



Research paper

Effects of digestate fertilization on *Sida hermaphrodita*: Boosting biomass yields on marginal soils by increasing soil fertility



Moritz Nabel, Silvia D. Schrey, Hendrik Poorter, Robert Koller, Nicolai D. Jablonowski*

Forschungszentrum Jülich GmbH, Institute of Bio- and Geosciences, IBG-2: Plant Sciences, 52428 Jülich, Germany

ARTICLE INFO

Keywords:

Perennial energy plants

Marginal lands

Soil fertility

Digestate fertilization

Nutrient cycling

Sida hermaphrodita (L.) Rusby

ABSTRACT

Perennial non-food energy crops are currently discussed as a more sustainable alternative to conventional energy crops like maize. As they can be cultivated on marginal soils, they reduce the risk of land use and food vs. fuel conflicts. In this study, we evaluated the perennial energy crop *Sida hermaphrodita* for its potential to be cultivated on marginal substrate and conventional agricultural soils over a three-year field and mesocosm experiment at agricultural conditions. Furthermore, we aimed for a closed nutrient loop by fertilizing plants with biogas digestate and using the carbon fraction of the digestate as soil amendment to ameliorate the overall soil fertility. As controls, plants were either untreated or fertilized with an equivalent amount of mineral NPK fertilizer. We found *S. hermaphrodita* to give highest DM yields of up to 28 t ha⁻¹ under favorable soil conditions when fertilized with mineral NPK. However, on marginal substrate digestate fertilization resulted in a clear biomass yield advantage over NPK fertilization. An increased soil carbon content, water holding capacity and basal soil respiration indicated improved soil fertility in the marginal substrate. These results demonstrate the great potential of *S. hermaphrodita* to be cultivated on marginal soil in combination with organic fertilization via biogas digestate.

1. Introduction

Energy crops have the potential to diversify our energy production [1]. However, cultivation of species like maize and oilseed rape on productive agricultural soils, causes land use conflicts and negatively impacts food security [2,3]. Perennial energy crops like *Populus* or *Miscanthus* are discussed as a more sustainable alternative [4,5]. More recently the perennial energy crop *Sida hermaphrodita* is coming to focus [6,7]. Compared to annual crops like maize and oilseed rape, it allows for an extensive production with minimal need for soil cultivation, weed and pest control [8,9]. Compared to other perennial energy crops, *S. hermaphrodita* has the potential to minimize land use conflicts, as it can be cultivated on light soils and marginal lands [3,6,10,11]. The European Environmental Agency (EEA) defines marginal land as being of low quality from an intensive agriculture viewpoint, where production barely covers cultivation costs [12]. *S. hermaphrodita* is a forb species from the North American prairies belonging to the Malvaceae family that develops a large root system allowing access to water and nutrients even when resources are limited. It grows well on sandy or rocky soils with low organic matter content and produces relatively high biomass yields even with low nutrient levels in the soil [9,13]. Assimilates stored in the large root system are instrumental for rapid

regrowth, rendering the plants competitive against weeds thus reducing the need for weed control in an established stand [8]. Its biomass can be used as a renewable resource, as solid fuel for direct combustion or as feedstock for biogas production [7,14]. The recorded biomass DM yields vary between 11 t ha⁻¹ on a light soil in eastern Poland to 25 t ha⁻¹ on a rich field soil in Germany [6,7]. In this study, we tested *S. hermaphrodita* for its potential cultivation on both a sandy marginal substrate and a pebbly field soil and compared the yield potential to a conventional rich field soil at agricultural conditions.

To allow for economic and sustainable use of soils for the cultivation of *S. hermaphrodita*, we aimed for an extensive cultivation system. As a key element, we fertilized plants with biogas digestate to facilitate a closed nutrient cycle and to render the cropping system independent from synthetically produced fertilizers [15,16]. An interesting asset of this closed-loop approach is that digestates from energy crops have a high concentration of organic matter derived carbon [17,18]. We investigate the potential of this carbon as a soil amendment to ameliorate the marginal substrate, which is naturally low in organic carbon and plant available nutrients. A soil amendment is any material, which, upon application to the soil, would improve or maintain its physical, chemical or biological properties [19]. Organic matter content is the main indicator that defines the status of a soil amendment [19].

* Corresponding author. Institute of Bio- and Geosciences, IBG-2: Plant Sciences, Forschungszentrum Jülich GmbH, 52425 Jülich, Germany.

E-mail address: n.d.jablonowski@fz-juelich.de (N.D. Jablonowski).

<http://dx.doi.org/10.1016/j.biombioe.2017.10.009>

Received 26 July 2017; Received in revised form 11 October 2017; Accepted 13 October 2017

Available online 21 October 2017

0961-9534/ © 2017 The Authors. Published by Elsevier Ltd. This is an open access article under the CC BY license (<http://creativecommons.org/licenses/by/4.0/>).

Accordingly, soil organic matter is a key element for increasing and sustaining soil fertility [20]. In combination with a no-tillage system digestate application could improve the soil properties in terms of water and nutrient retention, and thus increase the yield potential [21,22]. With very few exceptions, studies on the effects of digestate application on soil properties have consistently noted the improvement of soil quality [23].

Studies on *S. hermaphrodita* have been carried out either under greenhouse conditions [24–26], or under outdoor conditions [7,27]. So far, multi-year field research with *S. hermaphrodita* was mainly carried out in Poland, however, literature on this topic in the English language is limited to a few studies [6,8,28]. To our knowledge, a study focused on the potential of *S. hermaphrodita* for its cultivation on marginal soils and the potential increase of soil fertility via the reapplication of biogas digestates was not performed yet. To close this knowledge gap we designed a combined outdoor mesocosm and field experiment with three different substrates and three different fertilizer regimes, and evaluated plant biomass and soil data over three consecutive years. Additionally, two laboratory experiments were conducted to investigate the effects of digestate fertilization on the water holding capacity and basal soil respiration of a marginal substrate. The underlying research questions and hypothesis were the following:

Question 1. What is the yield potential of *S. hermaphrodita* on marginal soils and how does it scale to the yield on conventional field soils? To what extent is yield influenced by fertilization and substrate quality?

Hypothesis 1.1. Under optimal conditions, i.e. good soil quality and NPK fertilization, we expect biomass DM yield up to 20 t ha⁻¹, based on experiences from earlier field studies [7,8]. Yields from plants grown on substrate of lower quality are expected to be lower, which can partly be compensated by fertilization.

Hypothesis 1.2. Plants fertilized with mineral NPK fertilizer will perform better on rich field soil, whereas digestate-fertilized plants will perform better on marginal substrate due to the higher carbon share.

