

# Biogas Digestate Suitability for the Fertilisation of Young *Salix* Plants

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## Abstract

We analysed the possibilities to use the digestate from pig slurry anaerobic digestion as a nutrient supply for Short Rotation Coppice *Salix* species. Our greenhouse experiment revealed that the total biomass (roots, cutting and shoots) of one-year-old *Salix* plants treated with moderate amounts (hereafter 0.5F) of digestate during the vegetative period was more than twice that of control (hereafter C) plants and this difference increased in time. After the first vegetative period both shoot and root biomass production of 0.5F plants were also significantly higher than those of plants supplied with mineral NPK fertiliser at optimal N load (hereafter MIN). Although the digestate application at optimal N load (hereafter F) harmed the plants by damaging the roots during the first vegetative period, they were able to recover and annual production was comparable with C plants during the following year. The MIN plants supplied with the same N load as F plants died back during winter despite the larger supply of K. The largest load (hereafter 2F) of digestate caused serious problems and was lethal to one-year-old plants. By the end of the experiment the pH of the growing substrates treated with digestate or mineral fertiliser was lower than in control pots. Less ammonium N was available but the K and P content increased in growing substrates following digestate application. Our results reveal that digestate can be an alternative fertiliser in SRC and the nutrients from digestate are more available for *Salix* plants than those from mineral fertiliser.

**Key words:** biogas digestate, biomass, fertilization, *Salix*, shoot growth rate

## Introduction

Short Rotation Coppice (SRC) of willows (*Salix*) is assumed to be one of the most promising second generation energy crops for the northern Europe (Christersson and Sennerby-Forsse 1994). However, its economically feasible yield can be achieved only through supply of additional nutrients. Use of mineral fertiliser is costly and its production requires large fossil energy consumption. Therefore technologies that substitute mineral fertilisers with other sources of required chemical elements (mainly N, P and K) are favoured in modern and sustainable agriculture (Dimitriou and Rosenqvist 2011). To date the most popular alternative nutrient resources for SRC fertilisation are pre-treated sewage sludge, bottom ash from wood boilers and municipal wastewater. The application of sewage sludge to SRC supplies plants with P, which may increase the crop yield on poor soil sites (Hasselgren 1998; Holm and Heinsoo 2013). However, in this case hygienic risks or contamination of agricultural land with heavy metals, e.g. Cd must be avoided (Heller et al. 2003). Wood ash is usually exploited together with sewage sludge to provide additional K to the plants (Adler et al. 2008). The most promising al-

ternative nutrient resource for SRC is municipal wastewater after mechanical pre-treatment and reasonable storage time to minimize the hygienic risks. This kind of wastewater usually contains nitrogen (N) and phosphorus (P) in a ratio that is optimal for *Salix* growth. Application of these nutrients along with water to SRC increases the wood yield in most cases as water availability is considered a major limiting factor of *Salix* growth in the region (Aronsson et al. 2002). Technically the irrigation of SRC is limited by wastewater transportation costs. Moreover, the applicability of this particular SRC fertilisation method also depends on local legislation (Heinsoo and Holm 2008). Due to limitations in local monitoring techniques for groundwater quality some countries have difficulties in considering such vegetation filter methods to be sustainable or environmentally safe.

Recently a new co-product of bioenergy production has come to the market. The increase of biogas production from agricultural substrates (e.g. manure, maize) in Europe has resulted in large amounts of liquid co-product digestate, which in general has very high N content (Poeschl et al. 2010). This residue has to be utilised carefully in order to avoid contamination of watersheds, but on the other hand, it can be

used as an alternative fertiliser, if the application can be carried out in a sustainable way without significant environmental impact (Weiland 2010), particularly the emission of  $\text{NH}_3$  (Rehl and Müller 2011). In order to minimize pollution and diminish the cost of digestate utilisation and transportation, crops with high annual productivity and large N demand should be preferred. In addition, multi-annual crops should be favoured as in that case the growing substrate contains a larger proportion of roots for filtering the supplied material and well-developed rhizosphere enables faster aerobic degradation of N compounds. These requirements make SRC to become a reasonable choice for digestate utilisation.

