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Evaluation of mixed organic fertilizers based on solid digestate

Master´s thesis

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Declaration

I hereby declare that the Diploma Thesis “Evaluation of mixed ecological fertilizers based on solid digestate” I have elaborated completely by myself and under the supervision of Ing. Tatiana Ivanova, Ph.D. I also declare that all sources of information and data that have been used are listed in the references or they are my own outcomes of the research.

Prague, 27th April 2017 …………………………

Barbora Seidlová

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Abstract

Organic fertilizers represent an irretrievable role in a plant production system. In term of sustainable maintaining soil fertility in the long term, it cannot be successfully accomplished without intake of organic matter into the soil.

The aim of the study was to determine the properties and optimal composition of mixed fertilizers and further to evaluate the impact of these fertilizers on selected plants´ production.

The main methodological steps of the research were to determine chemical composition of all compounds involved in the study. Additional was to establish a weight ratio of fertilizers to each sample and to set up greenhouse trial. Practical part of research was focused on measurement of plants´ growth, subsequent processing of the plants in laboratories and determination the plants´ growth and yield by statistical analysis.

For the trial were determined five substrates with a various fertilizers´ composition. The weight ratios of the substrates were established according to plants´ requirements for nutrients intake. The selected tested crops were: Garden pea (*Pisum sativum L.*) and Spring barley (*Hordeum vulgare L.*). The statistically significant results for crops were evaluated.

The plants´ growth of garden pea had almost identical progress. Substrate with all fertilizers had the lowest average plants´ growth for whole observed period. Substrates with lowest fertilizer composition had highest plants´ growths during whole observed period.

The plants´ growth of spring barley had progressive growth for whole observed period. Enriched substrates had higher plants´ growths than the prior substrates in same week except for plants in substrate with separate, ash and poultry excrements. The growths within a week as well as between the weeks were statistically confirmed.

Highest grain yields for spring barley had substrates with separate and with separate and poultry excrements. Lowest yield indicated plants from substrate with separate and ash that was even lower than in non-fertilized variation.

All substrates had an effect on plants´ growth. The contribution of addition of ecological fertilizers was confirmed in form of increased nutrients´ content in the soil and positively influenced plants´ growth and plants´ yields. Further contribution represent a cheap nutrient input that improve crop yields.

Key words: ecological fertilizers, optimal composition of fertilizers, separate, straw ash, poultry manure, spring barley, garden pea

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1 Introduction

Intensive agriculture, which requires substantial amounts of soluble fertilizers and other inputs, has considerably increased global food production (Matson et al., 1997; Tilman et al., 2002). Nevertheless, it has also increased the risk of negative consequences on ecosystems, climate or public health (Matson et al., 1997; Tilman et al., 2002; Delgado and Scalenghe, 2008; Tscharntke et al.,2012).

Organic fertilizers play an irreplaceable role in plant production system. In terms of maintaining or improving soil fertility in the long term, it cannot be successfully managed without intake of organic matter into the soil. The main sources of organic substances are post-harvest residues and root residues (it makes up to 60 % of the total needs) and further organic fertilizers. (Vanek et al., 2012).

Organic fertilizers are considered as cheap nutrient inputs that may contribute to improving of crop yields (Maldonado-Montiel et al., 2003).

An imbalanced application of N, P and K. can occur in the soil. Also deficiency of secondary nutrient sulphur and micronutrient zinc can be widespread in the developing countries with poor soils and lack of other nutrients. While attempts are being made to increase the fertilizer use in states where level of application is low, the focus is on developing and promoting secondary and micronutrient customized fertilizers. To increase the use efficiency of nitrogen it is necessary to supply in adequate quantity for specific plant. Therefore, more efficient nitrogen fertilizers using low-cost nitrification inhibitors and coating materials need to be developed and produced. Consumption of nutrients is considered to be the main cause for decline in crop yield and crop response ratio. (Prasad, 2012).

Recycling of phosphorus is important because phosphate rock reserves are a limited resource (Stewart et al., 2005) and the demand for phosphorous fertilizers is increasing, especially in third world countries.

One of the most attractive organic fertilizers is digestate that is applied as biofertilizer into the soil. It gives the opportunity to recover the nutrients, primarily nitrogen and phosphorus. As well the loss of organic matter is attenuating the loss of organic matter suffered by soils under agricultural exploitation.

Investigation of properties and impact of digestate in combination with other ecological fertilizers on common agricultural crops is a focus of the present Thesis.

2 Literature review

2.1 Main observed nutrients of specified organic fertilizers

2.1.1 Nitrogen (N)

Organic nitrogen in the soil is made up of plant and animal residues, by biomass of microorganisms (bacteria, algae, fungi, actinomycetes, protozoa), by products with biological and chemical transformations of organic nitrogen and especially the hummus (Vanek et al., 2012).

The content of total nitrogen in the soils ranges from 0.1 - 0.2 % (Richter and Hlusek, 2003), but may vary in a significant range (0.03 - 0.5) (Ivanic et al., 1984). The largest proportion of nitrogen is bound in organic compounds. It is 90 - 99 % of the total amount of nitrogen as reported by Richter and Hlusek (2003). Only a small fraction represents the inorganic nitrogen. Both of these forms are subjected to constant changes in the soil.

The nitrogen content in plants is a significant indicator in the assessment of the crop´s quality. It is not only affected by the fertilization (Balik et al., 2012).

The claims of most plants to nitrogen are high, particularly for those species that produce large amount of biomass, which may also be a specific indicator (Vanek et al., 2012).

The amount of nitrogen in dry matter of plants is in average in the range of 1 - 3 %. Nitrogen is a most limiting nutrient for plant´s production, and therefore it is widely applied into the land in large quantity (Mikanova and Simon, 2013).

2.1.2 Phosphorus (P)

The total quantity of phosphorus in soil ranges from 0.01 - 0.15 %. The soil with higher content of biomass usually exhibit higher amount of P, while the light soil with low content of organic matter has low content of P. The bulk of the total P in soil is unacceptable for plants (Vanek et al., 2012).

Phosphorus is received relatively evenly by plants during whole vegetation. However, for a good yield and quality of production is crucial its content in young plants (mostly 0.4 % in dry matter). Later, the content is in the range of 0.3 - 0.4 % (Chen et al., 2000).

The claim on phosphorus of each plant´s species is not differ significantly. The phosphorus consumption by crops depending on the yield is usually in the range of 15 - 40 kg P/ha. Most of the P is concentrated in the seeds, thus its largest export is by harvest of crops for grain and other seed crops (Delgado and Scalenghe, 2008).

2.1.3 Potassium (K)

More than 98 % of potassium is bound in the soil minerals from total amount of K in the soil. Only less than 2 % is contained in soil organic matter. Organic fraction of K is included in non-composted plant tissues and 25 – 50 % kg K/ha is included in microbial biomass. The humus substances do not almost contain potassium (Madaras et al., 2012).

Potassium is accepted as a cation by plants actively (it is prevalent at lower concentrations of K in soil solution) and passively at inverse concentration. An excessive intake of K can lead to unwanted effect like accumulation of K in plant´s tissues. That is called a “luxury” consumerism and leads to restriction of other cations (Na, Mg, Ca). Intake of potassium is in addition to its concentration in the soil solution significantly affected by moisture, temperature and the intensity of solar radiation (Ghosh et al., 2006).

Total potassium need is different for each plant species. The crops with vegetative character of products have high demands and consumption of potassium. The crops with long vegetative period have the same needs (e.g. vegetables, particularly cruciferous, celery root or brassicaceous plants) (Vanek et al., 2012).

Grains use around 100 kg K/ha. The plants with lower biomass production (pea, bean and others) evince small consumption of potassium. Claim on potassium nutrition grows with biomass production and culminate before flowering. Majority of K is received in a very short time period (Vanek et al, 2012).

2.2 Organic fertilizers

Using of organic fertilizers has an irreplaceable role in the supply of organic matter and nutrients into the soil, and thus also maintaining and enhancing soil fertility. Organic fertilizers include manure, green manure, liquid manure, slurry and straw, as well as other residues of plant origin and other side products originated from livestock farming, emerging particularly in the primary agricultural production, if they are not further adjusted and processed. It is a fertilizer which consists of the organic material of vegetable or animal origin as a main ingredient (carbohydrates, cellulose, hemicellulose, lignin, amino acids, proteins, auxins, etc.) that cannot be replaced in no way in the context of increasing soil fertility (Kasal et al., 2010).

The main sources of organic substances are post-harvest and root residues (make up to 60 % of the total requirement) and further organic fertilizers (Vanek et al., 2012).

Among the organic fertilizers are included also industrial-made composts, substrates, but also digestate from biogas plants, or sewage sludge from waste water treatment plants (Kasal et al., 2010).

Further the fertilizer consists of the main macronutrients (N, P, K, Ca, Mg), from which the humus and soil nutrient supply arise. Organic fertilizers improve the quality of the soil, provide aeration to the soil and support its absorption capacity and plant growth. Organic substances are able to retain water and nutrients in the sandy soils and heavy soil are more loosen and aerated (Martiskova, 2009).

2.2.1 Digestate

Gomez et al. (2005) defined digestate as one of the most attractive options in terms of reducing environmental issues to apply digestate as biofertiliser to the soil, because it allows recovering the nutrients - primarily, nitrogen and phosphorus. As well, it allows limiting the loss of organic matter suffered by soils under agricultural exploitation. Moreover, digestate can be produced anywhere via anaerobic digestion and is cheap compared to other fertilizers (Owamah et al., 2014).

Digestate is a side fermented product of the biogas plants that come from organic waste. Digestate may be a solid or liquid material, the fugate. It depends on the biogas technology (Vana, 2009; Makadi et al., 2012). Separated fermented residue has around 30 % of dry matter (solid fraction) (Babicka, 2012). Digestate contains a high proportion of mineral nitrogen (N), and in particular in the form of ammonium, which is available for plants. In addition, it includes other macroelements and microelements needed for plants´ growth. Therefore, digestate can be a useful source of plant nutrients and seems to be an effective fertilizer for agricultural crops (Makadi et al., 2012).

Organic matter from digestate shall satisfy the essential requirements to be marked as organic fertilizer. The matter must be readily microbial-biodegradable to release the necessary energy for soil microorganisms (Kolar et al., 2009).

Further it is characterized such a solid residue of the anaerobic fermentation process containing biodegradable substances (Dostal and Richter, 2008).

2.2.1.1 Source of digestate fertilizer

Matter composition of the digestate is closely related not only to the parameters influencing the progress of anaerobic digestion inside the reactor, but also with the quality, composition and origin of raw materials entering into a process of anaerobic digestion (Poffet, 2008). These are substrates of primary agricultural production (e.g. livestock manure, aimed grown energy crops), biological waste from the agri-food processing industry and biodegradable municipal waste. The most commonly used types of manure usable in BGP[1] given to the representation of individual livestock farming technology in the Czech Republic are swine slurry, cattle slurry, cattle manure, swine manure and poultry manure (Kratochvilova et al., 2009).

A stabilized digestate can be only used to fertilizing and which is produced by the right technological procedure and an appropriate organic load of the fermenters. The main source of problems are substrates with high contents of organically bound nitrogen, and therefore it is necessary to pay great attention to the composition of input raw materials and mainly to the total ratio of C:N. Inputted material into fermenters should have the ratio of C:N minimally 10:1, better around 20:1 in the interest of quality digestate. It is possible to reach by co-fermentation of slurry with plant materials. The choice of digestate amount has to be based from nitrogen content. Very important is to keep in mind the percent of nitrogen in absolute dry matter (and thus hundred percent) or in fresh matter of digestate (Marada et al., 2008).

2.2.1.2 Digestate properties

Digestate contains relatively high amount of total nitrogen (0.2 % but even 1 % in the mass), the higher pH value (7-8), the lower carbon content and dry weight ranges from 2 % to 13 % in comparison to traditional livestock manure (Marada et al., 2008).

[1] Biogas plant

Usage and amount of digestate as a fertilizer is similar to slurry properties and its usage. Of course it is necessary to take into consideration the current nutrient content, mainly the nitrogen and needs of the plants (Marada et al., 2008). When the average nitrogen content is 0.5 %, then one ton of digestate is applied into soil in amount of 5 kg N/ha. More detailed properties of digestate are described in Table 1.

The digestate composition presents a risk of nitrogen losses in gaseous form. Digestate is a fertilizer that contains valuable organic matter and mineral nutrients. As well it manifests small features of odour, resp. it does not have no odour. This is achieved owing to the appropriate mix of inputted material, their pre-treatment and in particular sufficient time keeping of input raw material in a digester at mesophilic (approx. 40°C) or thermophilic (approx. 55°C) temperature. The content of phosphorus, potassium and calcium remains preserved in its entirety (Marada et al., 2008; Hezky, 2012).

Table 1: The average content of dry matter (DM), organic matter (OM) and nutrients in digestate

Origin	**DM**	**OM**	**C**	**N_{tot}**	**$N\text{-}NH_4$**	**Ratio $N\text{-}NH_4$**	**C:N**	**P**	**K**
Cattle slurry	4.40	3.20	1.90	0.38	0.22	58 %	4.9:1	0.06	0.22
Maize digestate	5.90	4.20	2.50	0.50	0.31	62 %	4.9:1	0.06	0.37
Fugate	5.60	4.20	2.40	0.48	0.27	56 %	5:1	0.07	0.30
Separate	22.60	19.10	11.10	0.68	0.26	38 %	19.8:1	0.14	0.59

Source: Dostal, 2010

The main advantages of the digestate utilization in comparison with the other manures, the authors state (Dostal and Richter, 2008; Vana, 2009):

- a significant reduction of odour,
- a decrease of corrosive action,
- an ability to improve the fluidity or the possibility of the effective drainage,
- a decrease of the load on air-greenhouse gases,
- a low content of disease bacteria by up to 90 %,
- a minimization in weed infestation of crops and arable land.