Question 2. In how far will digestate fertilization differ in its effect on properties of marginal sandy soil (e.g. soil carbon content, water-holding capacity) from NPK fertilization over numerous growing periods?

Hypothesis 2. Digestate fertilization will increase the soil carbon content and by that increase the water holding capacity, soil respiration and the overall yield potential of marginal substrates.

2. Materials and methods

2.1. Study sites and cultivation

The experimental field site was located at the Research Centre Jülich (50°53'47" north and 6°25'32" east; 80 m a.s.l.) and had a size of 1000 m². In May 2013 a stand of *S. hermaphrodita* (L.) Rusby was established by transplanting pre-cultivated seedlings of BBCH stage 14–16 from the greenhouse to the field in 0.7 m × 0.7 m planting distance [7]. The plants were left untreated for one year before the start of the experiment. Monthly average temperature, daily light integral (DLI) and precipitation were recorded (Fig. 1). The soil type of the field site is a luvisol with a clear gradient of pebble stone share (Table 1). Based on the stoniness and the higher share of the grain size fraction > 2 mm we defined an area of “rich field soil” and “poor field soil”. Sixty subplots of 2 m × 2 m with an additional 1 m border zone to the neighboring plots were implemented and fertilization treatments were applied in a fully randomized design.

Additionally, an outdoor mesocosm experiment in 21 containers, each filled with 250 L of a sandy substrate (0/1 fine aggregate sand, RBS GmbH, Inden, Germany; Ca: 0.3 g kg⁻¹; K: 0.2 g kg⁻¹; Mg:

0.8 g kg⁻¹; P: 0.1 g kg⁻¹; further information in Table 1), used as a marginal model substrate, was established nearby the field experiment (50°54'34"N 6°24'47"E). Mesocosms, filled with a sandy marginal model substrate were used, as a field area with a well-defined marginal substrate is not available in the area. The size of the containers was chosen to keep growing conditions of *S. hermaphrodita* as close to field conditions as possible, assuming a planting density of 20,000 ha⁻¹ [29,30]. Seedlings of *S. hermaphrodita* of BBCH stage 13–14 were transplanted into the mesocosms in May 2014 [7]. The detailed establishment of *S. hermaphrodita* plants into the mesocosms was described earlier [27].

2.2. Fertilization treatments

In May 2014, 2015 and 2016 the sixty subplots of the field and the 21 mesocosms received either digestate fertilization, mineral fertilization or no fertilizer supplement as a treatment. The digestate was obtained from a commercially operating biogas plant using maize silage as feedstock (digestate dry matter mass fraction: 7.2%; N_{total}: 0.53%; NH₄⁺: 0.32%; P: 0.14%; K: 0.68%; Mg 0.037%; Ca: 0.16%; S: 0.03%; organic matter: 5.3%; C:N ratio: 6; pH 8.2; all values referring to fresh weight; ADRW Naturpower GmbH & Co. KG, Titz-Ameln, Germany). NPK-fertilizer with a N:P:K-ratio similar to the digestate and a high share of ammonia was chosen to allow a comparison between the mineral and the organic digestate fertilization (NPK-fertilizer composition: N: 15% (1% nitrate; 9.5% ammonia; 4.5% isobutylidenediurea); P: 5%; K: 8%; Mg: 3%; Compo Rasendünger, Compo GmbH, Münster, Germany). Both fertilizers were calculated to provide a total nitrogen application of 160 kg ha⁻¹. We chose for this fertilization dose as it resulted in optimal plant growth in a previously published dose-response experiment for digestate fertilization of *S. hermaphrodita*, grown on the same marginal substrate used in this study [25,27].

2.3. Sampling and measurements

In October 2014, 2015 and 2016, the above ground biomass on both field sites and from the mesocosms was harvested and dry mass was determined after drying at 70 °C to constant weight. Soil samples were taken on each plot at 0–30 cm depth and 30–40 cm distance from the plants at the time of biomass harvest and were dried to constant weight at 30 °C for further analysis. N and C content of the soil and plant samples were determined by elemental analysis (VarioELcube, Elementar Analysensysteme GmbH, Langenselbold, Germany). Soil pH was determined using standard electrodes (Hanna Instruments pH 209 pH-meter, Vöhringen, Germany), using 0.01 M CaCl₂ solution at 20 °C as extractant.

2.4. Water holding capacity and soil respiration measurements

Complementary to the outdoor *S. hermaphrodita* cultivation experiments two laboratory experiments were conducted to investigate the effect of the biogas digestate on water holding capacity (WHC) and soil respiration of the marginal sandy substrate used in the mesocosm study. For WHC determination 300 g of dried sand were amended with varying digestate doses (30, 60, 90, 120, 150, 180 and 210 g kg⁻¹ substrate; n = 4) of biogas digestate, homogeneously mixed for 6 min in an end-over-end shaker and flooded with water until field capacity was reached [31]. The increasing doses were chosen to mimic multi-year application of digestate. After 24 h, when no more water was dripping out of the pots, the weight of the wet sample was determined [31]. Afterwards the wet samples were dried at 105 °C to constant weight to determine the soil dry weight. Water holding capacity (WHC) was calculated by using the following equation [31]:

$$\text{Water holding capacity} = \frac{\text{total water in the wet soil (g)}}{\text{oven dry weight of total soil (g)}} \times 100$$

