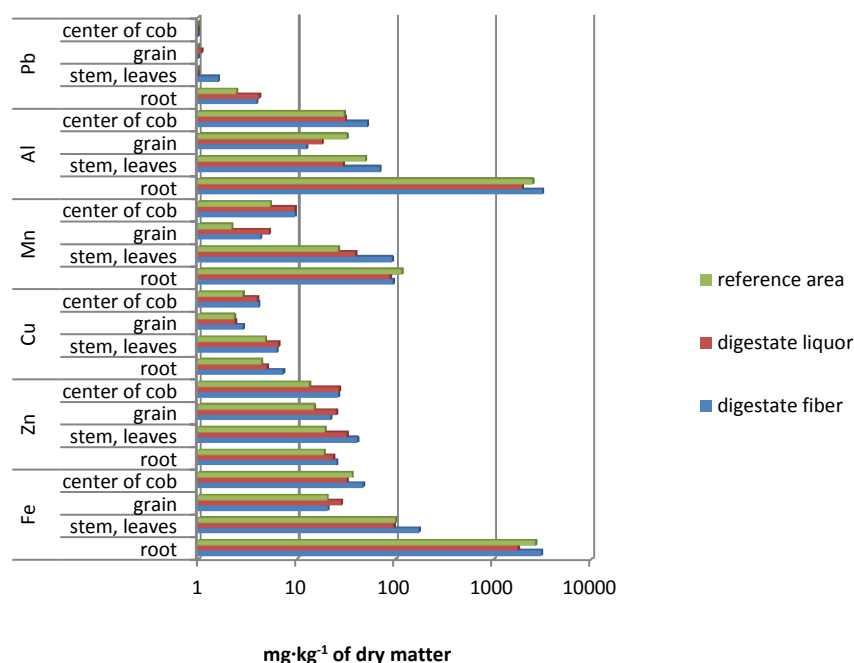


Figure 2: Distribution of micronutrients in maize

The above given data show that the distribution of **iron** in plant tissues is considerably uneven in favour of underground biomass. The highest absolute concentration in the plant roots was measured in DFF (3100 mg·kg<sup>-1</sup>). The highest relative concentration (in the distribution of iron in the body of the monitored plant) is exhibited in the root tissues in RF (94%), relative concentrations of iron in the plant roots in DLF and DFF are also above 90%. The roots thus appear to be the important reservoirs of iron in the plant.

Stem and leave tissues exhibit the second highest concentration, which corresponds to the important function of iron in the synthesis of chlorophyll, nucleotides and in the respiratory chain [19].

The highest absolute concentration in the leaves and stems was measured in the plants from DFF; the relative concentration was 5% in the leaves and stem tissues of plants in DFF and DLF, and 4% in the plants from RF. The remaining iron is stored in the grains and the centres of the cobs.

The distribution of **zinc** (Zn<sup>2+</sup>) in maize tissues is uniform in all three fields. Its content is around 20-36% in all examined tissues. The aboveground biomass has a higher concentration, especially the stem and leaf tissues, which corresponds to the function of Zn as cofactor photosynthetic enzymes in carbohydrate metabolism and in the production of auxin and gibberellin. A significant content of Zn is also in the roots.

Sekara et al. [20] studied the distribution of Zn in roots, leaves, cobs, stems, and grains and found the highest concentration in roots and leaves.

The highest relative concentration occurs in the tissues of plant stems and leaves in DFF (also with the highest absolute concentration of 41.5 mg·kg<sup>-1</sup>), underground biomass accounts for about ¼ of its content. Richter [15] states that Zn accumulates in roots at of Zn contents in soil, from roots otherwise are quite rapidly translocated into the shoot tissues. He further states that Zn is almost immobile in older leaves. The uptake of Zn by roots is also negatively influenced by the competition with Fe, Mn, Ca, Mg; its content corresponds to the content of organic matter, content of clay, redox potential, pH, macronutrient content (P), the content of Ca in the soil [15] and [21]. In our case, the content of Zn in plant tissues in all three fields was similar, which may be explained by its similar concentrations in the soil and low stocks in the fertilizers.

The high content of **copper** is also typical for the aboveground biomass. Copper occurs especially in chlorophyll, is also a part of many enzymes, and is important in the metabolism of proteins and carbohydrates [15]. Sekara et al. [20] and [22] reported that the highest concentrations are measured in roots and leaves, the smallest in stems.

Concerning the distribution of copper in our experiment, the roots show the highest relative concentration (about 30% in all three areas), followed by the leaves and stems (31-37%), the centres of the cobs (about 20%),



and the grains exhibit the lowest content. The highest absolute concentration was measured in the roots of plants from DLF (7.28 mg·kg<sup>-1</sup>).

The **manganese** content is relatively volatile. The highest concentrations occur in testa, leaves, and embryos of seeds. Poniedziałek et al. [23] focused on the distribution of manganese in maize and found that the highest Mn content was in roots, followed by leaves, grains, husks, and stems.

In our case, the distribution is similar to the above mentioned results. Plant roots exhibit the highest relative concentration (RF - 79%, DLF - 64%, DFF - 49%), followed by leaves and stems, the centres of cobs, grains (or, grains, centres of cobs in RF). The highest absolute concentration was measured in the roots in RF (117 mg·kg<sup>-1</sup>).

Plants accumulate **aluminium** in the roots. Higher concentrations of Al slow their growth. Al also inhibits the uptake of other ions, mainly of phosphorus, which binds tightly and thus causes its deficiency. The plants with toxic concentrations of Al in the aboveground parts exhibit generally simultaneously a high content of Fe and Mn, but a low content of Ca and Mg [15]; its high content also inhibits the intake of nitrate [24].