In recent years a lot of information about the digestate has become available. The main issue here is to give adequate input to the life cycle analysis of the biogas production as a whole (Patterson et al. 2011, Budzianowski 2012). Results can be variable as the content of organic material, N, P and K in digestate depends on the anaerobic digestion technology and feedstock used (e.g. Tambone et al. 2010). Furthermore, digestate from different sources may also have variable effects on the physical and mechanical characteristics of soil (Beni et al. 2012). In general, application of biogas digestate runs a lower hygienic risk than spreading fresh manure because of the thermal treatment during digestion process (Goberna et al. 2011). However, usage of slurry for biogas production may result in highly variable concentrations of critical chemical elements (e.g. Zn, Cu) in digestate. Digestate from the biogas plants that also use sewage sludge or other industrial wastes as primary raw material may contain such large amounts of heavy metals as to make the application of it to agricultural land impossible. Therefore the international harmonisation of the requirements for digestate quality acceptable for agricultural fertilisation is required (e.g. Albuquerque et al. 2012). So far national standards are available only in some countries. In the situation, where biogas technology will meet both the needs of sustainable agriculture and bioenergy production, the digestate application to farmland can improve the local N use efficiency and can be suggested for including in organic farming systems (Gunnarsson et al. 2011) and considered for rural economic development plans (Garfi et al. 2011).

Due to the high variability of the chemical characteristics of digestate, it is not easy to evaluate its influence on the yield of agricultural crops. There are data available that digestate application can increase the number of soybean pods and their yield even if digestate treatment has burned the plants and negatively influenced their growth (Makadi et al. 2008). In

organic farming, application of pre-digested ley crop and beet foliage effluent has increased the yield of marketable red beets by more than 50% (Gunnarsson et al. 2011). Also the yield of potatoes in digestate treated plots was higher than that of control area (Garfi et al. 2011).

As the information about the influence of digestate on SRC was very limited we conducted a greenhouse experiment to diminish the number of uncontrolled factors and avoid environmental risks. In our experiment we used *Salix viminalis* genotype, which has been studied in SRCs of different countries for years. The biogas digestate used, originated from a biogas plant treating only pig slurry. Such a choice seemed to be reasonable because pork production is one of the main branches of the Estonian agricultural industry and utilisation of large amounts of pig slurry via biogas production is one of the main interests of our farmers. The aim of the experiment was to find out the possible impacts of digestate utilisation on young *Salix* plants and the response of the plants to the treatment with different nutrient loads/resources. The questions we wanted to be answered during the experiment were:

- What is the optimal load of digestate that can be applied to the young *Salix* plants without negative impact to the plants?
- What impact has the digestate application on the biomass production of *Salix* plants?
- Does digestate application cause any change in the biomass allocation between different plant parts (mainly root/shoot ratio)?
- How does digestate application influence the growing substrate's chemical characteristics?

## Material and Methods

The experiment was carried out during two consecutive vegetative periods of 2006 and 2007. In May 31<sup>st</sup>, 2006, we planted 50 cuttings of 25 cm long originating from *Salix viminalis* into specially designed pots. The planting material originated from one-year-old shoots of the *Salix* clone 78183 according to Swedish clone numbering system. Each cutting was planted vertically into the growing substrate according to its original orientation so that no more than 1...2 cm of the cutting remained above the substrate surface.

In principle each pot consisted of two buckets put into each other. The inner vessel with the volume of 5 litres had three drilled holes in the bottom. Through each cavity approximately one third of 30 cm-long textile wick reached to the bottom of outer vessel. The outer vessel constantly contained water up to the bottom level of the inner vessel. The water level was

controlled and water added on demand at least twice per week. In case of heavy rainfall or excessive irrigation the extra water was removed from the outer vessel via the hole in vessel's wall at the bottom level of inner vessel. With the described design we ensured that the plants growing in pots had sufficient water supply and water was not a limiting factor of plant growth.