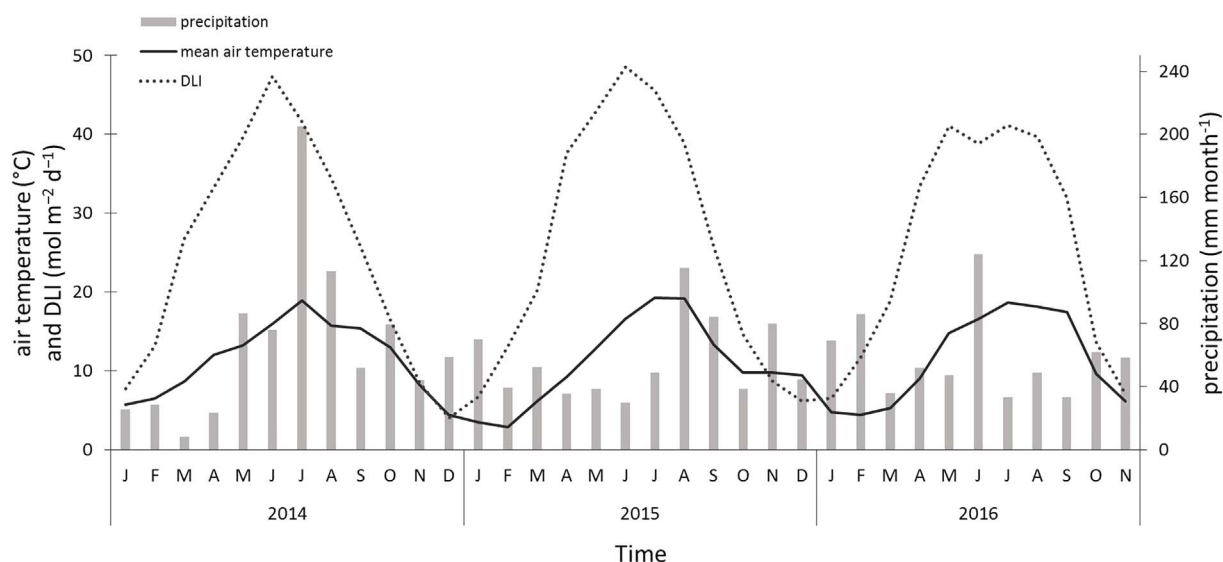


Fig. 1. Monthly mean temperature precipitation and daily light integral (DLI) values during the experimental time from 2014 to 2016 at the Research Center Jülich (50°53'47" north and 6°25'32" east; 80 m a.s.l.).

Basal soil respiration of the untreated marginal substrate and marginal substrate amended soil with two concentrations of biogas digestate (20 g kg⁻¹ and 40 g kg⁻¹) were measured in five biological replications. For sample preparation, 30 g of homogenized samples were adjusted to 40% WHC, filled into plastic vessels and slightly compressed [32,33]. Samples were incubated at 22 °C for 120 h to stabilize respiration rates [34]. Subsequently, basal soil respiration (CO₂) was measured with a Respicond VIII system (Nordgren Innovations AB, Sweden) at a constant temperature of 22 °C for 48 h [35,36].

2.5. Statistical analysis

The *S. hermaphrodita* cultivation experiment has a two-way factorial design with the factor fertilizer having three different levels in a completely randomized setup (control, NPK fertilization, digestate fertilization) and three different substrates (rich field soil, poor field soil and marginal sandy substrate) as second factor. The exact number of replicates per variant is given in Table 3. Statistical analysis was performed with analysis of variance (ANOVA) in R 3.0.3 (The R Foundation for Statistical Computing 2014) using the work package “Agricolae” with an *a posteriori* test as well as pairwise *t*-test [37].

3. Results and discussion

3.1. Biomass

Above-ground biomass continuously increased over the three-year duration of the experiment reaching a maximum of this three-year experiment in 2016 (Fig. 2). This corresponds well with findings of Borkowska et al. [6] who observed that above-ground biomass yields increased three to four years after planting and explained this effect with

Table 1

The three substrates differ mainly in their grain size fraction, while pH, total organic carbon (TOC) and total nitrogen (TN) content are in a comparable range. Data show the mean of n = 3 soil samples.

	Grain size fractions (%)					pH	TOC %	TN %
	0–0.002 mm	0.002–0.02 mm	0.02–0.2 mm	0.02–0.2 mm	> 2 mm			
Field (rich)	16	21	58	4	2	6.2	1.0	1.0
Field (poor)	11	15	47	4	22	6.2	0.7	0.8
Sand	1	1	40	58	0	6.3	0	0

Table 2

ANOVA results indicate that substrate quality had a greater effect on biomass yield of *Sida hermaphrodita* than fertilization. Digestate and NPK fertilization were adjusted to 160 kg N ha⁻¹. df: Degrees of Freedom; Sum Sq: Sum of Squares.

	df	Sum Sq	Effect (%)	p-value
Soil	2	31210066	58	< 0.01
Fertilization	2	9814541	18	< 0.01
unfertilized vs. fertilized	1			< 0.01
NPK vs. Digestate	1			0.26
Fertilization x Soil	4	3004071	6	< 0.01
unfertilized vs. fertilized	2			< 0.01
NPK vs. Digestate	2			0.018
Residuals	74	9466359	18	

the perennial growth of *S. hermaphrodita*, as plants mainly invest into the establishment of a deep reaching root system in the first years [8].

Fertilization generally had a positive effect on the biomass yield on all three substrates. Highest DM yields of up to 28 t ha⁻¹ were obtained on rich field soil in the third year of the experiment, when fertilized with mineral NPK (Fig. 2). Maximum yields were clearly higher than the expected yield presented in hypothesis 1.1. In mesocosms, filled with marginal sandy substrate a maximum DM yield of 9 t ha⁻¹ was measured for plants fertilized with digestate in their third year of growth and fertilization (Fig. 2). The relative biomass stimulation of fertilized plants compared to unfertilized control plants increased with decreasing substrate quality in the following order: rich field soil: +68%; poor field soil: +71%; marginal substrate: +597%. Despite the same nutrient application, the yield differences between the two field soils can be explained by the contrasting grain size fractions, i.e. the

Table 3

Soil carbon and nitrogen content was increased in digestate fertilized plots on all three substrate types. Control: no fertilization. Digestate and NPK fertilization were adjusted to 160 kg N ha⁻¹. \pm indicates the standard error. Variants with the same letter are not significantly different at $p < 0.05$ referring to data from 2016, within one treatment and substrate.

		Carbon mass fraction (%)			Nitrogen mass fraction (‰)			pH	n	
		2014	2016		2014	2016		2016		
rich field soil	Control	1.2 ± 0.1	1.3 ± 0.1	y	1.2 ± 0	1.4 ± 0.1	y	6.9 ± 0	x	26
	Digestate	1.1 ± 0.1	3.1 ± 0.6	x	1.2 ± 0	2.8 ± 0.5	x	7.0 ± 0	x	7
	NPK	1.2 ± 0.1	1.5 ± 0.1	y	1.2 ± 0	1.6 ± 0.1	y	6.3 ± 0.1	y	7
poor field soil	Control	1.2 ± 0.1	1.4 ± 0.1	n	1.2 ± 0.1	1.4 ± 0.1	n	6.9 ± 0	n	12
	Digestate	1.3 ± 0.2	2.8 ± 0.7	m	1.2 ± 0.1	2.6 ± 0.6	m	7.1 ± 0	m	6
	NPK	0.9 ± 0.1	1.3 ± 0.1	n	0.9 ± 0.1	1.4 ± 0	n	6.2 ± 0	o	6
marginal substrate	Control	0 ± 0	0.1 ± 0	b	0.1 ± 0	0.2 ± 0	b	7.4 ± 0.1	a	7
	Digestate	0.3 ± 0.1	0.5 ± 0.7	a	0.3 ± 0.1	0.5 ± 0.1	a	6.9 ± 0.1	b	7
	NPK	0.1 ± 0	0.2 ± 0	b	0.1 ± 0	0.3 ± 0	b	6.9 ± 0.1	b	7

stoniness (Table 1). The effect of grain size fraction on soil productivity is well known and corroborates our separation of the field soils into “rich” and “poor” soil [38].