2.2.1.3 Chemical properties of separate/dry digestate

Basic chemical properties of separate that affect plant growth include the value of pH, the value of electrical conductivity (EC) and the value of water extract, which shows the content of soluble salts and acceptable nutrients content. Primarily nitrogen in the form of ammonium and nitrate, phosphorus, potassium, magnesium and calcium, but also trace elements such as iron, manganese, zinc, copper, boron, molybdenum (Tlustos et al., 2013).

Furthermore, studies have claimed that digestate can still contain an organic fraction not yet completely biodegraded. As well as complex organic elements, salts or pathogenic bacteria that can affect the soil biota (Teglia et al., 2010).

It is worth mentioning that digestate from only animal manure or from waste has a different composition (Fuchs et al., 2008; Alburquerque et al., 2012; Tambone et al., 2015).

Pivato et al. (2016) set the physical and chemical properties of the solid sample and different methodologies were used for the relevant parameters: humidity, pH, conductivity, organic carbon, ammonia nitrogen, organic nitrogen. As well quantification of P, K, Zn, Cd, Cr, Cu, Pb, Hg, Ni and hexavalent chromium was made.

2.2.1.4 Acceptable content of nutrients in separate

Richter et al., (2010) state that the pH value ranged from 8.0 up to 9.1 and the value of electrical conductivity reached from 0.79 to 1.03 mS/cm. Furthermore, the values of macronutrient and micronutrient have been set.

Tlustos et al. (2013) defined the content of nutrients in separate. Klir and Kozlova (2015) and Eagri (2016) established content of nutrients in other additives.

Determination the appropriate amount of digestate according to Marada et al. (2008) is based on:

- the necessary nutrients of plants for the estimated crop yield and quality of production,
- the amount of accessible nutrients in the soil and site conditions (in particular the influence of climate, soil type and soil category,
- the soil reaction (pH), the ratio of major cations (calcium, magnesium, and potassium), and the amount of soil organic matter (humus),

- the growing conditions affecting the accessibility of nutrients (preceding crop, soil treatment).

Liquid digestate with a share of 6 - 10 % of dry matter is necessary to detach the screen or drum separators. The separate occur after the separation of solid parts and which contains approximately 29.3 % dry matter and the fugate (Pawlica, 2010).

An application of digestate into agricultural land as a fertilizer in liquid or separated form is the basic use of digestate from agricultural BGP (Babicka, 2012).

2.2.1.5 Directive

Digestate from the BGS is matter where raw materials are livestock manure or vegetable materials. Into these BGS are entering solely plant material such as straw, grass silage, corn silage corn and livestock manure as well. The digestate has to be registered according to § 4 of law No 156/1998 Coll., on fertilisers when is placed into circulation (Vecerova, 2008).

Digestate can be applied only in the case when it is registered in accordance with the act on fertilizers.

But the digestate is not subject to this requirement, if is made exclusively from livestock manure or bulky feeds and on the premises of the producer. Digestate is based on government regulation No. 103/2003 Coll. (the nitrates directive) the fertilizer with quickly released nitrogen (C: N ratio of less than 10). This regulation restricts or even prohibits its use in the vulnerable areas in a certain time period (PV-AGRI, 2012).

According to government regulation No. 262/2012 Coll., on the determination of vulnerable areas and the programme of action, it follows restrictions for digistate fertilizing. For example, for silage maize fertilization is allowed maximally an amount of nitrogen 230 kg/ha in digestate. To this restriction is related a law No. 254/2001 Coll., on waters where digestate is included among exceptionable substance and these substances cannot leak into underground water or threaten the environment (Marecek, 2010).

2.2.2 Other ecological fertilizers

Organic fertilizer has an irreplaceable role in plant production system. It cannot be successfully managed without intake of organic matter to the soil in terms of maintaining or improving soil fertility in the long term range (see Table 2).

According to Regulation 474/2000 Coll. we divide limits of risk in substrates, organic and state fertilizers in following table:

Table 2: Acceptable limits of selected elements in ecological fertilizers [mg/kg in DM]

Fertilizer	Cd	Pb	Hg	As	Cr	Cu	Mo	Ni	Zn	PAH
Organic and state fertilizers DM > 13 %	2	100	1	20	100	150	20	50	600	-
Organic and state fertilizers DM < 13 %	2	100	1	20	100	250	20	50	1200	-
Ash from biomass	5	50	0.5	20	50	-	-	-	-	20

Notice: PAH – polycyclic aromatic hydrocarbons, DM – dry matter

Source: Klir and Kozlova, 2015

2.2.2.1 Manure

Manure contains the macrobiogenic and microbiogenic elements. An organic fraction of manure is from 85 – 90 % in the form of half-decomposed organic matter. The rest consists of humus substances. The ratio of carbon to nitrogen is between 20-30:1, while high-quality manure has a narrower ratio below 17:1 and manure of lower quality ratio wider than 24:1. Plant nutrients in manure are contained in mineral and organic form. Nitrogen in ammonia form is 29 % and 70 % in organic form. Nitrate content shall not exceed 1%. Phosphorus and potassium is contained in variable organic forms (Richter and Kubat, 2003).

Sulphur is bound from 40 % in organic carbon, 20 % is in the form of sulphides and the remaining 40 % in the form of organic and inorganic sulphates (Pedersen et al., 1998).

Manure also contains a significant amount of microorganisms (1 – 2 % of dry matter), then some of the biologically active substances such as auxins, enzymes, etc. (Richter and Kubat, 2003). More nutrients in different manures see in Table 3.

Table 3: The average content of dry matter (DM), organic matter (OM) and nutrients in manure

Manure	**Fresh content (%)**						
	DM	**OM**	**N**	**P**	**K**	**Ca**	**Mg**
Cattle	24.00	17.00	0.48	0.11	0.52	0.37	0.08
Horse	25.00	20.00	0.65	0.13	0.52	0.21	0.11
Poultry	-	-	2.94	0.40	17.28	-	-

Source: Vanek et al., 2012

2.2.2.2 Slurry

The composition of the slurry is very different. Percentage of slurry is limiting for the nutrient content and which is mainly influenced by the proportion of technological water. Nitrogen and carbon ratio decides about the high value of the manure which is in the range of 4 – 8:1. The C: N ratio affects the speed of transformation of organic substances in the soil. Although, the release of nitrogen came about from organic bond, but also into organic compounds (immobilization) with the wider C: N ratio. During this process emerge more stable organic substances (Richter and Rimovsky, 1996).

The most nutrient contains slurry from poultry (see Table 4). The manure of cattle and pigs reported a significantly lower nutrient content. Higher content of N and P is in pigs´ slurry, bovine manure has higher contents of K (Vanek et al., 2012).

Dry matter content from 7.5 – 15 % is desirable for cattle and pig slurry and 15 – 20 % for poultry manure. Organic materials make up about 70 % to 80 % of the dry matter.

Nutrients contained in slurry are easily accessible for the plants. Swine manure contains essential microelements for plant nutrition, such as B, Cu, Mn, Co, Zn and Mo (Hlusek, 2004).

Table 4: The average content of dry matter (DM), organic matter (OM) and nutrients in slurry

Slurry	Fresh content (%)						
	DM	**OM**	**N**	**P**	**K**	**Ca**	**Mg**
Cattle	7.80	6.00	0.32	0.07	0.40	0.14	0.04
Swine	6.80	5.30	0.50	0.13	0.19	0.24	0.04
Poultry	11.80	8.10	0.96	0.28	0.32	0.94	0.06

Source: Vanek et al., 2012

2.2.2.3 Liquid manure

This organic fertilizer is considered as a very effective nitrogen-potassium fertilizer (see Table 5) according to the chemical composition of liquid manure (Richter and Rimovsky, 1996). The content of organic matter and phosphorus there is negligible. The dose of liquid manure is therefore followed by the demands of the crops on nitrogen or potassium (Vanek et al., 2007). A dose 10 t/ha of liquid manure in medium quality is equal to 23 kg of nitrogen and 33 kg of potassium in industrial fertilizers. Up to 85 % of the nitrogen is in the form of free ammonia, which easily escapes. Only 10 % of the nitrogen is bound to organic matter. Nutrients are contained in the liquid manure in an acceptable state for plants and fully usable immediately after fertilization (Richter et Rimovsky, 1996).

Table 5: The average content of dry matter (DM), organic matter (OM) and nutrients in liquid manure (LM)

LM	Fresh content (%)					
	DM	**OM**	**N**	**N-NH$_4$**	**P**	**K**
Cattle	12.0-30.0	7.0-15.0	1.5-2.5	1.0-2.0	0.1	2.8-6.0
Swine	11.0-20.0	5.0-10.0	2.5-3.2	1.8-2.6	0.1-0.2	2.0-4.0

Source: Beer et al., 1990

2.2.2.4 Straw

The chemical composition of the straw is different and depends on the type of crops (see Table 6), fertilisation level and content accessible nutrients in the soil. Straw contains on average 80 % of organic substances which are subject to degradation (mineralization), but they are also a valuable raw material for the creation of permanent humus. Soils fertile by straw are more aerial (looser), dry up more quickly and it is easier to cultivate them (Richter et Rimanovsky, 1996). It is desirable to apply in about 4 - 6 kg N per tonne of straw (Vanek et al., 2012).

Table 6: The average content of dry matter (DM), organic matter (OM) and nutrients in straw

Straw	Fresh content (%)							
	DM	**OM**	**N**	**P**	**K**	**Ca**	**Mg**	**C:N**
Cereals	86.0	82.0	0.45	0.09	0.79	0.24	0.06	80-100:1
Maize	85.0	80.0	0.48	0.16	1.26	0.32	0.14	60-80:1
Legumes	86.0	80.0	1.33	0.16	1.07	0.91	0.02	20-25:1

Source: Richter and Rimovsky, 1996

2.2.2.5 Green manure

Intercrops grown for green manure have a fertilize effect. They are an important contribution to the enrichment of the soil of organic matter and retention of mobile nutrients, in particular N and Ca in the organic matter (see Table 7). By ploughing of root and stubble residues occurs to a specific limitation of losses of nutrients by the washing out, further mobilization of phosphorus and other elements from the soil supplies from the difficult to reach forms (Vach and Javurek, 2007).

Intercrops have also significant effect in restoration of the microbial life of the soil and have a beneficial effect for increasing or at least maintaining the content of humus in the soil (Vach and Javurek 2007).

Among the most commonly cultivated intercrops include mustard, rape, radish or white clover (Vanek et al., 2012).

The amount of nutrients contained in above-ground mass (especially legumes) is accessible to majority of plants after degradation in the soil (mineralization) and roughly equal to the nutrients´ content in the same amount of manure (Vanek et al., 2012).

Table 7: Returning of nutrients into soil by green fertilizing

Crop	Nutrients (kg/ha)				
	N	**P**	**K**	**Ca**	**Mg**
Red clover	109.1	10.2	100.0	87.9	16.6
Pea, vetch	104.9	9.6	64.9	55.2	13.8

Resource: Vanek et al., 2016

2.2.3 Ash

Ash is an inorganic portion of the fuel that remains in the boiler after the burning of organic matter contained in the biomass and that contains most of the minerals of the original biomass (Khan et al., 2009). The amount of ash in the products from biomass is on average in the range of 1 - 6 %.

Table 8: Ash content in biomass

Wood	Bark	Straw	Grasses	Rice peels
0.3 - 1.0	3.0 – 4.0	5.0	7.0	40.0

Source: Biedermann and Obernberger, 2005; James et al., 2012

Wood usually contains a relatively lower amount of ash, while the significantly higher values can be found in the bark, straw or grasses (Biedermann and Obernberger, 2005). In the rice peels even up to several time higher value as shows Table 8 (James et.al., 2012).

Ash from straw, in comparison with wood, contains more K_2O and has a higher pH value (Obernberger and Supancic, 2009; Hinojosa et al., 2014). Higher contain of chlorine is typical especially for ash from straw, when during the combustion of biomass to gas release significant levels of chlorine and alkali metals, e.g. in the form of HCl (g), KCl (g), KOH (g) or NaCl (g). Obernberger et al. (1997) report that the flying ash from the incineration of cereals may contain up to 90 weight percent of KCl from share of inorganic matter (Hansen et al., 2001, Wei et al., 2005). Wooden biomass contains more calcium in comparison with the ashes from the burning of straw (Du et al., 2014).

2.2.3.1 *Chemical composition and nutrients in ash*

The ash contains many macro-elements and micro-elements that are necessary for plants´ growth. Most of these nutrients are originally derived from the soil and the atmosphere in the course of plant growth. The ash is generally alkaline with high levels of calcium, magnesium, phosphorus, potassium and other elements. Carbon content varies considerably depending on the use of combustion technology; effective combustion produces a light brown ash with minimum carbon content. The nitrogen content is low in ash. An input of nitrogen to the environment does not increase by the ash application. The average pattern of the wood ash in the concept of commercial fertilizers can has essential nutrients (N-P-K) in a proportion of 0-1-3. Ash is a source of many nutrients necessary for plant growth only in trace amount except to the macro-elements (Soucek and Spulak, 2006) that are stated in Table 9

Table 9: Main nutrients found in ashes

Nutrients in ash (%)			
Ca	**K**	**Mg**	**P**
7.0 – 45.0	3.0 – 14.5	1.0 – 6.5	0.3 – 1.4

Sources: Hansen et al., 2001; Siddique, 2012

Ashes from straw and cereals contain a high amount of potassium (Obernberger et al., 1997; Biedermann and the Obernberger, 2005).