Before planting the pots were filled with sandy soil. The substrate, which was very rich in silica, was used in order to diminish the amount of natural nutrients in the growing substrate. Moreover, this substrate was also assumed to be a reasonable option enabling the collection of all the fine roots of the plants according to the experiment scheme. During both vegetative periods the pots with plants were stored in the open-roof greenhouse, where they were protected from wind and vandalism but opened completely to the sunshine and precipitation. During winter 2006/2007 the pots with 25 plants that remained for experiment in 2007 were taken to the garden, where each inner vessel was partly buried into the ground so that the substrate level in the pot remained at ground level. With this treatment we simulated the natural growing conditions of plant roots during winter period. Throughout the vegetative periods the pots were regularly checked for mechanical elimination of weeds and pests (ants invaded some pots).

In the experiment, two different types of fertilizer were used. Liquid fraction of digestate originated from commercial biogas plant at Saaremaa Island, which was the first enterprise to produce biogas from pig slurry in Estonia. This liquid was the co-product of anaerobic digestion separated from the digestate by centrifuging. The liquid was transported in plastic containers to the greenhouse in both springs and stored in a cellar at a temperature of 5...10 °C throughout the experiment. The chemical composition of liquid digestate (Table 1) was analysed in an accredited laboratory of the Agricultural Research Centre of Estonia (L024) prior to application in the experiment. The amount of applied digestate was calculated on the basis of its nitrogen content. We assumed that optimal N requirement of plants corresponds to the typical fertilisation level in mature Short Rotation Coppice (SRC) (200...250 kg N per ha). In respect of an SRC plant density of 20 000 plants per ha the requirement of a single *Salix* plant corresponds to 10...13 g N per year. In order to analyse the impact of digestate load we had besides optimal digestate load (F) also half (0.5F) and double (2F) digestate load series in our experiment. To compare the test series with those without any additional fertilisation and with mineral fertilisation both treatments were included to the experiment (C and MIN,

respectively). The mineral fertilisation was adjusted to provide an N application load comparable to the optimal (F) treatment. In the beginning of our experiment 10 healthy young plants were randomly chosen for each of our test series. The fertilisation treatment started in a month after planting.

**Table 1.** Loading of critical chemical elements in applied fertilisers

	concentration in fertiliser mg l <sup>-1</sup>	liquid digestate				mineral fertiliser	
		C	0.5F	F	2F	concentration in fertiliser g kg <sup>-1</sup>	MIN g week <sup>-1</sup>
pH	8.66						
N	5000.00	0.00	0.50	1.00	2.00	170.00	1.00
P	530.00	0.00	0.05	0.11	0.21	31.00	0.18
Ca	890.00	0.00	0.09	0.18	0.36		
Mg	110.00	0.00	0.01	0.02	0.04		
K	2100.00	0.00	0.21	0.42	0.84	116.00	0.68
S	160.00	0.00	0.02	0.03	0.06		

Throughout vegetative periods the plants were supplied weekly with fertilisers according to the experiment scheme (Table 2). We diluted the dosage of the fertiliser in 1 litre of tap water for each particular plant and poured the solution to the pots gradually throughout the week taking into account the natural precipitation and avoiding over-irrigating and loss of the liquid from the emergency outflow of the outer vessel. Our initial aim was to supply the nutrients 13 times during both vegetative periods despite the treatment. However, in 2006 we did not achieve this goal as the pots treated with digestate formed a thick layer of biofilm on the surface of growing substrate. Therefore we had to skip the irrigation with digestate in some weeks during the vegetative period. The regular visual evaluation also confirmed that plants supposed to be treated with the highest load of digestate (2F) started to suffer from over-fertilisation during the first month of experiment. This was another reason why we decreased the number of irrigations of this treatment during July and August 2006. However, some plants of that treatment died during the first vegetative period and we had to exclude them from our study. In 2006 the last irrigation was performed in the beginning of September. At the beginning of the following vegetative period we had to postpone the digestate application for some weeks as the over-wintered plants suffered from drought and we wanted to be sure that the plants were in good health before continuation of our experiment. Unfortunately, we were not able to vitalize the plants treated with mineral fertiliser during 2006. As also the number of plants that got the maximum dosage of digestate (2F) had decreased drastically by the vegetative period of 2007. Therefore we had to exclude both MIN and 2F treatments from the experiment in 2007.