Notwithstanding the fact that we applied the same amount of N, digestate and mineral NPK fertilization differed in their growth-stimulating effects (Fig. 2, Table 2). NPK fertilization had the strongest effect on plant growth on the rich and poor field soil. A possible reason for the lower performance of digestate fertilization could be a partial immobilization of NH₄⁺ and immobilization of N in the biological biomass pool [39,40]. Microorganisms take up nitrogen when they have access to a carbon source like digestate [41]. The high proportion of NH₄⁺ in the digestate might also have resulted in losses of N via volatile NH₃ [42,43].

In mesocosms, filled with marginal substrate, we observed an opposite effect, i.e. plants fertilized with digestate produced 31% higher biomass yield than those with NPK (Fig. 3). The first reason for this might be that the marginal substrate contained no traceable amounts of plant nutrients. By NPK fertilization only macro elements were applied while the digestate fertilization also contains micro nutrients [18]. However, no specific nutrient deficiency symptoms were observed

throughout all substrates and fertilization treatments over the course of three years.

Secondly, nitrate leaching out of the root horizon was found to be significant in the marginal substrate when treated with NPK-fertilizer, as described in our previous study [27]. Nitrate leaching from soil following digestate fertilization is low because nitrogen is either organically bound or in the mineral form NH₄⁺ which is not as mobile as nitrate [44]. During the first year of this experiment we followed nitrate concentration of the leachate and found a reduction of nitrate leaching to deeper horizons when comparing digestate to mineral NPK fertilization [27].

The third explanation for the better performance of digestate on the marginal substrate, we see in the amendment of the substrate with organic carbon. Digestate amendment increased the soil carbon content of the marginal substrate five times more than NPK fertilization [40]. The importance of the soil carbon content and its influence on soil aggregation, water holding capacity and soil fertility has been described earlier [21,22] and our results will be discussed in more detail in section 3.2. The fact that the relative advantage of digestate fertilization over mineral NPK fertilization on biomass yield gets more and more

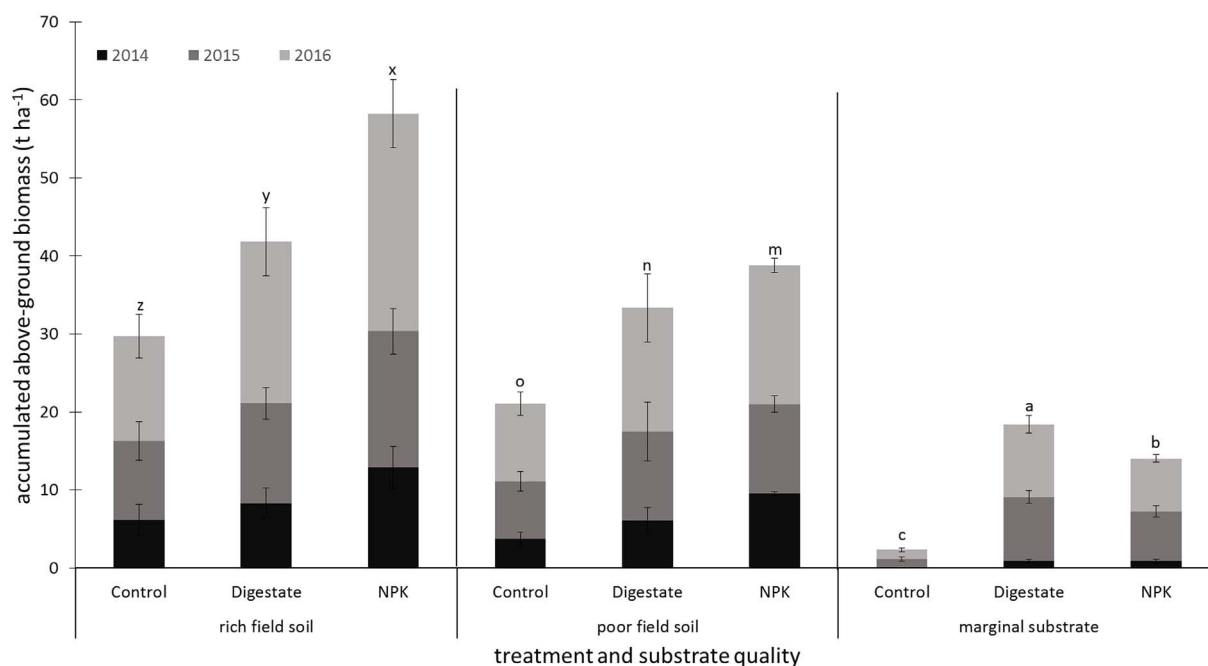


Fig. 2. The three-year cumulative biomass yield of *Sida hermaphrodita* was highly dependent on substrate type. Control: no fertilization. Digestate and NPK fertilization were adjusted to 160 kg N ha⁻¹. Planting density 20,000 plants ha⁻¹. Bars indicate the standard error (n = 6–26, indicated in Table 3). Within one substrate type, values of cumulative biomass with the same letter are not significantly different at $p < 0.05$.

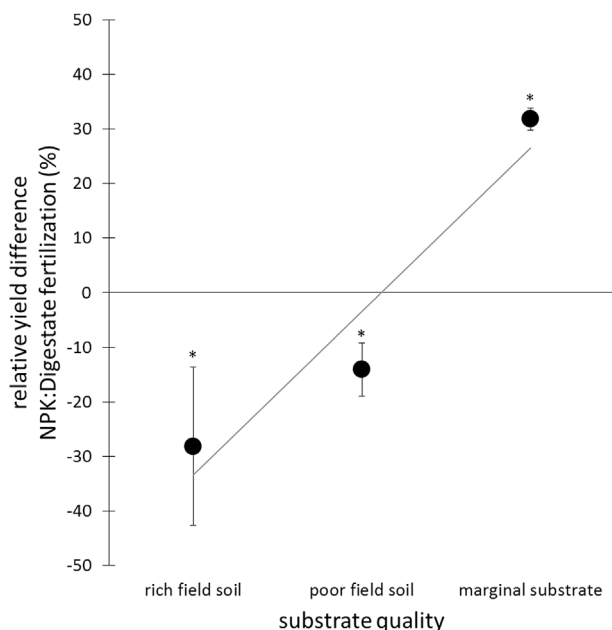


Fig. 3. The relative yield difference between digestate and NPK fertilization indicates the digestate yield advantage on marginal substrate. Digestate and NPK fertilization were adjusted to 160 kg N ha^{-1} . Bars indicate the standard error ($n = 6\text{--}26$, indicated in Table 3). Data points marked with * show a significant ($p < 0.05$) yield difference between digestate and NPK fertilization.