The dissolution of the ash in the soil and the rate of release of available nutrients to plants are more complicated than, for example, in the case of limestone. The ash from biomass contains cations in the form of oxides, hydroxides, carbonates and bicarbonates, which are dissolved under different conditions. Accessibility of nutrients is also influenced by the adsorption capacity of the soil. A certain proportion of elements found in soil solution after enrichment by ash may come from interaction of soil with Ca, Mg, or interaction with the soil exchange complex (Demeyer et al., 2001).

But changes in the availability of nutrients from the soil after application of ash from biomass are a combination of at least three factors: the quantity of nutrients supplied in the ash, changes in pH depending on the chemical balance of the soil, and microbial activity changes (Demeyer et al., 2001).

2.2.3.2 Combustion of ash

Chemical composition (nutrient content) and quantity of ash from biomass depends on the type of burned biomass, species of plant or its part, growth conditions, the age of the plants, but also from the applied fertilizers and pesticides, harvest techniques and season, pollution. However climate conditions of combustion, collection and storage are significant factors influencing chemical composition of ash (Vassilev et al., 2010; Pasquini and Alexander, 2004), the combustion conditions, combustion technologies, storage, etc. (Biedermann and Obernberger, 2005; James et al., 2012; Lovgren, 2012).

Similarly, as the proportion of ash, the levels of the various elements are significantly influenced by the biomass origin, plant species, or parts of it. Wood and bark are rich in calcium, while ash from straw and cereals contain a high amount of potassium (Biedermann and Obernberger, 2005).

Combustion conditions have a major effect on the composition of the ash. For instance, Etiegni and Campbell (1991) found out that ash production was reduce about 45 % after increasing the combustion temperature from 550°C to 1100 ° C, because the fuel and biomass was burnt better (Khan et al., 2009).

Microelements are present in smaller and variable quantities. The nitrogen content is negligible, as it escapes in the form of oxides to the atmosphere during the burning process (Lichtfouse et al., 2013). High pH value is the consequence of high content of calcium oxides, especially in grated ash after burning wood and bark, which leads to a reflection on the use of this material to modify soil reaction.

2.2.3.3 Cycle of nutrients in ash

The use of biomass as an energy source will be also kept in the future. This means that by the biomass utilization will increase the amount of residues (ashes) from bio-energetic processes. Reuse of ash in crop production is important for nutrient cycle in agriculture (Katai, 2006).

Application of ash for fertilizing that arose as a waste product when the burning of biomass, is less expensive than conventional mineral fertilization and liming (Ferreiro et al., 2011).

2.2.3.4 Phosphorus in ash

The presence of phosphorus in the ash is interesting from the point of view of limited world's resources of phosphorus and incessantly increasing prices in phosphate fertilisers. Alternative sources of fertilization, such as ash from biomass, could help to improve plant nutrition by phosphorus (Bhattacharya and Chattopadhyay, 2002).

The phosphorus content is higher in the ash based on cereals and oil seeds. The effect of phosphorus in ash depends on the crops as well (Krejsl and Scanlon, 1996). In the ash with relatively high phosphorus content (13 %) was found that the phosphorus is soluble from 80 % in citric acid, and is relatively available for plants. Ash had comparable or even better effect on the yield and quality of phosphorus in plants against highly soluble compounds of phosphorus that was verified by container experiments with different crops (Eichler-Lobermann et al., 2008). An increase of manganese content in plants was recorded and lesser extent of zinc and boron in the soil (Kabata-Pendias and Pendias, 2001).

2.2.3.5 Heavy metals and risk elements in ash

Elements representing the environmental problems are very sparsely represented in the ash. Concentration of heavy metals in the ash is generally low and similar to soils where the burning biomass grown up. Heavy metals can be separated from the ash during the processing and separation. However, it significantly increases the cost of production. How was mentioned above, ash can be considered as a suitable natural fertilizer supplying scarce elements. Its composition is similar to limestone utilization. But the content of substances in the ash is very variable, and therefore ash analysis has to prevent its use (Khan et al., 2009; Lichtfouse et al., 2013).

The potential use of ashes is influenced by the content of contaminants, including risk elements (Khan et al., 2009). Ash could be a source of contamination of soil by these elements along with the nutrients necessary for plant growth (Park et al., 2004; Patterson et al., 2004; Eichler-Loebermann et al., 2008). But the content of the risk elements in comparison with other substances used as fertilizer (e.g. sewage sludge) is relatively low (Omil et al., 2007). However, it depends on the used biomass (Park et al., 2004).

In general, the ash from the combustion of wood contains higher concentrations of As, Cd, Pb and Hg than ash from the incineration of straw, residues from the grain and fruit processing, etc. (James et al., 2012). Ashes from biomass contain less of heavy metals in comparison with coal (Campbell, 1990).

Some risk elements in the soil can be immobilized by an ash application by increasing the value of pH (Demeyer et al., 2001; Omil et al., 2007). But the risk of heavy metals release should not be underestimated. The solubility of heavy metals varies considerably depending on the chemical composition of ash and is affected by specific soil conditions (Ferreiro et al., 2011).

Cadmium is considered to be the most problematic element in ashes from biomass because of significant toxicity to biota and high mobility (Tulonen et al., 2012).

The ash from the combustion of biomass, regardless of origin, is typically alkaline (Campbell, 1990) and has a relatively similar effects as limestone (Pan and Eberhardt, 2011; Arshad et al., 2012), which is due to the high content and form of alkali metals in the ashes (Kuba et al., 2008). The pH values of ashes from wood biomass are generally higher than for ashes from straw and grain because of the higher content of calcium and a lower content of sulphur and chlorine (Van Loo and Koppejan, 2008).

2.2.3.6 Ash utilization in agriculture

One of the main use of the ash appears to be its application as fertilizer because of the high bazical cations content and nutrients (Ludwig et al., 2002; Arshad et al., 2012; Gomez-Rey et al., 2012; James et al., 2012; Moilanen et al., 2012; Norstrom et al., 2012), which remain in the ash after incineration of biomass (Tulonen et al., 2012).

The use of ash as fertilizers is provided, inter alia, for the following reasons:

- to promote the growth of plants, since the increased nutrient content (Ca, K, S, Mg, P, Na) and micronutrients (Mn, Zn, Fe, B, Cu, Mo) improves soil fertility,
- an alkaline nature of the ash provides an effect similar to liming,
- to decrease mobility and bioavailability of risk elements,
- to reduce the toxicity of aluminum, manganese and iron for plants by reducing the exchange contents of their ions in acidic soils,
- to support biological activity and conditions for certain micro-organisms,

- to improve the texture, aeration and water capacity of soils (Vassilev et al., 2013).

2.2.4 Poultry manure

Organic manures and crop residues can play a major role in recycling K. Concomitant use of organic manure and N fertilizer can reduce leaching losses of N (Prasad, 2012). As poultry waste contains a high concentration of nutrients and addition of small quantity of it in an integrated nutrient management (INM) system could meet the shortage of other fertilizers to some extent. Thus INM is essential for increasing food production (Ghosh et al., 2004; Prasad, 2012).

Poultry excrements are still considered as one of the most effective fertilizers of natural origin. Contain a significant amount of nitrogenous compounds and it is suitable for soil fertilizing. The average value of the nutrients in dried poultry droppings is following (Table 10):

Table 10: Average value of the nutrients in poultry manure

Nutrients in poultry manure (%)		
N	**P_2O_5**	**K_2O**
40	35	27

Source: Koubova, 2005

The poultry manure is a relatively cheap source of both macronutrients (N, P, K, Ca, Mg, S) and micronutrients (Cu, Fe, Mn, B). As well can increase soil carbon and N content, soil porosity and enhance soil microbial activity (Ghosh et al., 2004).

Eghball (2000) tested poultry manure as fertilizer to prevent diseases at cereals. The samples were collected and left for 6 weeks when they underwent the aerobic decomposition. The carbon:nitrogen (C:N) ratio of the compost was achieved to test the maturity of the compost and value of ≤25 % was taken as well matured compost ready to use. The mineralization rate of compost was not determined but was expected to be in the range of 13 – 18 %.

According to a study, Yoger et al. (2006) tested a fertilizer where dried samples of composted poultry and cattle manures were collected randomly from several piles, packed in polythene bags and transported to the laboratory for nutrient analysis. Oven dried samples (70°C) were milled to pass through 2 mm sieve. Samples were digested following the method described by the authors and total nitrogen (N) and potassium (K) in the digest were determined by Spectrophotometer (Pye-Unicam Model SP191). Phosphorus (P) was determined by Olsen method (Olsen et al., 1954; Lyimo et al., 2012).

2.2.4.1 Heavy metals in manure

One of the main limiting factors in the use of manure is the concentrations of heavy metals (Alvarenga et al., 2015). It has been reported that the concentrations of some heavy metals or metalloids in manure have increased during the last few decades due to the additions of metals to animal feed in intensive animal production systems (Paradelo et al., 2011; Wang et al., 2013). Heavy metals are non-degradable during the composting process and persist in the final application (Lopes et al., 2011; Moller and Schultheiss, 2015). Some studies have shown that applications of animal manures or manure composts containing high concentrations of heavy metals can result in excessive accumulation in soil, leading to adverse effects on soil quality (Hang et al., 2009; Zhejazkova and Warman, 2003).

Moreover, the concentrations of heavy metals such as Cd in soil have increased considerably over the last 3 decades (Zhao et al., 2015). Therefore, there is an urgent need to monitor and minimize the inputs of heavy metals to agricultural soils.

2.2.4.2 Potential of poultry manure and conducted experiments

Poultry manure is an excellent substrate for the growth of microorganisms, especially bacteria linked to the organic matter, which allow the development of a dense population of protozoa (Schroeder, 1980). Furthermore, it has a high content of nitrogen and phosphorus (Arredondo, 1993). Dissolving the poultry manure (previously dried and sieved) in water and maintaining it with constant aeration during 24 h may have allowed the establishment of certain microorganisms and, therefore, may accelerate the decomposition processes of the fertilizer. This is in addition to diminishing the biochemical oxygen demand (BOD) in the experimental systems, since according to the poultry manure has a high BOD.

Teicher-Coddington et al. (1990) state that organic fertilization in ponds can fulfil all conventional dietary requirements with no need for additional supplements. Arredondo (1993) points out that when fertilizers are applied to a pond, the organisms in culture can consume it directly or they can grow on the natural feed that is produced as a result of fertilization.

According to Edward and Daniel (1992) when poultry manure was applied in combination with chemical fertilizer, it supplemented all nutrients to crop, and increased the productivity of crop.

2.3 Garden pea (*Pisum sativum L.*)

Legumes do not have big demands on before-crops. However, it is inappropriate to grow them in a row or after clovers. They are suitable to plant before cereals. Manured root crops are proper preceding crops for legumes (Urban and Vasak, 2016).

The plant does not require excessive fertilization by nitrogen. Selgen (2016a) has investigated the demand of fertilization for garden pea.

Fertilization of legumes has specific peculiarities arising from biological fixation of N nodule bacteria and from soil pH requirements. To N fertilisation is usually dependent small amount of legumes. The total consumption of N by plants varies between 100 – 150 $kg.ha^{-1}$. Nitrogen is required in the first phase of growth to make the rhisobia. Phosphorus and potassium is used effectively from the soil than from direct fertilization (Urban and Vasak, 2016).

One of the main assumptions of the establishment of productive growth is high quality of seeds. Germination is between 75 – 80 %. It is necessary to treat the seeds as prevent from the occurrence of harmful agents and to increase performance (Saucke and Ackermann, 2006).

Pea is a traditional and most planted legume in the Czech Republic. Worldwide, it is an important legume, and one of the oldest crops (Eagri, 2016).

It is recommended to grow pea in a well aerated soils, biologically active, supplied enough by Ca and P, and with a neutral to slightly acidic pH (Saucke and Ackermann, 2006).

The pea belongs among the crops with less yield stability associated with considerable revenue dependency on weather. It is most often achieved the yield 2.0 - 3.0 $t.ha^{-1}$ (Urban and Vasak, 2016).

2.3.1 Potential of pea and conducted experiments

Crop yield and nutrient concentrations of pea grain yields in the control treatment of the pea plots were similar to those obtained under organic cultivation (Saucke and Ackermann, 2006), but lower than those produced under conventionally grown peas

(Hauggaard-Nielsen et al., 2001; Ghaley et al., 2005; Neumann et al., 2007; Neumann et al., 2009).

Kaye and Hart (1997) and Manson et al. (2009) found that organic fertilizer addition might not have to enhance the yields of peas. It led to a yield reduction in the pea plots. The added organic fertilizers were rich in easily decomposable components that may result in competition between soil microorganisms and plants for easily available nutrients, especially N.

As the organic fertilizers, especially horse manure, led to a significant decrease in S concentrations in pea biomass. S deficiency might be one reason for the absence of positive organic fertilizer effects, because legumes have a high S demand (Scherer et al., 2008; Varin et al., 2010).

2.3.2 Fertilizer effects on crop yields of pea

Prochazkova et al. (2002), Levy and Taylor (2003) and Roy et al.(2010) established several main factors that may influence positive organic fertilizer effect on crop growths and yields. E.g. poor germination, emergence, and subsequent early plant growth can influence further development of the plants.

These problems might be intensified by the generally low plant density that was below the recommended seed rate for field peas of 80 seeds per m^{-2} (Neumann et al., 2007).