**Table 2.** Fertilisation scheme of different treatments described by the N load applied

	year	C	0.5F	F	2F	MIN
amount of N per week (g)		0.0	0.5	1.0	2.0	1.0
total amount of N (g)	2006	0.0	6.5	11.0	12.0	13.0
total amount of N (g)	2007	0.0	5.0	10.0		

At the end of the first vegetative period we selected randomly 5 plants per treatment for biomass measurements. From each plant we removed all shoots with sharp scissors near the surface of initial cutting and chopped the shoots for packing into paper bags. The inner vessels of the pots were emptied into a sieve. With careful usage of running tap water and tweezers we separated the root system from the growing substrate. The exposed root system was carefully divided into two components, initial cutting (further on referred as stem) and the fine roots. The stem was cleaned from growing substrate by washing. All the fine roots of the plant were collected onto a sieve and bathed carefully several times in clean standing water. The last procedure was needed to ensure that none of the finest roots got lost during the washing. On the other hand, we wanted to be sure that all the mineral particles of growing substrate were separated from the roots. All the paper bags with shoots, stems and fine roots were labelled with treatment and plant index and dried at 85 °C for 3 days for dry weight estimation. In 2007 we repeated the same procedures with the survived plants from C, 0.5F and F treatments.

For statistical analyses SAS 9.2 software package (SAS Institute Inc., Cary, NC, USA) was applied. For analysing the influence of treatment and growing year on biomass production SAS GLM MANOVA procedure was used. Multiple comparisons of biomass and its allocation by treatments were carried out with the Ryan-Einot-Gabriel-Welsch Multiple Range (REGWQ) test. The confidence level of all analyses was set at 95%.

**Results**

The biomass production of studied plants depended significantly on the treatment and the year of measurement (GLM MANOVA  $p < 0.0001$  in both cases). The stem (initial cutting) biomass did not have significant influence on the root or shoots biomass in any treatment in 2006 (GLM ANOVA  $p > 0.05$  in all cases). During the 1<sup>st</sup> year of experiment, the largest biomass was gained by 0.5F plants followed by MIN and C plants (Table 3). The average sum of shoot, stem and root dry biomass per plant were 85.1, 55.1 and 35.4 g for 0.5F, MIN and C treatments, respectively. The average biomass of F plants was a little smaller than that of C plants, 30.2 g. The poorest growth was performed by the 2F plants with average dry weight of 12.8 g. The majority of plants of this treatment were dead in the first autumn and therefore excluded from the measurements of the year 2007. The MIN plants did not survive the winter 2006/2007 and were also excluded from the experiment in 2007. At the end of the 2<sup>nd</sup> vegetative period the largest average biomass (189.2 g) was obtained once again by 0.5F plants with their biomass almost threefold compared with C plants (65.8 g). The annual increment of 0.5F plants was more than 110%, whereas for both C and F plants it remained less than 100%.

The contribution of different plant parts to the total biomass was also significantly treatment-specific (Table 3). In 2006 the stems of 2F plants were on average significantly smaller than those of C plants. Among the remaining treatments no significant differences in the stem biomass were found in 2006. More significant differences were found in the biomass production of newly-sprouted shoots of the studied plants. In 2006 the largest average shoot biomass was achieved by 0.5F plants, 46.3 g. This was almost twice as large as the biomass of the second best shoot-producer, MIN treatment. Moreover, the shoot biomass

**Table 3.** Biomass production (g) of various *Salix* parts for different treatments, total biomass production (g) and the shoot/root ratio of *Salix* in case of different treatments. *S.E.* is standard error ratio from the mean; REGWQ is different letters indicating significant differences between treatments according to SAS GLM REGWQ test (n = %)