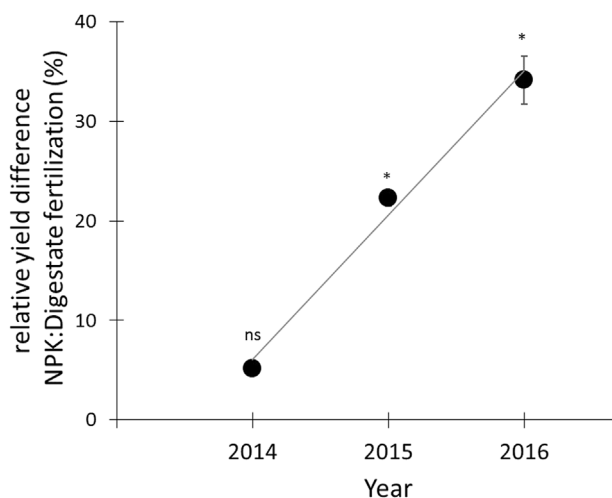


Fig. 4. Relative yield difference between digestate and NPK fertilization on marginal substrate. Yield difference of digestate vs. NPK fertilization is constantly increasing over time. Digestate and NPK fertilization were adjusted to 160 kg N ha^{-1} . Bars indicate the standard error ($n = 7$). Differences marked with * are significant at $p < 0.05$.

pronounced over time supports the linkage to the crucial role of the soil carbon content on plant performance (Fig. 4).

In conclusion, the DM yield expectations of 20 t ha^{-1} stated in hypothesis 1.1 were exceeded for the rich field soil. Nevertheless, the strong yield reduction in the poor soil and the marginal substrate could only be partially compensated by fertilization within this three-year study. Hypothesis 1.2, foretelling better performance of NPK fertilized plants on rich substrate and better biomass yields achieved via organic fertilization on the marginal substrate, was confirmed.

3.2. Soil

Throughout the entire experimental period of three years, the soil carbon content in the top 30 cm increased in mesocosm and field

substrates, however the increase was not found to be statistically significant within this three-year study. (Table 3). Similar results were described by Zan et al. [45]: the authors compared perennial cropping systems for bioenergy purposes with annual corn cropping systems and found that perennial systems without tillage are beneficial for carbon accumulation in the soil. A conversion from annual to perennial cropping systems favors soil carbon accumulation [46]. In our experiments, NPK fertilization did not result in different carbon accumulation rates compared to the unfertilized control, whereas digestate fertilization resulted in an enhanced soil carbon accumulation in mesocosm and field substrates (Table 3). We conclude that the additional carbon applied via the digestate partly remained and was incorporated into the soil carbon pool [18,40].

Soil nitrogen showed the same pattern and development as the soil carbon content (Table 3). At the end of each growth season, NPK fertilization resulted in the same soil nitrogen content as found in the unfertilized control plots. As fertilization took place at the beginning of the growth period, the added nitrogen was probably already taken up by the plants or leached into deeper soil layers before samples were taken. This might be particularly the case in the poor field soil and mesocosms filled with the marginal sandy substrate due to their higher porosity and low amounts of organic carbon [27,44]. However, digestate fertilization resulted in an increase of the soil nitrogen content in all three substrates. The NH_4^+ nitrogen present in the digestate gets partly immobilized by clay particles or bound to the organic fraction of the digestate [17,39]. A significant nitrogen immobilization in the case of anaerobic digestates from bark chips and organic kitchen wastes was reported already earlier [47]. Also humic compounds, that are part of the soil organic carbon are able to sequester nitrogen [48,49].

Mineral fertilization with the NH_4^+ -rich fertilizer resulted in lower soil pH values compared with unfertilized control plots on all three substrates. The soil acidification effect of NH_4^+ is well-known [50]. On marginal substrate digestate application resulted in a similar acidification. However, on the two field substrates digestate did not cause acidification like NPK. The high pH of the digestate itself as well as humic acids in the digestate can buffer the acidification effect of the NH_4^+ in the digestate [18,23]. Unfortunately, long-term studies about the impact of digestates in soil chemical and physical properties are limited. A three year study for different digestates, performed on a loamy Retisol showed no effect on pH after three years [51]. However, Giusquiani et al. [52] found comparable results on soil pH for composts with a similar pH, and Mäder et al. [53] even measured a slight increase of pH by the application of farm yard manure. Nevertheless, these studies were performed on different soil types and did not consider perennial cultures.

The results for soil carbon content, soil nitrogen content and pH indicate the potential of the combination of perennial cropping systems with organic fertilization for soil carbon accumulation and increased soil fertility as stated previously [21,22,47]. Hornick and Parr showed that the productivity of marginal soils with stony and sandy texture was strongly increased by its amelioration with composted manure and sewage sludge both having a positive effect on soil pH, the soil water content and nutrient status of the substrate [54,55]. However, to allow a deeper understanding of the processes that lead to soil carbon accumulation and increased soil fertility a much longer timespan and soil analysis also to a sampling depth of up to 90 cm would be necessary and should encourage further research. In addition, the question to what extent the fertility and productivity of a marginal substrate can be increased would be essential to allow assessments on economic feasibility of the broader cultivation of marginal soils.

3.3. Water holding capacity and basal soil respiration

The complex term of soil fertility cannot be expressed merely based on plant performance. In order to get a better understanding of the interaction between organic fertilization and the marginal sandy

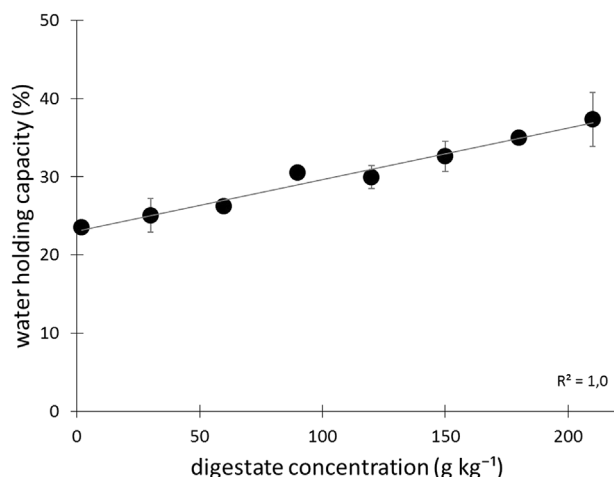


Fig. 5. The water holding capacity of the marginal substrate was positively influenced by the digestate concentration. Bars indicate the standard error ($n = 4$).