Jannoura et al. (2014) researched organic fertilizer effects on grown peas and oats manured by horse manure and yard-waste compost at 10 t/ha each. By comparison the effects of these organic fertilizer and cropping system on pea yield, grown as the sole crop or intercropped, as well as N_2 fixation and photosynthetic rate. In general, organic fertilizer application improved nodule dry weight, N_2 fixation and photosynthetic rate of peas by an average of 15 %. As well soil microbial biomass was positively related with pea dry matter yields. Organic fertilizers increased the contents of microbial biomass C, N, P.

2.4 Spring Barley (*Hordeum vulgare L.)*

2.4.1 Technology of growing

More suitable for spring barley planting are suitable black earths, degraded black earths or brown earths. Medium heavy soils, earthy - sand-earthy soils are suitable in terms of soil types. The best barley thrives on soils with a pH from 6.0 to 7.1 (Urban and Vasak, 2016).

The best front-crops are manured potatoes and sugar beet, after which it achieves stable revenue. There is a risk of higher N content in grains after cereals´ growing and after ploughing of above-ground parts of sugar beet. Barley is extremely sensitive to fluctuations in the weather, the soil reaction, to inappropriate soil, on soiled seed or on uneven spreading of fertilizing (Urban and Vasak, 2016).

2.4.2 Growing requirements

The term of sowing is crucial to reach a good yield. The most appropriate term is in the spring as soon as possible, preferably in March – according to the soil moisture. Sowing depth is 20 – 30 mm. It is necessary to sow into the matured land. Recommended sowing rate is 3.5 millions of germinable grains per ha for better conditions (Urban and Vasak, 2016).

Spring barley is one of the most sensitive cereals for fertilizing. Any unevenness in the soil or fertilizer is reflected in imbalances of plants, the yield of grain or its quality. Increase in the protein content of the grain is caused by higher dose of nitrogen, which is again not suitable for malting barley. Nitrogen fertilization is done usually before sowing. On the very fertile habitats is applied to about 30 kg $N.ha^{-1}$ (for yield of 1t/ha), in poorer conditions and after cereals approx. 80 kg $N.ha^{-1}$ (Urban and Vasak, 2016).

Richter and Ryant (2015) have published the demand of fetilization for spring barley. Seed grains are treated against diseases of barley. Leaf diseases occur such as powdery mildew, brown staining, rust barley and so on.

2.4.3 Potential of barley and conducted experiments

Oelofse et al. (2015) state that soil organic carbon (SOC) has no significant effect on potential winter wheat, whilst effect for spring barley was found. However, only for a sandy loam soil type was found a significantly positive effect of SOC on potential yields. A significant negative relationship was found between SOC and nitrogen use efficiency (NUE) for both winter wheat and spring barley.

Soil organic matter (SOM) influences soil biological, physical and chemical properties, therefore from an agronomic perspective. SOM is considered as an important contribution in a variety of ways to improve some of the factors influencing crop yield. SOM has been associated with: better plant nutrition, particularly as a potential source of nutrients, improved soil structure, improved water holding capacity and soil buffering capacity (Loveland and Webb, 2003; Johnston et al., 2009). SOM levels are thus closely linked to main soil parameters and to an economically and environmentally sustainable agriculture (Christensen and Johnston, 1997).

The organic manuring experiment in Woburn (Johnston et al., 2009) shows that yields for a rotation of potatoes, winter wheat, sugar beet and spring barley were always larger on soils holding more organic matter, despite equal levels of nitrogen application. Similarly, cultivation of spring barley (Hoosfield continuous barley experiment) resulted in higher yields on fields with higher levels of SOM for three of four cultivars reported (Christensen and Johnston, 1997; Johnston et al., 2009).

Alvarez et al. (2002) found that SOM content (averaging 45 $Mg.ha^{-1}$ for 0 - 20 cm in the experiment) was the most important explanatory variable of wheat yields (ranging from 1,000 to 5,000 $kg.ha^{-1}$). However, other variables which also correlated positively to yields, included rainfall and potential mineralization N. In these experiments, higher SOC content would also be associated with better nutritional status of the soil and therefore the improved yields may be an effect of crop nutrient supply rather than an effect of the SOM itself. Loveland and Webb (2003) conclude there is some evidence that SOC reduction leads to a reduction in yield potential, although these reductions are small (Korschens et al., 2013).

Ter Meulen (1924) determined the soil organic carbon according to the ter Meulen´s method based on loss on ignition at 550 °C in a LECO IR-12 furnace.

One of the hypotheses of study provided by Oelofse et al. (2015) was that an increased level of SOC leads to an increase in potential yields i.e., the yield that can be attained when sufficient levels nutrients are available to not limit biomass production.

Korschens et al. (2013) found that the SOM effects on yields of winter wheat were in the order of 3 %. This implies that 90 – 97 % of potential yields can be obtained by adding sufficient mineral fertiliser. The potential yield is the yield achieved at sufficient nitrogen application and was assumed other nutrients to be non-limiting.

Schjonning et al. (2009) stated that whilst a common border cannot be defined for SOC, low levels of SOC can have serious implications for aggregate stability. In the Hoosfield continuous barley experiment, the authors attribute the observed higher yields in fields with higher SOM levels primarily to improved soil structure from SOM, although they recognize the potential addition of N mineralized late in the season (Johnston et al., 2009).

2.5 Effect and importance of biomass in soil

Including of organic fertilizers into soil purpose a large and rapid increase in the soil microbial biomass (Ghoshal and Singh, 1995; Heinze et al., 2010) that form only a small fraction of soil organic matter. However the soil microbial biomass plays an important role in nutrient cycling and plant nutrition, due to its fast turnover (Jenkinson and Ladd, 1981). For this reason, some studies have found a close relationship between the soil microbial biomass and crop yields under greenhouse conditions (Chen et al., 2000) as well as under field conditions (Insam et al., 1991; Goyal et al., 1992; Khan and Joergensen, 2006; Mandal et al., 2007).

Reduced light may affect nodule biomass by restricting photosynthesis of peas and consequently the energy supply to the roots (Ghosh et al., 2006). Moreover, legumes are less competitive for available inorganic soil N than cereals (Jensen,1996).

3 Hypothesis and objectives of the Thesis

3.1 Hypothesis

The hypothesis for this Thesis is based on literature review knowledge and observations of previous studies about ecological or other fertilizers and theirs properties. Hypothesis is: Addition of ecological fertilizers (such as ash from grain straw, poultry excrements) may improve mixed fertilizer(s) properties based on solid digestate.

3.2 Objectives of Thesis

The main objective of the Thesis is determination of properties and optimal composition of mixed fertilizer(s). The secondary objective is evaluation of mixed fertilizers' impact on selected plants´ production.

4 Materials and methods

4.1 Methodology of literature review

For elaborating of research review, the theoretical part of the Thesis was used literature primarily from foreign authors, but as well from Czech authors. Most of the articles and other scientific thesis were used in electronic version from scientific databases. Draw on databases were mainly from: Scopus, Science Direct or Scholar Google. The articles were searched based on combination of keywords as: solid digestate, separate, ecological fertilizers, optimal composition of fertilizers, straw ash, poultry manure, spring barley, garden pea, etc.

Used scientific articles were mostly published in journals as: Agronomy Journal, Bioresource Technology, Plant and Soil, Soil Biology and Biochemistry, European Journal of Agronomy, Journal of Plant Nutrition and Soil Science or Field Crops Research.

4.2 Methodology of practical research

The idea of the thesis was to develop and analyse mixed fertilizer(s) based on solid digestate from biogas plant and ecological fertilizers (poultry excrements, ash from grain straw).

The main methodological steps of the research was:

1) to classify a tested fertilizer

2) to determine a chemical composition of fertilizers – C, H, N

3) to determine a microelement content in fertilizers

4) to determine an ash content in fertilizers

5) to classify a soil origin and composition

6) to prepare an ecological fertilizer samples - ratio/weight

7) to set up a field trial and test the fertilizer(s) on spring barley and garden pea

8) to measure a plant´s growths

9) to harvest the field trial and prepare the plants for drying

10 to determine a plant´s dry matter/yields

11) to determine a plant nutrients´ content - crushing of samples

Determination of all above mentioned fertilizers' properties was carried out according to the given standards.

Research was conducted in cooperation with pellet producing company and Research Institute of Agricultural Engineering in Prague (RIAE).

Subsequently, the tested samples were processed in laboratories and were applied quantitative statistical methods. Before the application of the intended statistical testing, the data were checked (based on the outputs of descriptive statistics) for the normality. As was seen from the results of normality tests within the p-values were lower than set significance level 0.05, thus was rejected the null hypothesis that the data have normal distribution and accepted alternative hypothesis.

One-way ANOVA was applied on collected data and further the Post hoc analysis (Tukey test) was performed that show, which specific groups differ.

4.3 Materials I.

Materials for setting up the field trial were originated from the Heesters s.r.o., the pellet producing company. All materials - digestate, ash from biomass (wheat straw), and fermented poultry excrements were the fundamental compounds of tested ecological fertilizer. All these samples contained certain amount of moisture content between 10 % - 15 % that was taken into consideration during sample processing. These source as the parts of fertilizer were used due to good potential as organic fertilizer. The compounds are waste/residues that are produced during other agriculture processing, e.g. crop harvesting, combustion of residues from harvesting or residues from livestock production.

Thus, input materials have been included into study after initial measuring of element´s content. The short description with compounds´ origin and properties is following:

4.3.1 Digestate

Digestate is considered as a basis of the fertilizer composition in this research. It regards a digestate, from biogas station that is produced by separation, i.e. mechanical draining. Such a made separate is dried. The used dry digestate/separate is a fermentation residue after an anaerobic digestion. Digestate used in this fertilizer is composed mainly from the feed materials and farm/state fertilizers. A ratio between both constituents is in a range 6:4 up to 8:2, respectively. Applied material into tested mixture of fertilizer was dry with value of moisture of 13.6 %.

4.3.2 Poultry manure

Fermented poultry faeces are an additive that is considered as main source of nitrogen. Due to the fermentation are preserved properties of faeces – the fertilizer is stable, homogenous with high fertile impact that increase use of nutrients and their release, also decrease a smell.

4.3.3 Ash from wheat straw

Next additive of tested organic fertilizer is ash. Grain straw was used for combustion due to adequate amount of potassium in its ash. Applied ash had moisture content 15 %.

4.4 Materials II.

In research were selected two model experimental plants and certain variety for research purposes that are described below: Garden pea (*Pisum sativum L.*), Spring barley (*Hordeum vulgare L.*). Garden pea was a first plant representative originated from leguminous plant/*Fabaceae* family involved into study Spring barley was selected as the second most spread cereal in Czech Republic and in European Union, and also because of fast growing properties (Chloupek et al., 2004; Eagri, 2016)

Concise description of model plants is following:

4.4.1 Garden pea (*Pisum sativum L.*)

The variety that was use is “Impuls”. The supplier of seed grain is Selgen, a.s. company as well. The variety is semi-late (semi-serotinous), green-seeded, type semi-leafless. The plant does not require excessive fertilization by nitrogen. The demand of fertilization is up to 20kg N/ha, 50-70 kg P_2O_5/ha, 80-120 kg K_2O/ha (Selgen, 2016a).

4.4.2 Spring barley (*Hordeum vulgare L.*)

Used variety is “Sebastian”. The supplier of seed grain is Selgen, a.s. company. The demand of fertilization for barley yield equal to 1 t of grain is 24 kg N, 5.2 kg P, 19.9 kg K (Richter and Ryant, 2016).

4.5 Methods

This practical review was carried out in RIAE and quantitative methods of research were applied. There were used the greenhouse, laboratories and appropriate tools. The compounds of tested ecological fertilizer were supplied into RIAE by the pellet producing company. Next part of research, specifically laboratory measurement of heavy metals, was provided by Laboratory of Environmental Chemistry in the Interfaculty Centre of Environmental Sciences, CULS[2].

Chemical properties of fertilizer´s constituents and soil applied in the trial were determined as the first. Then the field trial was set up – a mixture of soil and samples of fertilizers were prepared. Afterward the seeds of tested crops were sowed and the plants were measured in regular intervals for diverse time. Finally the trial was completed and the harvest was evaluated. For the data processing was used MS Excel and statistical software Statistica12.

4.5.1 Classification of tested fertilizer

The intention was to develop a fertilizer, whose predominant component is a separate from biogas plant (BGS). This material is a source of organic matter and

[2] Czech University of Life Sciences Prague

applicable as a fertilizer. To this base material were added additives to increase potassium, nitrogen and phosphorus content in form of ash from biomass and poultry excrements.

4.5.2 Determination of chemical composition of ecological fertilizers – CHN[3] macroelements

For all additives and separate were established physico-chemical compositions of base elements. Elementary analysis was performed in RIAE by automatic device LECO CHN628 Series Carbon/Hydrogen/Nitrogen Elemental Determinator from LECO Corporation. The determination of all basic elements is important to set right ration of fertilizer composition (see Figure 1).

The LECO device works with dry homogenized samples that are pack into tin foil (weight of foil is 0.1 g). The foil prevent from humidity absorption during hydrogen detection. The analytic sample is pressed into small globule when the air is pushed out. First sample contains a calibration powder (EDTA) to ensure correct work of analyser after last measurement. It is important to measure exactly given values. Then the analytic sample is put into autoloader and move into purge chamber to remove atmospheric gas. Next step is a dual-stage furnace system operated with temperature about 1,050°C. It is necessary a pure oxygen intake to ensure complete sample combustion without using any metal oxidizing reagents. The results of three repetitions were automatically calculated by computer software (Azom, 2016).