Year	plant part	C			0.5F			F			2F			MIN		
		Mean	S.E.%	REGWQ	Mean	S.E.%	REGWQ	Mean	S.E.%	REGWQ	Mean	S.E.%	REGWQ	Mean	S.E.%	REGWQ
2006	stem	18.90	0.10	a	17.96	0.15	a; b	17.74	0.16	a; b	10.17	0.09	b	17.92	0.12	a; b
	shoots	6.51	0.15	c	46.28	0.11	a	9.95	0.19	c	2.06	0.45	c	25.70	0.14	b
	roots	9.99	0.21	b	20.90	0.17	a	2.66	0.17	c	0.59	0.45	c	11.44	0.10	b
	total biomass (g)	35.40			85.13			30.35			12.82			55.05		
	shoot/root ratio	0.65			2.21			3.74			3.52			2.25		
2007	stem	20.32	0.05	a	20.18	0.10	a	16.16	0.11	a						
	shoots	24.98	0.08	b	118.14	0.06	a	29.08	0.20	b						
	roots	20.48	0.12	b	50.84	0.07	a	15.42	0.12	b						
	total biomass (g)	65.78			189.16			60.66								
	shoot/root ratio	1.22			2.32			1.89								

of C, F and 2F plants was more than fivefold lower than that of the most productive 0.5F series. At the end of second vegetative period 0.5F plants had still the largest shoot biomass. However, in this year the relative differences between 0.5F plants and C or F plants were decreased to fourfold. This was the result of the very rapid relative growth of shoots by C and F plants in 2007 (3.8 and 2.9 times by C and F plants, respectively). The annual biomass increment of 0.5F plants was 2.5 times in 2007.

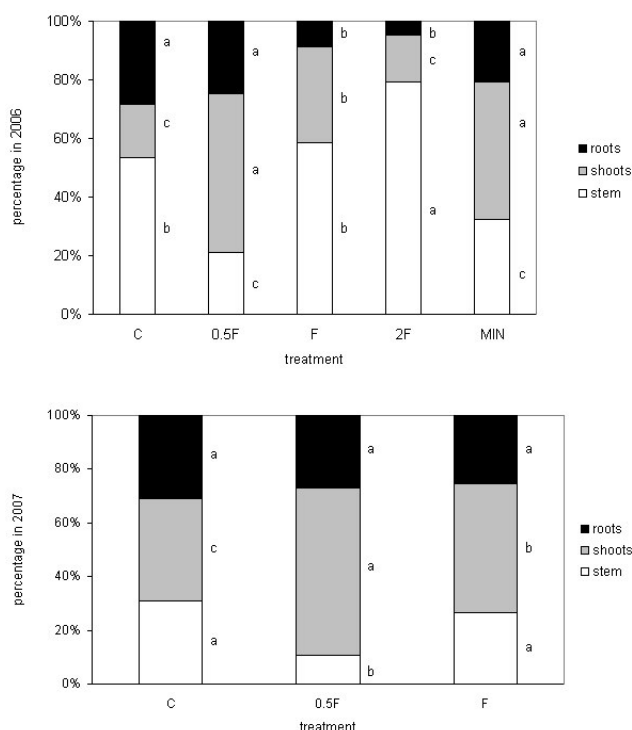
Large differences were also found in the root biomass of different treatments. The most productive were the 0.5F plants with an average of 20.9g of root biomass in 2006 and 50.8 g in 2007. There were no large differences in the root biomass between MIN and C plants. Both F and 2F plants demonstrated poor root growth in 2006. During the second vegetative period the largest annual increment of roots was found in F plants (5.8 times) and the root biomass of F and C plants was comparable (annual root increment twofold in both cases).

For biomass allocation strategy analysis between different plant parts we used the relative biomass distribution data of studied plants (Figure 1). For both study years we found different patterns here. In 2006 the smallest proportion of roots was developed by F and 2F plants. For MIN, 0.5F and C plants the relative

biomass of roots was 20.8%, 24.6% and 28.2% of total biomass, respectively. In 2007 the plants from F treatment also increased their root biomass up to 25% of total biomass and no statistically significant differences in biomass allocation to roots were found between the treatments. In 2006 the shoot biomass ratio was the largest for 0.5F and MIN plants, 54.4% and 46.7% respectively. Both C and 2F plants had shoot biomass less than 20% of the total plant biomass in this year. In 2007 the shoot biomass ratio of 0.5F plants increased up to 62.5% and the shoot biomass ratio of F plants was also found to be significantly higher than that of C plants (48% and 38%, respectively). No statistically significant differences in the stem biomass ratio between treatments were detected.