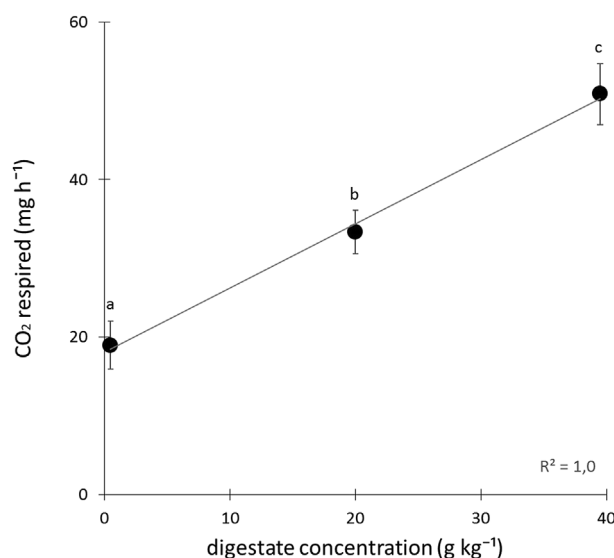


Fig. 6. Basal soil respiration of the marginal substrate increased by the addition of digestate. Bars indicate the standard error ($n = 5$). Values with the same letter are not significantly different at $p < 0.05$.

substrate, we set up two laboratory studies focusing on water holding capacity and basal soil respiration. Here, the amendment of the marginal substrate with increasing doses of digestate showed a positive correlation for basal soil respiration and water holding capacity (Figs. 5 and 6). Albuquerque et al. [56] found similar effects on soil respiration after the amendment with digestate and argued that digestates consist of two fractions of organic matter. The first fraction is easily degradable and triggers microbial activity, whereas the second fraction is more resistant to microbial degradation, contributing to the increase of soil organic matter [40]. Furthermore, Albuquerque et al. [56] were able to positively correlate the increased soil microbial activity and soil respiration with the formation of soil aggregates, resulting in a positive effect on the water holding capacity. In line with our results, Reeves [22] describes the importance of organic fertilization to maintain or increase soil organic matter. As the biological and physical soil properties like microbial activity and water holding capacity highly depend on carbon, organic fertilization is essential for a sustainable use of soils. Based on our results, we confirmed hypothesis 2, stating a generally positive influence of organic fertilization via digestate on the soil properties of the marginal substrate, as we can prove increased soil respiration and enhanced water holding capacity, both essential

indicators for increased soil fertility of the marginal sandy substrate.

4. Conclusion

Digestate fertilization resulted in higher plant biomass yields of *Sida hermaphrodita* on the marginal substrate. Furthermore, the relative yield advantage of digestate over NPK fertilization got more and more pronounced over the three-year experiment. Digestate fertilization increased the soil carbon content especially on the marginal substrate and thus had a beneficial effect on basal soil respiration and water holding capacity in this substrate.

Under favorable soil conditions and fertilization, i.e. “rich” field soil and NPK fertilization maximum biomass DM yields of 28 t ha^{-1} of *S. hermaphrodita* were reached. Not surprisingly, we found that with declining soil quality, the yield was reduced which could not be fully compensated by fertilization.

Even though NPK fertilization performed better on the rich soil compared to digestate fertilization, the organic fertilization is the favorable choice for the cultivation of the perennial energy crop *S. hermaphrodita* on marginal substrates. The combination of the perennial crop *S. hermaphrodita* and organic fertilization via digestate allows for an increase of the soil carbon content and an improvement of the soil fertility, resulting in an increased biomass yield over the first three years of this combined field and mesocosm experiment.

Acknowledgements

This study was financed by Forschungszentrum Jülich, IBG 2: Plant Sciences core funding. The digestate and the sand was kindly provided by ADRW Naturpower GmbH & Co.Kg, Ameln, and Rheinische Baustoffwerke, Inden, respectively. The kind provision of mesocosms by EGN mbH, Viersen, used as mesocosms for plant cultivation is highly appreciated. Many thanks to Lucy Harrison, Sabine Willbold and colleagues from ZEA-3 for the sampling and chemical analysis of the plant materials and soil samples. We thank Axel Knaps, Nele Meyer, Gerd Welp, Achim Kunz and Thorsten Kraska from the University of Bonn for their generous help with the soil respiration measurements and the provision of climate data. We thank Andre Schallenberg and the gardeners' team of FZ-Jülich for the help of setting up and maintaining the experimental sites. We thank the anonymous reviewers for their valuable comments and suggestions for the improvement of this manuscript. We highly acknowledge the financial support of numerous students' apprentices by the DAAD and IAESTE program, providing great support for this experiment.

References

- [1] W. Zegada-Lizarazu, A. Monti, Energy crops in rotation. A review, *Biomass Bioenergy* 35 (2011) 12–25, <http://dx.doi.org/10.1016/j.biombioe.2010.08.001>.
- [2] FAO, *The State of Food Insecurity in the World Economic Crises – Impacts and Lessons Learned 2009 Key Messages*, (2009).
- [3] D. Graham-Rowe, Agriculture: beyond food versus fuel, *Nature* 474 (2011) S6–S8, <http://dx.doi.org/10.1038/474S06a>.
- [4] S. Fang, J. Xue, L. Tang, Biomass production and carbon sequestration potential in poplar plantations with different management patterns, *J. Environ. Manage* 85 (2007) 672–679, <http://dx.doi.org/10.1016/j.jenvman.2006.09.014>.
- [5] I. Lewandowski, J.C. Clifton-Brown, J.M.O. Scurlock, W. Huisman, Miscanthus: European experience with a novel energy crop, *Biomass Bioenergy* 19 (2000) 209–227, [http://dx.doi.org/10.1016/S0961-9534\(00\)00032-5](http://dx.doi.org/10.1016/S0961-9534(00)00032-5).
- [6] H. Borkowska, R. Molas, A. Kupczyk, Virginia fanpetals (*Sida hermaphrodita* Rusby) cultivated on light soil; height of yield and biomass productivity, *Pol. J. Environ. Stud.* 18 (2009) 563–568.
- [7] N.D. Jablonowski, T. Kollmann, M. Nabel, T. Damm, H. Klose, M. Müller, et al., Valorization of *Sida (Sida hermaphrodita)* biomass for multiple energy purposes, *GCB Bioenergy* (2016) 1–13, <http://dx.doi.org/10.1111/gcbb.12346>.
- [8] H. Borkowska, R. Molas, Two extremely different crops, *Salix* and *Sida*, as sources of renewable bioenergy, *Biomass Bioenergy* 36 (2012) 234–240, <http://dx.doi.org/10.1016/j.biombioe.2011.10.025>.
- [9] H. Borkowska, K. Wardzinska, Some effects of *Sida hermaphrodita* R. cultivation on sewage sludge, *Pol. J. Environ. Stud.* 12 (2003) 119–122.
- [10] P. Schröder, R. Herzig, B. Bojinov, A. Ruttens, E. Nehnevajova, S. Stamatiadis, et al.,