Figure 1: Leco analyser – CHN

Source: LECO, 2016

[3] Carbon, Hydrogen, Nitrogen

4.5.3 Determination of microelements´ content and other trace elements in fertilizers

The freeze-dried and homogenized samples were decomposed in a microwave-assisted wet digestion system with focused microwave heating. An aliquot (~0.3 g of dry matter,

in 3 replicates) of the sample was weighed in a quartz-glass digestion vessel (volume 35 ml) and 6.0 ml of concentrated nitric acid was added; the mixture was heated at maximum power 300 W, temperature 180 °C and maximum pressure 21 bar for 12 minutes. After cooling, the solution was quantitatively transferred to plastic polyethylene tubes, filled to 30 ml with MilliQ water and kept at laboratory temperature until measurement. Elements´ content (P, K, Al, V, Cr, Mn, Fe, Co, Ni, Cu, Zn, As, Se, Cd, Hg, Pb) in the digests was measured by inductively coupled plasma mass spectrometry using no gas mode or a collision cell mode to reduce potential interferences (Vlachosova, 2016).

4.5.4 Determination of ash content in ecological fertilizers

Samples of fertilizers were analysed in laboratory of RIAE. Ash content was established after combustion of sample by calculation of fertilizer weight. The samples of fertilizers were combusted in Muffle furnace LAC (see Figure 2). These furnaces are used for heat-treatment testing of various samples.

Figure 2: Muffle furnace LAC

Source: LAC, 2016

First the empty porcelain crucibles were heated up to 550°C for 60 minutes, after that were cooled and moved into desiccator. Further the combustion process was able to begin. One gram of certain analytical sample was weighted with cold crucibles. Then the crucibles were placed into cold furnace and heated up to 250°C for 30 minutes and maintained for 60 minutes to leave volatiles before sample ignition. In further 30 minutes the temperature was raised up to 550°C and maintained for 120 minutes to ensure complete combustion. Such a result of ash content was determined an average of nearest measured results (EN ISO 18122, 2016).

The temperature was regulated during whole combustion. Ash content was calculated according to formula:

$$A_d = \frac{(m_3 - m_1)}{(m_2 - m_1)} \cdot 100 \cdot \frac{100}{100 - M_{ad}} [\%] \qquad (1)$$

$\mathbf{m_1}$–weight of the empty porcelain crucible [g]
$\mathbf{m_2}$–weight of porcelain crucible with wet sample[g]
$\mathbf{m_3}$–weight of porcelain crucible with ash [g]
$\mathbf{M_{ad}}$–average moisture content of used sample [%]

4.5.5 Classification of soil origin and composition

The soil involved into field trial originated from RIAE sources. Primarily, the sample of soil was taken to measure soil chemical properties. The determination alike initial soil composition. It is necessary to know how much others nutrients is need to add.

To determine the content of nutrients in the soil was used XRF[4] analyser, which works on principle of rentgeno-fluorescence analysis (Figure 3) from Niton Corporation.

Lowest limits of detection, non-destructive analysis, low size and weight are the assets of this device (NitonUK, 2016).

[4] X-Ray Fluorescence

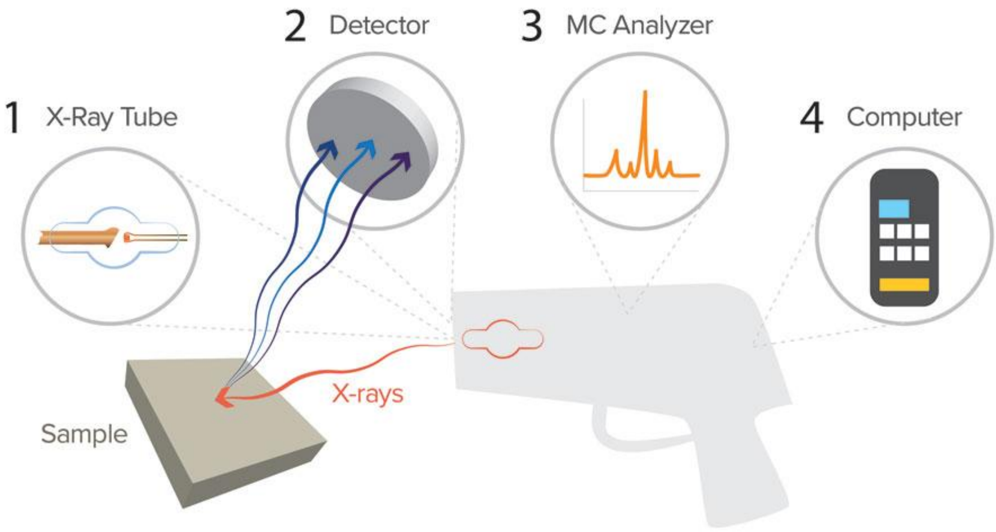

Figure 3: XRF process

Source: Niton, 2016

XRF refers to the emission of ‘secondary’ x-ray radiation from an element which has been bombarded with ‘primary’ x-ray radiation, i.e. radiation from an external source. The emission of this secondary radiation is known as ‘fluorescence’, and its measurement provides the mechanism for XRF spectrometry (NitonUK, 2016).

XRF spectrometry is widely used to determine the elemental composition of materials, particularly in fields such as metallurgy, geology, environmental analysis and forensics. This device allows obtain fast, accurate and non-destructive results in detection or analysis of elements. Each element can be identified by emissions and its intensity can be used to determine the amount of each element that is present in sample (NitonUK, 2016).

4.5.6 Preparation of ecological fertilizer samples – ratio/weight

After initial elementary determination it was counted the amount of each nutrient contained in fertilizers according to plant´s demand. The required amount of fertilizer was set after previous establishment of nutrient quantity. Also was set a ratio of each fertilizer according to requirement of trace elements/nutrients in soil. The ratio and weight was re-counted from ha to m^2 of the container.

A fertilizer ration was defined to ensure and to increase NPK intake into soil. Basic ratio of elements N:P:K for plants´ requirements is equal to ratio 5:4:1.

A mixture of fertilizers was proposed totally in 5 variations. First sample was used without any additional fertilizer. It is a test sample of soil. In second sample was used a separate. The other three samples were in different combination and weight ratio of separate, ash from grain straw and poultry manure. The weight ratio was determined on the basis of chemical analysis of the fertilizer and additives and plants´ requirement.

For pea sample was established two groups of tested samples as a repeating of the trial. Group A (Experiment 1) and group B (Experiment 2) that had the same composition of soil and fertilizers inside the containers. However, these containers were randomly placed on different sites inside the greenhouse. Specific marking of all container´s samples was 0A – 4A and 0B – 4B. The very soil was applied in container with zero number as test sample.

For barley sample was established one group of tested samples. Concentration of sowing was higher according to standards. It was applied 5 variations of fertilizer as well. The containers were marked as 0 – 4.

4.5.7 Setting up of field trial

The field trial with pea was set up on the 22nd of April 2016 and barley was sawed on the 13th of May 2016 in RIAE Prague. For model and relatively stable conditions was use air-conditioned greenhouse (GH) with regulated irrigation. Used tools: scale, 15 containers, trowel for soil and fertilizers, mix container, containers with fertilizers, permanent marker.

Prior to sowing, all samples of fertilizers were weighed and put into boxes. The soil was sorted from stones and waste and weighed as well. The mixture of ecological fertilizers was homogenized in mixing container. Then the soil was added and mix equally together with the fertilizers. Further the mixture of enriched soil was put into prepared containers with marked description of certain fertilizer and plant.

All the containers were placed in GH with south-west orientation. Exact position in GH for each container was selected accidentally because of dissimilar air condition due to the air flow from ventilators. As well the direct sunshine conditions change during whole day.

Pea and barley seeds were sawed into sowing containers with size 27 cm × 37 cm, i.e. area 1 m^2, depth of substrate was 15 cm. Weight of substrate was 15 kg in each container. The amount of seeds was determined according to requirements for density of sowing, which is 5 – 10 plants (or higher according to the type of soil) per m^2 for pea. Desired depth of sowing is 5 – 7 cm (Selgen, 2016a). 120 seeds of pea were sawed into 10 containers in two lines.

Required density of sowing for barley is 250 – 350 seeds per m^2. The depth of sowing is recommended in 3 cm (Selgen, 2016b). 300 seeds of barley were sawed into 5 containers in two lines.

4.5.8 Measurement of plants´ growths

A field (GH) experiment with pea was measured for the first time after two weeks from sawing. Barley is plant with fast growth and therefore it was possible to measure the plants already after one week. A germination capacity success of pea was 94 seeds in total, for barley the success represented 273 seeds in total. The heights of plants (plant´s grow) were measured regularly each week. The record period lasted for 6 weeks for each plant until the growth stop. Pea had begun to bloom and barley had stopped to grow after grow up the leaves of the plants.

4.5.9 Harvesting of the field trial and preparation of plants to drying

The field trial had been lasted until July 2016. Pea was harvested on the 22nd of July after ripen seeds in yellow maturity and dry plant without watering. Barley was harvested on the 8th of July.

First the boxes were moved out of GH and each plant was put with whole root ball out of the container. The support of pea was also removed. The roots of barley were washed in pure water to remove left soil. Further the plants were counted according to belonging to certain container and divided into drying basket and described. At barley the roots were separated from main plant stem. The plants were dried inside for one week on air with natural air flow.

After one week the samples were processed into dryable form to insert into dryer. The roots of pea were separated and the stems were cut on smaller pieces. The hulls with seeds were also separated from mother plant and counted. The leaves of barley were separated from main stem and both were cut on maximum 5 cm of length. Each matter was dried separately in drying dishes.

All plants from certain container (substrate) were kept in the same drying dishes.

4.5.10 Determination of plants´ dry matter/yields

Each dish had identifying number and a table for writing notes and result was made up in MS Excel. The table contained chain of formulas to count final dry matter: dry matter in grams and in percents, moisture in percents and total percentage average of dry matter.

First all drying dishes (tarra) were weighted. Then the cut plants´ samples were weighted together with the drying dishes. Very detailed scale with three thousandths of decimal places was used.

The samples of phytomass were divided into drying dishes according to: roots, stems and leaves (at barley) and on roots, whole stem and hulls (at pea).

Subsequently the phytomass was dried at a temperature of 105°C until constant weight.

After drying process the samples were weighted once more. All the data of weights were put into prearrange Excel table and the dry matter of both plants was counted. The hulls were counted and seeds originated from certain substrate were counted and weighted as well. It was possible to found out the yield of pea per container and tested substrates/fertilizers.

4.5.11 Determination of plants´ elements´ content – homogenization of samples

Firstly on the beginning of the field trial the content of nutrients was measured. And after harvesting and drying of plant samples the content of nutrients was measured as well. It was used dry matter of all samples.

From each sample/container was selected dry mix of all plant´s constituents – part of roots, stems and leaves or hulls, and then mixed and blended by grinder. This homogenized powder was used to determine a nutrient content in pea and barley that provides information how many nutrients the plant had exhausted. The results were compared to results of soil nutrients´ content.

The content of main elements/macro-elements was measured in RIAE and in CULS laboratories. The elements are as following: N, P, K, C, H, Ca, Mg.

The content of other elements/micro-elements was measured in RIAE such as Cd, Pb, Hg, As, Cr, Cu, Mo, Ni, Zn, Al.

To determine the content of these nutrients in the soil was used XRF[5] analyser as well as for soil nutrients´ analysis (Figure 3).

These elements were compared and statistically evaluated.

[5] X-Ray Fluorescence

5 Results and discussion

This chapter provides the findings from the practical research according to the hypothesis and objectives of the thesis and compares it with the relevant findings of other authors. Results of input materials as the ecological fertilizers and nutrients´ demand highly affect plants´ growth. Values of nutrients/elements were established according to the previous measurements of input compounds and ratio of required nutrients was found according to the plants´ demand. This was followed by main results from plants´ growth, amount of dry matter and plants´ yield. Majority of results were noted as arithmetic means and its standard deviation was determined.

The fertilizers were applied in the same composition for both plants garden pea and spring barley. The aim was to insert elementary intake of nutrients into soil, but mainly the organic matter into soil. All the fertilizers are based on the organic matter that provides nutrients that are returned into soil as a fertilizer.

5.1 Classification of tested fertilizers

Determination of Carbon, Hydrogen and Nitrogen content were done according to EN ISO 15104 (2011). Results are shown in following table (Table 11):

Table 11: Content of main elements in classified organic fertilizers [%]

Mark of fertilizers	Fertilizer	N	P	K
1	**Separate**	1.98	0.34	3.52
2	**Separate + ash**	1.40	0.36	8.10
3	**Separate + poultry excrements**	2.34	0.60	4.02
4	**Separate + ash + poultry excrements**	1.68	0.48	6.48

Source: Author, 2016

The effect of carbon and nitrogen content in the fertilizer is crucial to adequate plant´s growth and profitability. The demand on fertilization of spring barley is in quantity optimally about 150 kg N/ha, 30 kg P/ha and 120 kg K/ha. Garden pea requires the amount of nutrients about 20 kg N/ha, 50-70 kg P/ha and 80-120 kg K/ha (Richter and Ryant, 2015; Selgen, 2016a). Garden pea does not require excessive fertilization by nitrogen compared to cereals. According to these requirements was determined a specific quantity of tested fertilizer for various testing samples (see Table 12).

Table 12: Weight of fertilizer samples and element´s content at garden pea/spring barley [kg/ha]

Fertilizer	**Sample weight** (dry matter)	**N**	**P**	**K**
1	**Separate (186 g)**	**Spring barley**		
2	**Separate + ash (220 g)**	150:30:120		
3	**Separate + poultry excrements (170 g)**	**Garden pea**		
4	**Separate + ash + poultry excrements (195 g)**	20:50-70:80-120		

Source: Author, 2016

5.2 Analysis of chemical composition of ecological fertilizers – CHN – macroelements

Determination of Carbon, Hydrogen and Nitrogen content were done according to EN ISO 15104 (2011). Results are shown in Table 13. The effect of nitrogen and carbon in the fertilizers is crucial from viewpoint of nutrients which are required by plants.