The shoot/root ratio varied between the treatments more than six-fold in 2006 (Table 3). The smallest (0.7) ratio was for C plants. Both 0.5F and MIN plants had 2.2...2.3 times larger shoot biomass compared with root biomass. The largest ratio was found for 2F and F plants (3.5 and 3.7, respectively). After the end of the second vegetative period the shoot/root ratios were 1.2, 1.9 and 2.3 for C, F and 0.5F plants, respectively.

Limited number of soil samples did not allow statistical analyses of the soil data and therefore only general trends in the treatment-induced changes were detected (Table 4). Despite the application of digestate with a strong alkaline character (pH=8.7) all soil samples that were treated with this material had lower pH than that in control pots. Moreover, the proportion of NH<sub>4</sub> content was larger in the soil of C and MIN trails. By the end of the first vegetative period the amount of P in soil that was treated with mineral fertiliser was larger than that in the soil of 0.5F treatment scheme after two years. On the other hand, fertilisation with digestate in F treatment over two years resulted in a larger amount of P in soil than after one year of experiment with mineral fertiliser. At the same time, the concentration of K in the soil of F treatment after two years was only ca. 10% higher of that in the soil after one year of mineral fertiliser application.



**Figure 1.** Biomass distribution between different Salix parts in 2006...2007. Values marked with distinct letters differ from each other significantly

**Table 4.** Chemical characteristics of growing substrate after completion of the experiment

	C	0.5F	F	2F*	MIN*
pH <sub>KCl</sub>	6.59	6.05	5.94	6.07	5.09
organic matter %	2.96	4.72	4.01	4.50	4.00
NH <sub>4</sub> -N (mg/kg)	14.10	8.70	2.10	6.00	18.20
N %	0.15	0.26	0.23	0.25	0.22
P (mg/kg)	97.50	218.40	314.00	214.30	260.60
K (mg/kg)	260.50	432.90	597.90	922.00	533.40

\* For C; 0.5F; F treatments the duration of experiment was two vegetative periods, for 2F and MIN treatments one vegetative period

## Discussion and Conclusions

### *Plant dieback analysis*

In order to avoid environmental risks we carried out the experiment in a greenhouse. Therefore the trial was conducted with young, recently sprouted plants instead of mature SRC having well-developed root system. Youth of the plants is the most obvious reason why the plants in the experiment suffered from digestate and mineral fertiliser overloads. When designing the experiment we assumed that the optimal load of N for *Salix* plants corresponds to 200...250 kg per ha per year (Perttu and Kowalik 1997, Heinsoo et al. 2002). However, the results of our experiment revealed that for young *Salix* plants this load was an overestimate and application of that amount of N with digestate probably burnt the newly formed roots of 2F plants during the first months of the experiment.

The other technical drawback that we faced during the digestate application was the very poor infiltration of the digestate solution to the growing substrate. According to the literature (Beni et al. 2012) digestate application does not change the macroporosity of the soil. Complications caused by biofilm layer forming on the growing substrate surface and low absorption rate of digestate in our experiment, were most probably caused by the larger amount of undigested organic matter in the digestate.

Another unexpected problem in our experiment was the death of MIN plants during the winter 2006/2007. Although their root and stem biomass were as large as those of control plants in 2006 and the plants looked healthy in autumn, most of them did not resprout in spring 2007. This result may indicate that the young plants did not tolerate periodical N application during the whole first vegetative period and their physiology was not prepared for wintering. Even the relatively large amount of K in mineral fertiliser that is used to support plant preparation for wintering in agriculture (e.g. Bilderback and Bir) did not diminish the problem in our experiment. Most obviously the similar amount of N in F series compared with MIN treatment was in a form that was easier to uptake by plants and therefore the F plants were able to prepare for winter. Larger amount of ammonium N contained in digestate compared with manure is reported earlier (e.g. Holm-Nielsen et al. 2009). The risk of lower survival of MIN plants should be kept in mind while designing a wastewater irrigation scheme for SRC, because nitrogen content is the main limiting factor in typical municipal wastewater used for vegetation filter irrigation. Therefore the irrigation of young SRC plants or during the final weeks of the vegetative period can be critical for vegetation filter overwintering as well.