- Bioenergy to save the world. Producing novel energy plants for growth on abandoned land, *Environ. Sci. Pollut. Res. Int.* 15 (2008) 196–204.
- [11] T.B. Voigt, O.K. Lee, G.J. Kling, Perennial herbaceous crops with potential for biofuel production in the temperate regions of the USA, *Cab. Rev.* 7 (2012) 45–57, <http://dx.doi.org/10.1079/PAVSN>.
 - [12] European Environmental Agency, Eionet, (2015) <http://www.eionet.europa.eu/gemet/concept/5023>, Accessed date: 25 July 2015.
 - [13] D.M. Spooner, A.W. Cusick, G.F. Hall, J.M. Baskin, Observations on the distribution and ecology of *Sida hermaphrodita* (L.) Rusby (Malvaceae), *Contrib. Bot.* 11 (1985) 215–225.
 - [14] M. Dębowski, M. Zieliński, M. Kisielska, M. Krzemieniewski, Anaerobic co-digestion of the energy crop *Sida hermaphrodita* and microalgae biomass for enhanced biogas production, *Int. J. Environ. Res.* (2017), <http://dx.doi.org/10.1007/s41742-017-0024-4>.
 - [15] J.J. Walsh, D.L. Jones, G. Edwards-Jones, A.P. Williams, Replacing inorganic fertilizer with anaerobic digestate may maintain agricultural productivity at less environmental cost, *J. Plant Nutr. Soil Sci.* 175 (2012) 840–845.
 - [16] T.K. Haraldsen, U. Andersen, T. Krogstad, R. Sørheim, Liquid digestate from anaerobic treatment of source-separated household waste as fertilizer to barley, *Waste Manag. Res.* 29 (2011) 1271–1276.
 - [17] J.A. Albuquerque, C. de la Fuente, A. Ferrer-Costa, L. Carrasco, J. Cegarra, M. Abad, et al., Assessment of the fertiliser potential of digestates from farm and agroindustrial residues, *Biomass Bioenergy* 40 (2012) 181–189.
 - [18] K. Möller, T. Müller, Effects of anaerobic digestion on digestate nutrient availability and crop growth: a review, *Eng. Life Sci.* 12 (2012) 242–257.
 - [19] European Committee for Standardization, Brussels, Belgium: CEN CR 13456 Soil Improvers and Growing Media — Labelling, Specifications and Product Schedules, (1999).
 - [20] H. Tiessen, E. Cuevas, P. Chacon, The role of soil organic matter in sustaining soil fertility, *Nature* 371 (1994) 783–785, <http://dx.doi.org/10.1038/371783a0>.
 - [21] M.H. Beare, P.F. Hendrix, D.C. Coleman, Water-stable aggregates and organic matter fractions in conventional- and no-tillage soils, *Soil Sci. Soc. Am. J.* 58 (1994) 777–786.
 - [22] D.W. Reeves, The role of soil organic matter in maintaining soil quality in continuous cropping systems, *Soil Tillage Res.* 43 (1997) 131–167.
 - [23] R. Nkoa, Agricultural benefits and environmental risks of soil fertilization with anaerobic digestates: a review, *Agron. Sustain. Dev.* 34 (2014) 473–492, <http://dx.doi.org/10.1007/s13593-013-0196-z>.
 - [24] D.B.P. Barbosa, M. Nabel, N.D. Jablonowski, Biogas-digestate as nutrient source for biomass production of *Sida hermaphrodita*, *Zea mays* L. and *Medicago sativa* L., *Energy Procedia*, 2014, pp. 120–126, <http://dx.doi.org/10.1016/j.egypro.2014.10.357>.
 - [25] M. Nabel, D. Bueno, P. Barbosa, D. Horsch, N.D. Jablonowski, Energy crop (*Sida hermaphrodita*) fertilization using digestate under marginal soil conditions: a dose-response experiment, *Energy Procedia* 59 (2014) 127–133, <http://dx.doi.org/10.1016/j.egypro.2014.10.358>.
 - [26] J. Franzaring, I. Holz, Z. Kauf, A. Fangmeier, Responses of the novel bioenergy plant species *Sida hermaphrodita* (L.) Rusby and *Silphium perfoliatum* L. to CO₂ fertilization at different temperatures and water supply, *Biomass Bioenergy* 81 (2015) 574–583, <http://dx.doi.org/10.1016/j.biombio.2015.07.031>.
 - [27] M. Nabel, V.M. Temperton, H. Poorter, A. Lücke, N.D. Jablonowski, Energizing marginal soils - the establishment of the energy crop *Sida hermaphrodita* as dependent on digestate fertilization, NPK, and legume intercropping, *Biomass Bioenergy* 87 (2016) 9–16, <http://dx.doi.org/10.1016/j.biombio.2016.02.010>.
 - [28] E. Krzywy-Gawronska, The effect of industrial wastes and municipal sewage sludge compost on the quality of Virginia fanpetals (*Sida hermaphrodita* Rusby) biomass Part 1. Macroelements content and their uptake dynamics, *Pol. J. Chem. Technol.* 14 (2012) 9–15, <http://dx.doi.org/10.2478/v10026-012-0064-7>.
 - [29] H. Poorter, F. Fiorani, R. Pieruschka, T. Wojciechowski, W.H. van der Putten, M. Kleyer, et al., Pampered inside, pestered outside? Differences and similarities between plants growing in controlled conditions and in the field, *New Phytol.* 212 (2016) 838–855, <http://dx.doi.org/10.1111/nph.14243>.
 - [30] H. Poorter, J. Bühler, D. Van Dusschoten, J. Climent, J.A. Postma, Pot size matters: a meta-analysis of the effects of rooting volume on plant growth, *Funct. Plant Biol.* 39 (2012) 839–850, <http://dx.doi.org/10.1071/FP12049>.
 - [31] R. Viji, P.P. Rajesh, Assessment of water holding capacity of major soil series of Lalgudi, Trichy, India, *J. Environ. Res. Dev.* 7 (2012) 393–398.
 - [32] N. Meyer, G. Welp, L. Bornemann, W. Amelung, Microbial nitrogen mining affects spatio-temporal patterns of substrate-induced respiration during seven years of bare fallow, *Soil Biol. Biochem.* 104 (2017) 175–184, <http://dx.doi.org/10.1016/j.soilbio.2016.10.019>.
 - [33] ISO-Secretariat, Geneva, Switzerland: ISO/DIS 17155: Soil Quality - Determination of Abundance and Activity of the Soil Microflora Using Respiration Curves, (2001).