C:N ratio in the soil is required in a proportion 20-30:1 ideally. Oelofse et al. (2015) state that negative relationship was found between soil carbon and nitrogen use efficiency for spring barley when is used wrong ration of elements.

For separate was the ratio 20:1 that was acceptable. For poultry manure it was 10:1 and the composition of fertilizer had to be adjusted according to the ratio demand. Parallel situation was for ash from biomass where the ratio was 40:1 and the quantity of fertilizer required lower total amount of applied fertilizer (see Table 13). Remain relating elements in table were described in following subchapter.

Table 13: The contents of the main elements in the materials´ compounds of organic fertilizer, anhydrous state [%]

Fertilizer	N	P	K	C	H	Ca	Mg
Separate BGS	1.98	0.34	3.52	39.20	4.82	4.30	0.43
Poultry excrements	2.94	1.11	4.99	32.50	4.50	-	-
Ash from biomass	0.23	0.40	17.28	7.82	0.39	-	-

Source: Author, 2016

From the measured values is visible that separate and excrements contain high amount of carbon compared to ash from biomass. An amount of nitrogen in separate and excrements are considerable high compared to content in ash.

Tlustos et al. (2013) state content of nutrients in separate that are N 1.5-3 mg/kg, P 0.7-1.4 mg/kg, K 0.3-0.8 mg/kg and Ca 1.5-4.5 mg/kg.

Amount of calcium is almost the same as upper limits. It is one of the important elements to optimize system of nutrients (Hinojosa et al., 2014).

Compared to Marada et al. (2008) digestate in the experiment contained relatively high amount of total nitrogen, but carbon content was lower compared to other organic fertilizers. However, in the present experiment nitrogen content in separate was measured twice higher than the maximum published value and carbon content in digestate was relatively high.

Ash from biomass is an important source of potassium nutrition and contained higher amount of this nutrient than in other fertilizers Obernberger et al., 1997; Biedermann and the Obernberger, 2005) that was confirmed by this study. As well the ash from straw, in comparison with other biomass, has higher pH value (Obernberger and Supancic, 2009; Hinojosa et al., 2014). From the Table 13 is visible that the ash contains four times more potassium in comparison with digestate and excrements.

5.3 Analysis of microelements´ content and limits for other trace elements

Microelements´content was established according EN ISO 15104 (2011). Some of the results (P, K, Ca, Mg) are shown in Table 13.

From analysing is seen that excrements contained significant value of phosphorus, which is almost three times higher than for other fertilizers.

Poultry excrements were considered as a main source of nitrogen due to its considerable value. Klir and Kozlova (2015) and Eagri (2016) established amount of nutrients N 1.8 mg/kg, P 1.2 mg/kg, K 7.1 mg/kg.

Other trace elements (Table 14) were as well important for plants´ growth. Nevertheless these elements have required limits to apply them into soil or presence in organic fertilizers.

The allowed quantity of heavy metals in fertilizers is determined by Regulation No. 474/2000 Coll., assessment of requirements for fertilisers as amended according to the Regulation 131/2014 Coll. (Nitrat, 2016).

Table 14: The content of other trace elements in the materials for organic fertilizer production, anhydrous state [mg/kg]

	V	**Cr**	**Mn**	**Co**	**Ni**	**Cu**	**Zn**	**As**	**Se**	**Cd**	**Hg**	**Pb**
Separate BGS	4.0	13.6	472.1	2.1	1.4	23.9	370.8	0.4	0.3	0.6	0.1	1.4
Poultry excreme	3.2	49.0	357.1	1.0	10.7	50.5	463.2	2.0	0.3	0.4	0.2	1.9
Ash from biomass	6.0	48.1	407.1	1.1	0.7	23.0	149.2	3.1	0.5	1.2	0.4	2.4

Source: Author, 2016

The solubility of heavy metals varies considerably depending on the chemical composition of ash and is affected by specific soil conditions (Ferreiro et al., 2011), nevertheless the ecological fertilizers and other additives tested in the research had an adequate quantity of these elements that were under standard.

The content of the risk elements in fertilizers is determined during the registration or notification of the fertilizer, which is put into circulation. If the fertilizer is not put into circulation, it is recommended to provide laboratory analysis of fertilizer to determine the amount of nutrients and risk elements (see Table 14 and Table 15) (Klir and Kozlova, 2015).

Lansche et al. (2012) defined the content of micronutrients in separates and from their results show that the levels were the following: Mn 450 mg/kg and Zn 560 mg/kg, Ni 12.6 mg/kg. Further Bonetta et al., (2011) and Valeur (2011) set the contents of the risk elements that were the following: Pb 40 mg/kg, Cd 0.4 mg/kg, Hg 0.2 mg/kg.

According to Tulonen et al. (2012) and Zhao et al. (2015) cadmium is considered to be the most problematic element in ashes from biomass because of significant toxicity to biota and high mobility and its concentrations had increased considerably over the last 3 decades. However, the value in this study was in standard limits to use the tested fertilizers.

For comparison to the standards see Table 15 below:

Table 15: The allowed limits of certain elements in the selected fertilizers in dry matter [mg/kg]

Type of fertilizer	**Cr**	**Ni**	**Cu**	**Zn**	**As**	**Cd**	**Hg**	**Pb**	**Mo**
Organic and state fertilizer with dry matter > 13 %	100.0	50.0	150.0	600.0	20.0	2.0	1.0	100.0	20.0
Ash from biomass	50.0	-	-	-	20.0	5.0	0.5	50.0	-

Source: Klir and Kozlova, 2015

Some risk elements in the soil can be immobilized by an ash application by increasing the value of pH (Demeyer et al., 2001; Omil et al., 2007) and thus was necessary to determine its content. The risk of heavy metals release should not be underestimated. The solubility of heavy metals varies considerably depending on the chemical composition of ash and is affected by specific soil conditions (Ferreiro et al., 2011).

Some studies had shown that applications of animal manures or manure composts containing high concentrations of heavy metals can result in excessive accumulation in soil, leading to adverse effects on soil quality (Hang et al., 2009; Zhejazkova and Warman, 2003).

5.4 Analysis of ash content in ecological fertilizers

Ash was a source of many nutrients (K, C, P) necessary for plant growth. The highest amount of ash contains ash from biomass. However, nitrogen content is low in ash (Table 13) and the input of nitrogen to the environment does not increase by the ash application that stated Soucek and Spulak (2006).

Table 16: Ash content in the materials for the production of organic fertilizers, anhydrous state [%]

	Ash content
Separate BGS	18.61
Poultry excrements	29.16
Ash from biomass	89.67

Source: Author, 2016

Ash content was determined to find out a biomass quantity in combusted sample. The considerable value of ash content had ash from biomass that was three times higher than for excrements. Ash content in excrements was twice higher than for separate. The average pattern of ash utilization in commercial fertilizers can has essential nutrients (N:P:K) in a proportion of 0:1:3. (Soucek and Spulak, 2006).

5.5 Evaluation of soil origin and composition

Soil applied in the field trial had pH value 7.32. The best barley thrives on soils with pH from 6.0 to 7.1 (Urban and Vasak, 2016; Selgen, 2016b). For pea is recommended pH value from 6.2 to 7.0 in the soil (Selgen, 2016a). From above mentioned is visible that pH of soil used in the current research is higher than the recommended ones for both crops.

Table 17: Average value of macronutrients in the soil – before/after set up the field trial [mg/kg]

Substrate composition	Garden pea		Spring barley	
	before	after	before	after
P	1,490.85	1,588.16	1490.85	1513.56
K	17,889.60	18,171.38	17889.60	19069.62
Mg	4,693.73	4,597.22	4693.73	5307.58
Ca	15,549.25	15,606.68	15549.25	12469.80

Source: Author, 2016

Table 17 and Figure 4 show values of nutrients that were measured in the soil before fertilizers´ application. Subsequently, the content of elements was measured in the soil after field trial termination when the fertilizers were used in the substrate. Value of fertilizers was taken into consideration and nutrient content was compared according to the planted crops and nutrients´ intake.

More phosphorus was used by barley although it requires higher amount than pea. Compared to these result, pea had utilized more potassium from soil than barley. This is worth mentioning because both plants require similar amount of potassium, which is about K 120 kg/ha (Selgen, 2016b).

Value of nitrogen content in the soil was 4,000 mg/kg before set up of the trial. Urban and Vasak (2016) recommended provide nitrogen fertilization before sowing, which was kept in this research.

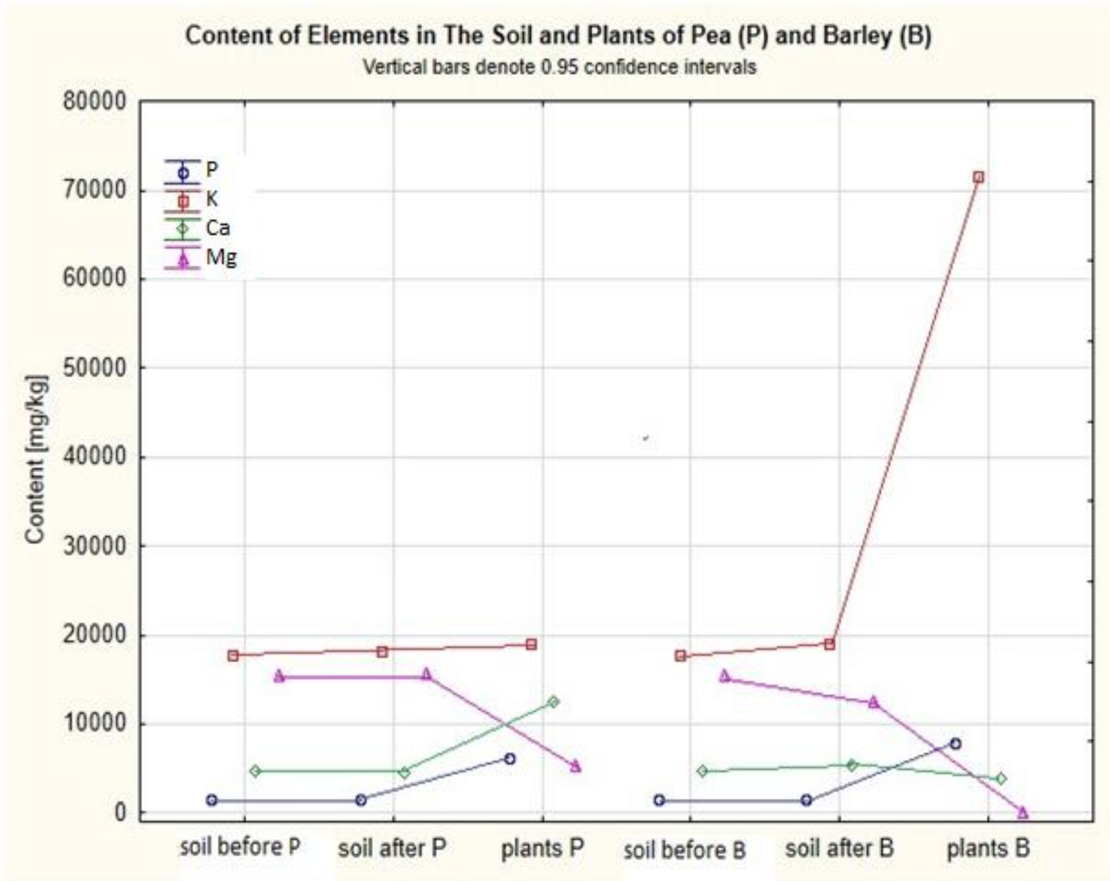

Figure 4: Content of elements in the soil and plants

Source: Author, 2017

5.6 Preparation of ecological fertilizer samples – ratio/weight

A mixture of fertilizers was proposed totally in 5 variations as follows:

Table 18: Ratio of tested ecological fertilizers, weight ratios are given for anhydrous state [%]

Mark of fertilizers	Mixed fertilizers	Weight ratio
0	**Test sample (soil)**	-
1	**Separate**	-
2	**Separate : poultry excrements**	2 : 1
3	**Separate : ash**	2 : 1
4	**Separate : ash : poultry excrements**	4 : 2 : 1

Source: Author, 2016

First sample was used without any additional fertilizer. It is a test sample and testing crops were planted in sole soil. Separate was established as the main part of each tested substrate (1-4). The quantity and nutrients´ content of separate was taken into consideration during determination of weight ratio in combination with other additives.

Nevertheless the fertilizers were applied in the same composition for both plants garden pea and spring barley. The aim was to insert elementary intake of nutrients into soil, but mainly the organic matter into soil. All the fertilizers were based on the organic matter that provides nutrients that are returned into soil as a fertilizer.

5.7 Setting up of field trial

The field trial was performed in greenhouse and lasted for several months in total. It was applied four various ecological fertilizers (see Figure 5) and homogenized with previously analysed soil. Identical substrates were prepared for each test sampling and for two tested crops.

Garden pea and spring barley seeds were sawed into sowing containers into two lines, covered by upper layer of soil and adequately watered.

Figure 5: Samples of fertilizers

Source: Author, 2016

GH trial with spring barley was set up in three weeks after previous trial with garden pea (Figure 6). All containers were marked with appropriate marking for each substrate.

Figure 6: Sawing of spring barley, three weeks plants of garden pea

Source: Author, 2016

5.8 Evaluation of plants´ growths

Both crops garden pea and spring barley were measured regularly once per week which indicate axis X of following figures. On the axis Y is shown height of plants during growing time in mm.