Aluminium is an element with the largest absolute concentrations in the root tissues which is probably connected with its high supply in the soil (the fertilizers used have minimum concentrations). Its share in the roots is 96 % in all study areas, in the monitored parts of the shoots is 1-2%.

**Lead** occurs in plants in normal concentrations of 2 to 3 mg·kg<sup>-1</sup>. Lead is almost immobile in the soil. It firmly binds in the soil sorption complex, and exhibits low bioavailability. In plants, lead is a little mobile too [15]. High concentrations of lead disrupt the metabolism of calcium and negatively affect photosynthesis; the plant species resistance to high concentrations is very variable. Maize is considered a relatively resistant species [25]. The distribution of lead in maize was dealt with by [20] who found the highest concentrations in leaves and roots.

Relative concentrations of lead were the highest in the root tissues in all three areas (approximately 60%). The highest absolute concentration in plant roots was in DLF (4.17 mg·kg<sup>-1</sup>). We measured the second highest relative concentration in the tissues of the leaves and stems in DFF (25%) and DLF (22%). In the case of DLF, the grains exhibit the second highest relative and absolute concentrations (15%; 1.08 mg·kg<sup>-1</sup>). The relative concentration of Pb in the centres of the cobs was 15% in RF and 8% in DFF; the rest of lead was in the grains. The concentration of lead in DLF is distributed as follows: 13% in the leaves and stems, and 11% in the centres of the grains.

The micronutrients Fe, Mn and Al, Pb showed significantly higher concentrations in the underground biomass, whereas Zn and Cu in the aboveground biomass were less abundant.

### Growth characteristics of maize

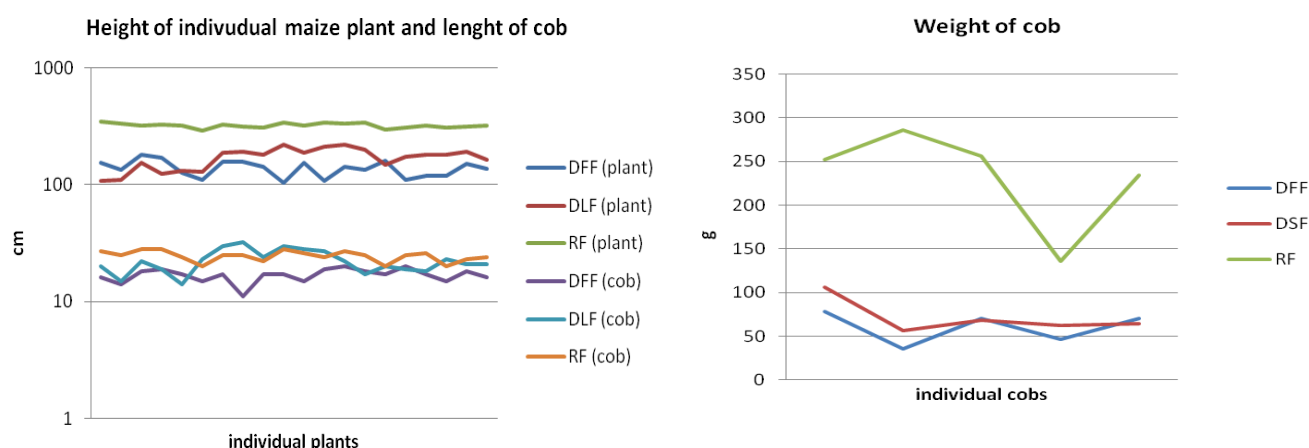


Figure 3: Increments in maize

Figure 3 shows that the plants from RF achieve higher yields than the plants in DFF or DLF. Moreover, we can see that the plants from RF achieve higher yields than the plants in DFF or DLF.

Our experiment also showed that DL and DF, at this moment, cannot fully substitute a standard fertilizer. The fertilizing effect of DF can be compared with mineral fertilizers [26-27], because they are a valuable source of mineral nitrogen (see Table 2). DL has a better fertilizing effect than DF due to the higher content of nitrogen

(mainly mineral nitrogen) and the lower content of Fe and Al in the roots. The experimental part brought a significant finding that the organic material of DF is stable and therefore it is nearly worthless as an organic fertilizer [8].

The results of our research could be influenced by environmental factors (especially soil conditions). Results could also be distorted by edge effect, because DFF and DLF were situated at the edge of the field and nearby a forest. That is why we recommend a better-situated area with larger subplots for further research. It would be more appropriate to determine the amount of nitrogen of each processed dose for DL and DF, but it was not possible due to process and finance conditions, so the amount of nitrogen (and any other elements) determined only the form of mixed samples of individual doses. We applied no herbicidal preparations on DFF and DLF and it may have caused lower competitiveness of maize mainly at the beginning of the growing season, although we removed weeds regularly.

#### 4 CONCLUSION

At present, the vast majority of digestate from agricultural biogas plants is used by farmers for application on their land. The problem is its liquid form, through which, when applied to the soil, contamination of surface water with nitrogen compounds may occur.

In our experiment, the possibility of using digestate as fertilizer on the basis of field cultivation of maize during growing season was evaluated. Digestate was separated by centrifugation into two fractions – digestate liquor and digestate fibre. Both of these fractions were applied to the soil in a field trial and the results were compared with artificial fertilizers. The analyses of each fraction of the digestate, soil, and phytomass were carried out by an accredited laboratory. The results showed that one of the positive aspects of digestate liquor and digestate fibre is their effect on increasing the pH value of the soil after its application (in contrast to the artificial fertilizer). Digestate enriches the soil with macrobiogenic and microbiogenic elements, but cannot fully replace chemical fertilizers.

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