 - [34] S.A. Blagodatsky, O. Heinemeyer, J. Richter, Estimating the active and total soil microbial biomass by kinetic respiration analysis, *Biol. Fertil. Soils* 32 (2000) 73–81, <http://dx.doi.org/10.1007/s003740000219>.
 - [35] A. Nordgren, Apparatus for the continuous, long-term monitoring of soil respiration rate in large numbers of samples, *Soil Biol. Biochem.* 20 (1988) 955–957, [http://dx.doi.org/10.1016/0038-0717\(88\)90110-1](http://dx.doi.org/10.1016/0038-0717(88)90110-1).
 - [36] H. Schiedung, S. Bauke, L. Bornemann, G. Welp, N. Borchard, W. Amelung, A simple method for in-situ assessment of soil respiration using alkali absorption, *Appl. Soil Ecol.* 106 (2016) 33–36, <http://dx.doi.org/10.1016/j.apsoil.2016.05.002>.
 - [37] R package version 1.2-0. Felipe de Mendiburu, agricolae: Statistical procedures for agricultural research, (2014) <http://cran.r-project.org/package=agricolae>.
 - [38] M. Lothar, S. Uwe, M. Wilfried, T.S. Graham, C.B. Bruce, H. Katharina, et al., Review article Assessing the productivity function of soils, *A Rev. Inf. Prod.* 30 (2010) 601–614, <http://dx.doi.org/10.1051/agro/2009057>.
 - [39] H. Kirchmann, A. Lundvall, Relationship between N immobilization and volatile fatty acids in soil after application of pig and cattle slurry, *Biol. Fertil. Soils* 15 (1993) 161–164, <http://dx.doi.org/10.1007/BF00361605>.
 - [40] J.A. Albuquerque, C. de la Fuente, M.P. Bernal, Chemical properties of anaerobic digestates affecting C and N dynamics in amended soils, *Agric. Ecosyst. Environ.* 160 (2012) 15–22.
 - [41] A. Hodge, D. Robinson, A. Fitter, Are microorganisms more effective than plants at competing for nitrogen? *Trends Plant Sci.* 5 (2000) 304–308, [http://dx.doi.org/10.1016/S1360-1385\(00\)01656-3](http://dx.doi.org/10.1016/S1360-1385(00)01656-3).
 - [42] M.H. Chantigny, D.A. Angers, P. Rochette, G. Bélanger, D.M. Agriculture, A. Canada, Gaseous Nitrogen Emissions and Forage Nitrogen Uptake on Soils Fertilized with Raw and Treated Swine Manure, (2006), pp. 1864–1872, <http://dx.doi.org/10.2134/jeq2007.0083>.
 - [43] R. Gutser, T. Ebertseder, A. Weber, M. Schraml, U. Schmidhalter, Short-term and residual availability of nitrogen after long-term application of organic fertilizers on arable land, *J. Plant Nutr. Soil Sci.* 168 (2005) 439–446, <http://dx.doi.org/10.1002/jpln.200520510>.
 - [44] H. Di, K. Cameron, Nitrate leaching in temperate agroecosystems: sources, factors and mitigating strategies, *Nutr. Cycl. Agroecosyst.* (2002) 237–256.
 - [45] C.S. Zan, J.W. Fyles, P. Girouard, R.A. Samson, Carbon sequestration in perennial bioenergy, annual corn and uncultivated systems in southern Quebec, *Agric. Ecosyst. Environ.* 86 (2001) 135–144, [http://dx.doi.org/10.1016/S0167-8809\(00\)00273-5](http://dx.doi.org/10.1016/S0167-8809(00)00273-5).
 - [46] W.M. Post, K. Wron, Soil carbon sequestration and land-use change: processes and potential, *Glob. Chang. Biol.* 6 (2000) 317–327, <http://dx.doi.org/10.1046/j.1365-2486.2000.00308.x>.
 - [47] T. Larsen, J. Luxhøi, J. Magid, L.S. Jensen, P.H. Krogh, Properties of anaerobically digested and composted municipal solid waste assessed by linking soil mesofauna dynamics and nitrogen modelling, *Biol. Fertil. Soils* 44 (2007) 59–68, <http://dx.doi.org/10.1007/s00374-007-0178-x>.
 - [48] D.W. Johnson, Nitrogen retention in forest soils, *J. Env. Qual.* 21 (1992) 1–12.
 - [49] F.J. Stevenson, Humus chemistry: genesis, composition, reactions, *Nature* 303 (1983) 835–836, [http://dx.doi.org/10.1016/0146-6380\(83\)90043-8](http://dx.doi.org/10.1016/0146-6380(83)90043-8).
 - [50] P. Barak, B.O. Jobe, A.R. Krueger, L.A. Peterson, D.A. Laird, Effects of long-term soil acidification due to nitrogen fertilizer inputs in Wisconsin, *Plant Soil* 197 (1997) 61–69, <http://dx.doi.org/10.1023/A:1004297607070>.
 - [51] T. Vanden Nest, G. Ruysschaert, B. Vandecasteele, M. Coughon, R. Merckx, D. Reheul, P availability and P leaching after reducing the mineral P fertilization and the use of digestate products as new organic fertilizers in a 4-year field trial with high P status, *Agric. Ecosyst. Environ.* 202 (2015) 56–67, <http://dx.doi.org/10.1016/j.agee.2014.12.012>.
 - [52] P.L. Giusquiani, M. Pagliai, G. Gigliotti, D. Businelli, A. Benetti, Urban waste compost: effects on physical, chemical, and biochemical soil properties, *J. Environ. Qual.* 24 (1995) 175–182, <http://dx.doi.org/10.2134/jeq1995.00472425002400010024x>.
 - [53] P. Mäder, A. Fließbach, D. Dubois, L. Gunst, P. Fried, U. Niggli, et al., Soil fertility and biodiversity in organic farming, *Atlantic* 296 (2008) 1694–1697, <http://dx.doi.org/10.1126/science.1071148>.
 - [54] S.B. Hornick, J.F. Parr, Restoring the productivity of marginal soils with organic amendments, *Am. J. Altern. Agric.* 2 (1987) 64–68, <http://dx.doi.org/10.1017/S0889189300001594>.
 - [55] S.B. Hornick, Use of organic amendments to increase the productivity of sand and gravel spoils: effect on yield and composition of sweet corn, *Am. J. Altern. Agric.* 3 (1988) 156–162, <http://dx.doi.org/10.1017/S0889189300002435>.
 - [56] J. a. Albuquerque, C. de la Fuente, M. Campoy, L. Carrasco, I. Nájera, C. Baixauli, et al., Agricultural use of digestate for horticultural crop production and improvement of soil properties, *Eur. J. Agron.* 43 (2012) 119–128.