### *Biomass production*

It has been reported previously that *Salix* shoot production during the first months after planting depends on the size of the cutting used (Verwijst et al. 2012). In our experiment the stem (initial cutting) biomass did not correlate significantly with shoot or root biomass production in any of treatments at the end of the first vegetative period (after three months of growing). In four of five groups ANOVA indicated  $p > 0.5$ . Most probably this can be explained by a longer vegetative period in our experiment. Besides random choice of plants in the beginning of the experiment also the fact that shoot/root biomass was not correlated with that of the stem at the end of the first vegetative period enabled us to exclude the cutting size parameter from further biomass production analysis.

Throughout the experiment the most productive group in terms of biomass production was 0.5F. During the first year of experiment the stem growth of 0.5F plants was similar to that of C and MIN plants, but the production of roots of that group was twice that of C or MIN treatments. Even larger differences with similar pattern were detected in shoot growth. While the reason for different shoot growth between C and 0.5F groups is obviously the lack of nutrients in growing substrate without fertilisation, the differences between 0.5F and MIN plants are more difficult to explain as MIN plants got more nutrients (Table 1). Hence we have to assume that in digestate these were in a more available form for plants than in mineral fertiliser. Advantages of digestate compared with mineral fertilisers have been reported also earlier (e.g. Weiland 2010). Another explanation for the differences in biomass production of MIN and 0.5F plants in 2006 is that different microorganisms become available to plants during digestate application and this facilitates nutrient assimilation. This assumption sounds credible to explain also why MIN plants were not able to overcome the winter. The 0.5F and F plants with similar treatment duration survived the winter successfully, even when the F plants indicated poor growth and most probably suffered from applied digestate load in summer 2006.

The rationale for poor growth of 2F plants in 2006 was already discussed in the section about plant survival. Significantly smaller production of fine roots in this group compared with most of other treatments confirms our hypothesis of unsuitability of such a large digestate load to young plants during their active rooting period.

Despite the significantly smaller biomass of roots of F plants compared with most of the other treatments in 2006, these plants were able to recover and their

biomass was in the same range with C plants in the end of 2007. Very rapid growth of this group in 2007 leads to the assumption that with an even longer experiment their biomass would have exceeded that of C plants. Therefore we can conclude that for fertilisation of *Salix* for bioenergy purposes mineral fertilisation can be substituted with the use of liquid digestate. Special care has to be taken not to burn the young plants by applying too large a digestate load and not to pollute the plantation area with heavy metals that can be easily taken up and removed from the SRC plants (Adler et al. 2008).

Both from the physiological and economical point of view it is interesting to analyse the biomass allocation between different parts of plants. Usually it is assumed that smaller shoot/root ratio indicates the shortage of water or nutrients and the larger ratio is a prerequisite for plants to capture more light and C through photosynthesis (e.g. Grace 1997). This pattern has also been confirmed for plants growing in fertilised/unfertilised plots of SRC (Heinsoo et al. 2009). The current study revealed that the shoot biomass of one-year-old plants (0.5F and MIN) was twice as large as that of fine roots. After the second vegetative period plants from F treatment that had very high shoot/root ratio of 3.7 in 2006 and also gained a shoot/root ratio comparable to that of 0.5F plants. In C plants this ratio increased from 0.7 to 1.2 by the end of second vegetative period. Therefore we can assume that for growth modelling shoot/ root ratio in a range 1.9...2.3 can be applied for SRC plants anticipated to have sufficient water and nutrient supply. In order to increase the harvestable wood yield of SRC, moderate additional fertilisation of young plants during the first years to minimize the biomass allocation to roots can be suggested.