The measurements lasted for 6 weeks for garden pea (see Figure 7 and Figure 8) and 5 week for spring barley (see Figure 8). Samples, marked as A and B (in case of pea), were placed randomly in the greenhouse.

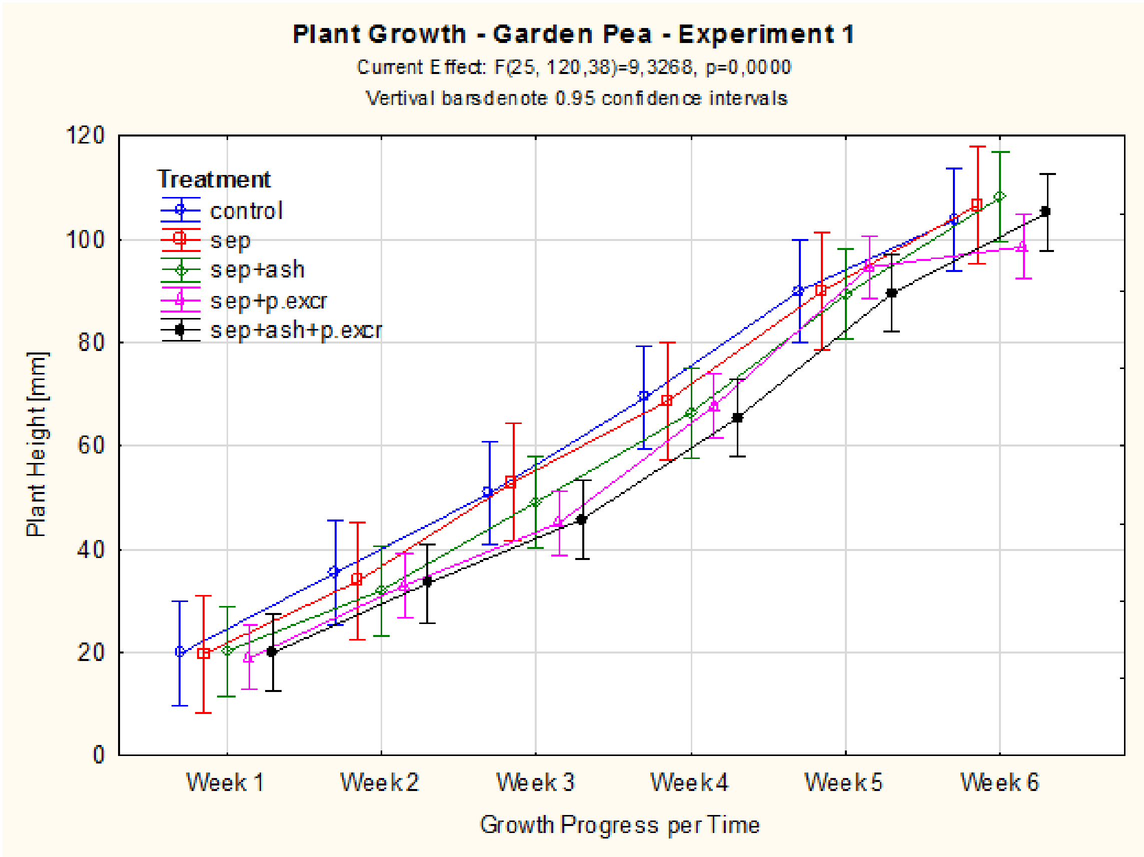

Figure 7: Plants´ growth of garden pea – Experiment 1 (group A)

Notice: One-way ANOVA. Confidence interval determined on the level of significance α=0.05
Source: Author, 2016

Figure 7 shows growth of garden pea in various substrates in regular measurements after weeks. The average growths of peas were very similar in first week of measurement.

Plants in substrate with sole soil and substrate with separate had highest growth almost during whole observed period compared to other in fertilized substrates (from week 2 to week 4).

The growth of plants in substrates with test sample (sole soil) and also separate were highest from the second to fourth week. Combination of fertilizers with separate and excrements had highest increase of growth in week 5. But it was completely inversed in the last week when the substrate with separate and ash had highest total plants´ growth.

Nevertheless these average growths for specific substrates are not statistically significantly. The error lines (values of standard deviations) are intersected between groups (substrates) so it means that values of growths in each substrate are similar.

The requirements for nitrogen intake are lower for pea compared to valuable demands of other crops, thus it may cause lower growths of pea in substrates with higher amount of nitrogen (Vanek et at., 2012; Selgen, 2016a). However, nitrogen is a most limiting nutrient for plant´s production, and therefore it is widely applied into the land in large quantity according to statement of Mikanova and Simon (2013).

Saucke and Ackermann (2006) recommended to grow pea in a well aerated soils, biologically active, with a neutral to slightly acidic pH and supplied enough by Ca and P. In the research was used soil with slightly alkaline pH, which may influence processes in the soil as well as utilization of nutrient. But it is assumed that the nutrient were in right ratio according to initial elements´ analysis.

The absence of positive organic fertilizer effect on crop growths might influence the following factors whose cause a poor germination, emergence, and early plant growth:

- inadequate seedbed after distribution of the organic fertilizers by hand and incorporation by a rotary cultivator,
- formation of large clods especially in manure,
- water deficiency due to water consumption by decomposition (Prochazkova et al., 2002), and
- production of phytotoxic substances during further decomposition (Levy and Taylor, 2003; Roy et al., 2010). This is suggested by delayed seedling emergence and the lower yields per plant in comparison with the control treatment. These problems might be intensified by the generally low plant density that was below the recommended seed rate for field peas of 80 seeds per m^{-2} (Neumann et al., 2007). However in this research was the seed rate adequate to the sowing are which was 0.1 m^2.

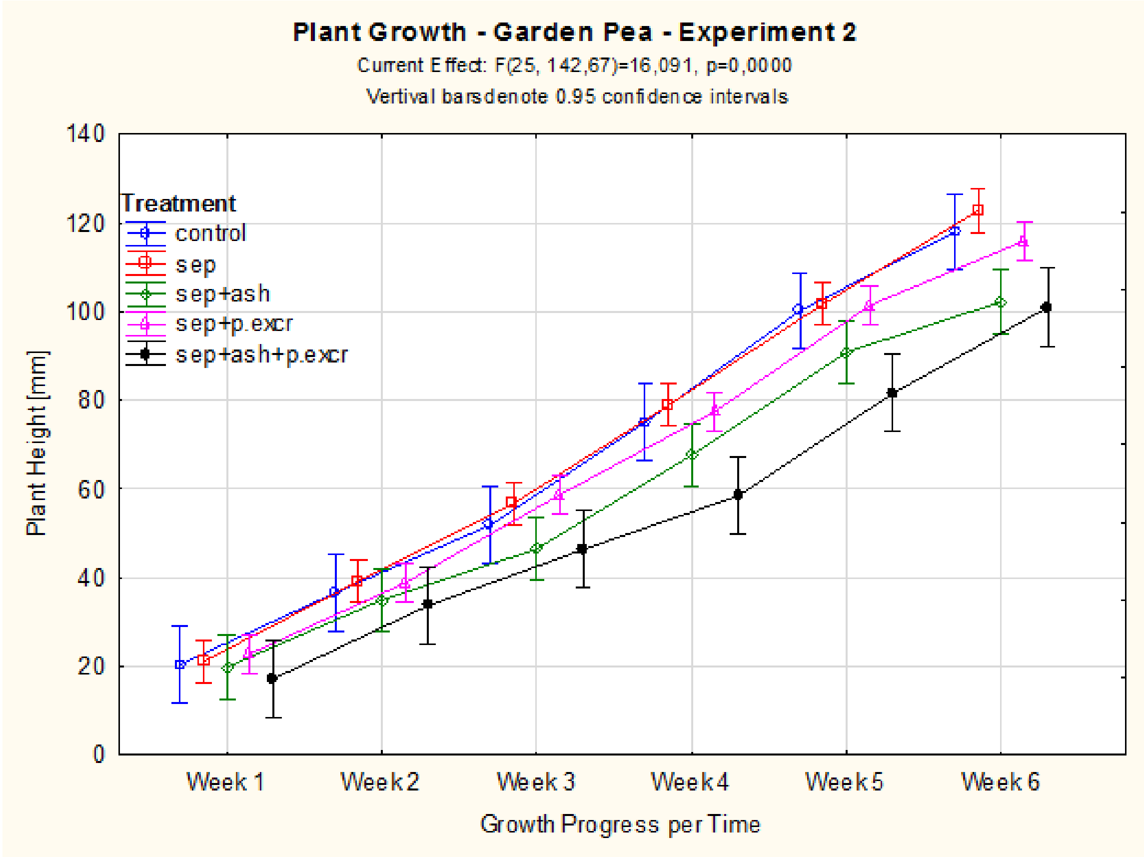

Figure 8: Plants´ growth of garden pea – Experiment 2 (group B)

Notice: One-way ANOVA. Confidence interval determined on the level of significance α=0.05

Source: Author, 2016

Figure 8 shows growth of garden pea in various substrates in regular measurements as previous figure. The growths in first week of measurement are very similar and statistically non-significant as well as in week 2. The differences between plants´ growths became to be higher in week 3, but are not statistically significant.

Surprisingly the substrate with combination of all fertilizers had the lowest average plants´ growth for whole observed period. Vice versa the substrates with lowest fertilization had highest plants´ growths.

Substrate with separate and ash is significantly different to substrate with separate and excrements in week 3.

Plants´ growths (week 4 and week 5) in substrate with combination of separate, ash and excrements are statistically significant compared to the soil and to the substrate with separate and substrate with separate and excrements.

Considerable amount of significant differences occurred in last week. Both, substrate with separate and ash and substrate with separate, ash and excrements, are statistically significant compared to all other substrates (see Figure 8).

Vanek et al. (2012) stated that the phosphorus is received relatively during whole vegetation by plants and it is important to intake adequate amount of phosphorus. According to the Figure 8 is visible that the combination of separate and excrements, which had more significant differences, contains valuable amount of phosphorus for plants´ growth. It was establishes in the amount 1.11 mg/kg in poultry excrements and 0.60 mg/kg in total substrate (separate and excrements) and which represent the highest value in tested fertilizers.

The plants with lower biomass production (pea) evince small consumption of potassium according to Vanek et al. (2012), which was confirmed. Substrate with separate contains lower amount of potassium 3.52 mg/kg and create comparable growths to other fertilizers.

Various differences between tested samples of pea (group A and B, see Figure7 and Figure 8) may occur due to different external effects on plants in the greenhouse (as watering, direct sunshine, air conditions, etc.). However due to the contribution of ecological fertilizer as intake of organic matter into soil was significantly increased nutrients´ content in the substrate (Kaye and Hart, 1997; Mansson et al., 2009). But it was confirmed that garden pea is able to grow in enriched substrates as well as in test sample (sole soil).

Jannoura et al. (2014) researched organic fertilizer effects on grown peas. By comparison the effects of this organic fertilizer and cropping system on pea yield, grown as the sole fertilized crop or intercropped. In general, organic fertilizer application improved properties of substrate and growth of plants that was confirmed by Experiment 2 for garden pea. Organic fertilizers increased the contents of microbial biomass C, N, P.

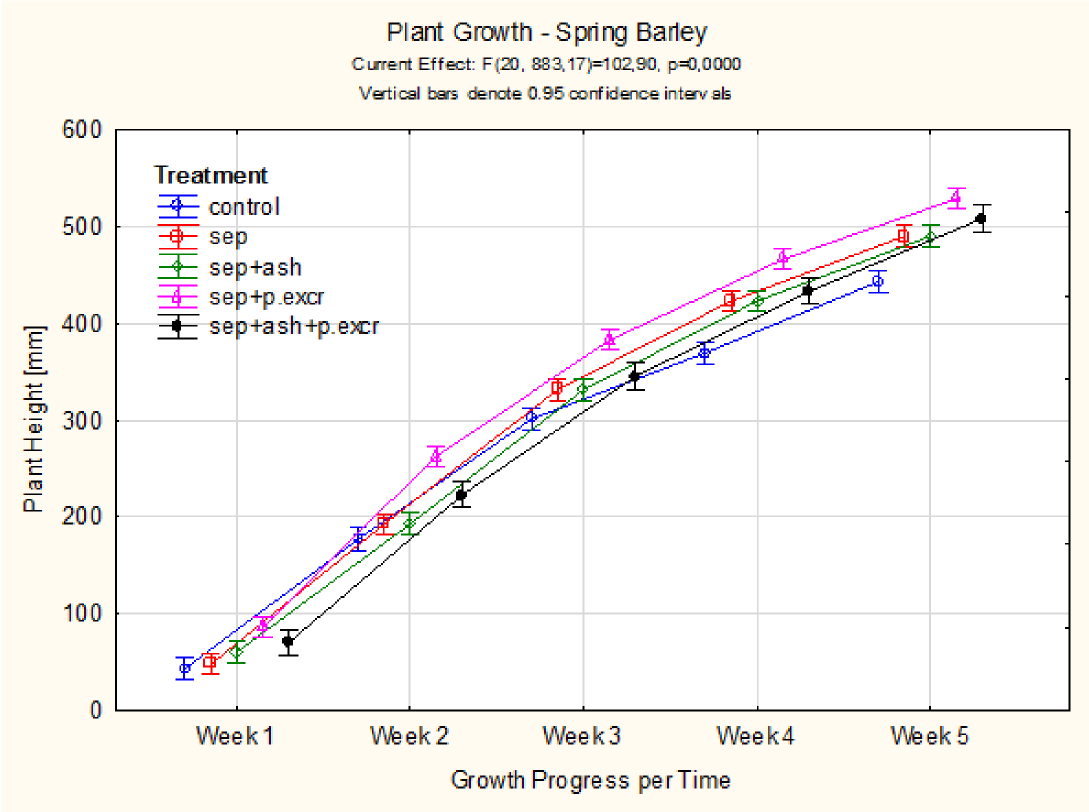

Figure 9: Plant growth of spring barley

Notice: One-way ANOVA. Confidence interval determined on the level of significance α=0.05

Source: Author, 2016

Figure 9 states the growths of spring barley that were progressively increasing between the groups (substrates) during whole observed period. Each enriched substrate had higher plants´ growths than the previous substrate except for plants in substrate with combination of separate, ash and poultry excrements. All growths within a group are statistically significant. As well as the growths between the groups is statistically significant.