#### *Substrate characteristics*

Limited number of soil samples and shortage of sampling procedures enabled us to draw only general conclusions about treatment-induced changes in soil main characteristics. Soil analysis confirmed previous knowledge indicating that digestate application is an option to increase C deposition in soil and to improve the soil quality. Besides the direct mechanical effect of organic substrate application rapid development of microbial activity could also be expected in the growing substrate (Albuquerque et al. 2012). Increased microbial activity is also most probably the reason for organic material growth in MIN trial growing substrate. According to the literature, the microbial biomass content and enzymatic activity of different organic fertilisers or treatments can be variable (Albiach et al. 2000). Our previous study has revealed that

wastewater application did not significantly affect the amount of microorganisms in soil but changed its diversity (Truu et al. 2009). Therefore, in further studies it would be necessary to analyse the changes in microorganism diversity after digestate or mineral fertiliser application as well. Most likely the majority of them belong to the group that produce acidic residues, because this could be the simplest explanation to the decrease in the pH after continuous application of alkaline digestate in our study. Another possibility is that the change of pH is caused by the surplus of K in the substrate caused by digestate application. The additional deposition of P and K in the substrate with digestate application should be taken into account while planning long-term experiments in the field or practical usage of digestate for SRC fertilisation in order to avoid long term decline in soil quality or the leakage of nutrients. Therefore local soil characteristics should be taken into account when planning for digestate application. The total N content was also slightly higher in fertilised substrates, but the proportion of which was tied up in ammonium was smaller compared with control substrate. Most probably this reflects the increased uptake of N by larger plants. However, it is possible that control plants without biofilm on the growing substrate surface are able to fix air N by endophytic bacteria. Such endophytes have been reported to increase the growth of both *Salix* and *Poplar* species (Doty et al. 2011, Wuehlisch, G. 2011; Knoth et al. 2014). However, as long as atmospheric N was not available for fixation by treated plants due to biofilm, we have to conclude that the atmospheric N was insufficient to support biomass production levels comparable with plants that were treated with biogas digestate.

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## ПРИГОДНОСТЬ БИОГАЗОВОГО ДИГЕСТАТА В КАЧЕСТВЕ УДОБРЕНИЯ ДЛЯ МОЛОДЫХ РАСТЕНИЙ ИВЫ

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*Резюме*

Мы проанализировали возможности использования дигестата от анаэробного брожения жидкой свиной фекальной суспензии, как питательную добавку для культивирования ивовых плантаций с коротким циклом ротации. Как показали проведенные нами исследования в экспериментальной оранжерее, общая биомасса однолетнего растения семейства ивовых, обработанного умеренным количеством (далее – 0,5F) дигестата на протяжении вегетативного периода была более чем в два раза больше, чем в контроле (далее – С) и эта разница на протяжении времени только возрастает. После первого вегетативного периода производительность молодых деревьев и корневой биомассы растений 0,5F была значительно выше, чем у растений, удобряемых минеральным комплексом NPK при оптимальном количестве N (далее – MIN). Несмотря на то, что применение дигестата при оптимальном количестве N (далее – F) вредит растениям, способствуя разрушению корней в период первого вегетативного периода, тем не менее они смогли восстановиться и продолжить ежегодную продукцию, сопоставимую с растениями С на протяжении следующего года. Растениям MIN поставлялось такое же количество N, как и растениям F, отмиравшим на протяжении зимы, несмотря на длительное использование удобрения К. Большое количество дигестата (далее – 2F) вызывает серьезные проблемы и является летальным для однолетних растений. В конце эксперимента pH питательного субстрата, обработанного дигестатом и минеральными удобрениями был выше, чем в контрольных горшках. После внесения дигестата было доступным незначительное количество аммония N, но содержание К и Р в питательном субстрате увеличилось. Таким образом, полученные нами результаты показывают, что дигестат может быть использован в качестве альтернативного удобрения для растений с коротким циклом ротации и питательные вещества из дигестата более доступны растениям ивы, чем таковые минеральных удобрений.

**Ключевые слова:** биогазовый дигестат, биомасса, удобрение, *Salix*, рост побегов