We can say that all fertilizer have an effect on height of plant included growing in sole soil.

Barley has significant requirements for nitrogen intake, which is 150 kg/ha (Richter and Ryant, 2016). Thus the fertilizer combination with separate and poultry excrements had considerable effect on plant growth and which had highest value of nitrogen in substrate.

An excessive intake of potassium in substrate with combination of all fertilizers can lead to unwanted effect like accumulation of K in plant´s tissues. That is called a “luxury” consumerism and leads to restriction of other cations (Na, Mg, Ca). Intake of K is in addition to its concentration in the soil solution significantly affected by moisture, temperature and the intensity of solar radiation (Vanek et al, 2012) that correspond to plants with highest average and total growths in substrate with separate and excrements. This container was most likely placed on the best site in the greenhouse.

Urban and Vasak (2016) defined the barley is one of plants that claims considerable nitrogen intake mainly because of production large amount of biomass, and which may also be a specific indicator. However the most sizeable amount of nitrogen was contained in poultry excrements. Poultry excrements are still considered as one of the most effective fertilizers of natural origin according to Ghosh et al. (2004) and Eghball (2000) recommend it to apply as fertilizer to prevent diseases at cereals that can cause slower growths´ increases or damage the plants. Other studies on cereals´ diseases provided in the RIAE greenhouses might cause the slower growth of barley.

Spring barley is one of the most sensitive cereals for fertilizing. Any unevenness in the soil or fertilizer is reflected in imbalances of plants, the yield of grain or its quality. Evenly SOM influences soil biological, physical and chemical properties therefore it is considered as an important contribution in a variety of ways to improve some of the factors influencing crop yield (Christensen and Johnston, 1997).

According to Eichler-Lobermann et al. (2008) the ash might have comparable or even better effect on the yield and quality of plants from substrate with separate and ash that had significantly high growths.

5.9 Harvesting of the field trial and preparation of plants to drying

Figure 10: Drying of plants on air after harvesting

Source: Author, 2016

Garden pea and spring barley were harvested and dried on air for one week on airy sieves. The roots of plants were cleared in water and separated. Further the plants were measured and compared (Figure 10). How was mentioned in previous figure (Figure 9) the range of plants´ height was very similar.

Figure 11: Preparation of plants before drying in oven

Source: Author, 2016

Crops planted in specified substrates/containers were processed and dried separately. The leaves and roots were separated as well before drying in oven and cut for small pieces that were better to use in next processing with the samples.

5.10 Evaluation of plants´ dry matter/yields

Table 19: Yield of grain of garden pea [g]

Variation of fertilizer	Grains´ weight
0	14.70
1	19.65
2	13.19
3	19.32
4	15.11

Notice: 0 – sole soil, 1 – separate, 2 – separate and ash, 3 – separate and poultry excrements, 4 – separate, ash and poultry excrements

Source: Author, 2016

According to the results in Table 19 is seen that highest yields had plants from substrate with separate and with combination of separate and poultry excrements. These results were unexpected because of the various substrate compositions. The both fertilization was very different (separate compare to combination with poultry excrements) and compare to the Figures 7 and 8 with plants´ growth was dissimilar because the growth of plan.

Lowest yield indicated plants from substrate with separate and ash that was even lower than in non-fertilized variation. However other fertilizers had positive influence on the grain yield.

Vanek et al. (2016) stated that phosphorus and potassium is used effectively from the soil and thus leads to adequate amount of yields and whole plant´s growth.

The pea belongs among the crops with less yield stability associated with considerable revenue dependency on weather, which might caused this unexpected yield of grain (Urban and Vasak, 2016). However, the conditions in the greenhouse were expected to be more consistent. One of the main assumptions of the establishment of productive growth is high quality of seeds. In the research were used stained grains stored in a dark dry place, so it is assumed to be in adequate quality.

According to other opinion about cause of the instability of the crop yields and nutrient concentrations of pea grain is not considerable difference in the control treatment. Thus the crop yields were similar to those obtained under organic cultivation (Saucke and Ackermann, 2006).

Kaye and Hart (1997) and Mansson et al. (2009) state that surprisingly, organic fertilizer addition did not enhance the yields of peas. This is the same situation for this study. Moreover it can mean that the added organic fertilizers were rich in easily decomposable components that may result in competition between soil microorganisms and plants for easily available nutrients, especially N.

However, the lack of response cannot be solely attributed to N and P deficiency. As the crop grown in the organic fertilizer treatments exhibited significantly higher P and N concentrations in grain and straw than those grown in the control treatments. Furthermore, pea, as a legume crop, had a relatively low soil N requirement and a low dependency on soil organic N (Mansson et al., 2009).

An imbalance in soil nutrient content might influence the grain yield of peas.

Table 20: Yield of dry matter of spring barley [g]

Variation of fertilizer	Plants´ weight
0	18.75
1	23.18
2	24.12
3	30.30
4	27.82

Notice: 0 – sole soil, 1 – separate, 2 – separate and ash, 3 – separate and poultry excrements, 4 – separate, ash and poultry excrements

Source: Author, 2016

Quantity of dry matter of spring barley (see Table 20) is highest for plants from substrate with combination of separate and poultry excrements. These results correspond to plants´ growth in Figure 9.

Considerably lowest quantity of dry matter had plants from substrate with sole soil. There is confirmed an effect of fertilizers on plants for both, evaluation of plants´ growths and dry matter content.

The higher amount of dry matter of barley might be caused inter alia by soil organic matter that was higher in enriched substrates, mainly in separate with excrements. Further these substrates provide more nutrients for plants, especially the nitrogen in required amount for barley.

The organic fertilizing experiment provided by Johnston et al. (2009) shows that yields of spring barley are higher after regular rotation of more crops. It is possible that the nutrients in the soil had contributing effect on the barley in the present research.

Further contribution to higher yields is due to adequate balance of C:N ratio. This ratio was adjusted according to requirement of plants that was about 20-30:1. Oelofse et al. (2015) stated the significantly positive effect of SOC on potential yields as well. The yield can be attained when sufficient levels of nutrients are available to not limit biomass production.

5.11 Analysis of plants´ element´s content – crushing of samples

Table 21: The contents of the main elements in the materials´ compounds of research plants, anhydrous state [mg/kg]

Plant	P	K	Ca	Mg
Garden pea	6,227.89	19,069.62	12,469.80	5,307.58
Spring barley	7,849.51	71,640.37	3,841.22	-

Source: Author, 2016

Table 21 and Figure 4 show values of nutrients that were measured in the plants after field trial termination. Value of elements was taken into consideration and nutrient content was compared according to the planted crops and nutrients´ intake.

Further the Table 21 and Figure 4 indicates elements´ content in dry matter of the plants after field trial harvesting. Value of potassium in barley is considerable high, which can means storage of this element in the plant due to some imbalances in the soil.

Spring barley is one of the most susceptible cereals for fertilizing. Any unevenness in the soil or fertilizer is reflected in imbalances of plants, the yield of grain or its quality.

Higher values of potassium and magnesium for pea can indicate storage of elements in grains.

The requirements for fertilization by phosphorus for garden pea are almost three times higher than for spring barley (Richter and Ryant, 2016; Selgen, 2016a). From Table 21 is visible that pea contains lower amount of P due to the higher nutrients intake. Although the demand of potassium is the same for both crops, the content of K is more than three times higher compared to the content in pea. It might be cause by storage the element in the plants due to some external effect.

Calcium is one of the important elements to optimize system of nutrients (Hinojosa et al., 2014). The content of Ca is three times higher than for pea. Urban and Vasak (2016) recommend grow pea in a well aerated soils, biologically active and supplied enough by Ca and P. As well soil biomass content was positively related with pea dry matter yields according to a study Jannoura et al. (2014).

6 Conclusion

Ecological fertilizers made from agricultural residues or waste are very important sources of nutrients that can be restored into the soil. This source of nutrients is considered as accessible and uncostly plant origin contributing to increase crops yields. The utilization of high-quality organic fertilizers is required in adequate classification and composition to plants´ demand. Presented Diploma Thesis was focused on the above mentioned problematic, especially on observation of ecological fertilizers´ effect on specified crops and its properties.

Specific quantity of nutrients´ demand was determined for tested crops according to required standard.

C:N ratio in the soil is required in a proportion 20-30:1 ideally. For separate was the ratio 20:1 that was acceptable. Ratio for poultry represented 10:1 and the composition of fertilizer had to be adjusted. Parallel treatment was provided in case of ash from biomass when the ratio was 40:1 and the required quantity of fertilizer was decreased according to the plants´ demand for nutrient intake.

Further nutrient analysis was stated that the measured values in separate and excrements contained high amount of carbon compared to ash from biomass. An amount of nitrogen in separate and excrements was considerable high compared to content in ash. Opposite to this the ash from biomass contained greatly higher value of potassium relatively to the other observed fertilizers.

From analysis was seen that poultry excrements contained significant value of phosphorus, which is almost three times higher than for other fertilizers. It was as well considered as a main source of nitrogen in present study.

Risk element and other trace elements important for plants´ growth had required limits to apply them into the soil or presence in organic fertilizers according to standards.

The risk elements in the soil can be immobilized or raised by an ash that might affect the pH value.

Ash content in fertilizers was a source of many nutrients (mainly K, C, P) necessary for plant growth. The considerable value of ash content had ash from biomass that was three times higher than for excrements. Ash content in excrements was twice higher than for separate. However, nitrogen content was low in ash and the input of nitrogen to the environment did not increase by the ash application.

Garden pea did not require an excessive dose of nitrogen that was confirmed in case of substrate in combination with separate and poultry excrements, which had significantly high plants´ growth.

According to intake of nutrients by plants was evaluated that more phosphorus was used by barley although it required higher amount than pea. Compared to these results, pea had utilized more potassium from soil than barley. It was worth mentioning because both plants required similar amount of potassium 120 kg/ha

The fertilizers were applied in the identical composition for both plants garden pea and spring barley. The aim was to insert elementary intake of nutrients into soil, but mainly the organic matter into the soil. Garden pea was tested in two groups with repeating, which corresponds to the amount of plants sowed per ha. The plants´ growth had similar progress, however, only the repeating measurement was statistically confirmed. The statistically different growths for garden pea were evaluated as following:

- the substrate with combination of all fertilizers (sample 4) had the lowest average plants´ growth for whole observed period,
- the substrates with lowest fertilizer composition (sample 0, sample 1) had highest plants´ growths during whole observed period,
- the substrate with separate and ash (sample 2) was significantly different to substrate with separate and excrements (sample 3) in week 3,
- the plants´ growths (week 4 and week 5) in substrate with combination of separate, ash and excrements (sample 4) were statistically significant compared to the soil (sample 0) and to the substrate with separate (sample 1) and substrate with separate and excrements (sample 3),
- a considerable amount of significant differences occurred in last week, when substrate with separate and ash (sample 2) and substrate with separate, ash and excrements (sample 4) were statistically significant compared to all other substrates.

The plants´ growth of spring barley had progressive growth for whole observed period. The statistically significant results for spring barley were evaluated:

- the enriched substrates had higher plants´ growths than the prior substrates except for plants in substrate with combination of separate, ash and poultry excrements (sample 4),

- the growths within a group were statistically significant,
- the growths between the groups were statistically significant,
- all fertilizers had an effect on height of plant included growing in sole soil.

The values of yields of garden pea were evaluated that highest yields (weight of grain) had plants from substrate with separate (sample 1) and with combination of separate and poultry excrements (sample 3), which had absolutely various composition of substrate. Lowest yield indicated plants from substrate with separate and ash (sample 2) that was even lower than in non-fertilized variation (sample 0).

The weight quantity of dry matter for spring barley was evaluated as highest for plants from substrate with of separate and poultry excrements (sample 3). Considerably lowest quantity of dry matter had plants from substrate with sole soil (sample 0). Higher amount of dry matter of barley might be caused by soil organic matter that was presented in enriched substrates, which provided more nutrients for plants.

Limitations of the study and recommendations

The recommendation to grow of both plants is to use well aerated soils with neutral to slightly acidic pH and supplied enough Ca and P, which may affect process in the soil in case of Experiment 1 for garden pea. The pea belongs among the crops with a less stabile yields influencing considerably by external conditions. Spring barley is considered as susceptible cereal affecting by soil unevenness in the soil or fertilizer is reflected in imbalances of plants, the yield of grain or its quality.

As well an inadequate seedbed after distribution of the organic fertilizers by hand and further formation of large clods especially in manure. Additional effect might be water unevenness in containers.

Thus, the various differences between tested samples might occur due to different external effects on plants in the greenhouse.

However, by the hypothesis of the present Thesis *“addition of ecological fertilizers (such as ash from grain straw, poultry excrements) may improve mixed fertilizer(s) properties based on solid digestate”* was confirmed in case of the contribution of ecological fertilizers as intake of organic matter into the soil that significantly increased nutrients´ content in the tested substrates and positively influenced the plants´ growths and plants´ yields.

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