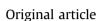
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Effect of digestate and fly ash applications on soil functional properties and microbial communities





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ABSTRACT

Digestate from biogas production contains a significant amount in organic matter and N, whereas fly ash from biomass combustion is rich in P and other nutrients. The use of both, as suitable fertilizers, has been shown to improve crop yields and soil properties. A Chernozem soil was used in a pot experiment in which wheat was grown to evaluate the impact of digestate (organic amendment) and fly ash (inorganic amendment) application on soil microbial activity, biomass and community structure. Soil samples were collected after two time-term treatments (30 and 60 days). Digestate application significantly improved soil chemical variables, microbial activity, the substrate average well colour development (SAWCD) and physiological diversity. Bacterial, actinobacterial, and fungal communities were positively affected by digestate addition, as well as microbial biomass. These effects were evident after a short period (30 days), but were more significant after long-term treatment (60 days). However, fly ash did not provoke significant changes in soil microbial activities, qPCR or PLFA at any time of treatment. Nevertheless, with this amendment, some chemical variables were positively affected after 60 days of treatment and the SAWCD revealed different patterns in the use of C sources compared with un-amended and digestateamended soil at both treatment times. These results suggest that the chemical properties present in digestate may determine its use as a suitable organic amendment because it improves soil health and quality, whereas fly ash did not have a relevant impact on soil microbial activities, gPCR, or PLFA.

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1. Introduction

The intensive agricultural and industrial practices generate a large production of different organic wastes which have has detrimental consequence into the environment if they are not appropriately managed. The use of organic wastes for biogas production (CO_2 and CH_4) by an anaerobic digestion leads the re-use and/or valorization of them. In Czech Republic, the phenomenon of biogas plants has become a great importance during the last decade since at least 13% of energy produced is derived from such sources [1]. Therefore, the production of energy from biogas plants

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will take a major role in the substitution of fossil fuels with renewable resources [2]. The rapid development of an anaerobic digestion will result in the increased production of biogas and a residual material known as digestate. The adequate management or disposal of this residue must be immediately addressed to avoid a limitation in the development of anaerobic digestion systems. The use of digestate with agronomical purposes seems to be a suitable strategy for its recycling [3]. Digestate also presents some advantages compared with other untreated residues due to its microbial stabilization [4]. A study conducted by García-Sánchez et al. [5] has observed that digestate soil application improved the input of inorganic nutrients in plant-available forms due to mineralization of organic compounds during the anaerobic digestion. Moreover, soil spreading of digestate results in higher amounts of organic-C compounds and N (present as ammonium), which take an

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important role enhancing the soil organic matter (SOM) content and thus, improving the biological activity and crop yield [3,6]. The maintenance of soil functionality and diversity is essential for sustainable agricultural production. It is generally accepted that the application of organic amendments stimulates soil microbial biomass, basal respiration, and enzyme activities [7–10]. In fact, studies performed on digestate application to soil have observed as this practice provided a source of available nutrients and had a positive impact on soil biology properties such as microbial biomass and activity as previously was reported by Alburguergue et al. [6]. Others studies have investigated the influence of anaerobic digestion on the fertilizer value of digestate observing a positive impact on soil microbial enzymes and nutrients uptake by plants [11]. A considerable increase was detected in the bacterial growth respect to fungal communities in soil previously amended by anaerobic digestate [12].

In the context of renewable energy, the production of other byproduct from biomass combustion in thermal power plants, such as fly ash, take also special attention [13]. The world annual production of fly ash is estimated to be around 600 million tons. This residue is one of the most complex and abundant anthropogenic materials and has been considered as a problematic solid waste because it is difficult to safely dispose [14]. Fly ash shows a wide variation in physico-chemical and mineralogical properties depending on the nature of the parent coal, the conditions of combustion, type of emission control devices, as well as storage and handling methods [15]. The principal components in fly ash are silica, aluminum and iron oxide along with significant percentages of Ca. K and Na. Ca was found to be the dominant cation in fly ash followed by Mg, Na, and K [16]. Al in fly ash is mostly bound in insoluble aluminosilicate structures, which considerably limits its biological toxicity. Also, quantities of trace metals (Cu, Zn, Mn, and Mo) and toxic elements (V, Se, As, B, Cd, Pb, Hg, and Cr) have been detected in the composition of fly ash [17]. The fly ash application, as inorganic amendment, have significantly increased the yield in different types of crops as recently have been reviewed by Ram and Masto [18]. In fact, there are some reports that mention its potential use as amendment to increase soil pH, availability of nutrients and stimulate the microbial activities in the soil [19,20]. However, the use of fly ash in agriculture may be limited due to its low content in N, C, and high pH, since these factors have been suggested to reduce the microbial activity [14]. Nevertheless, some studies have shown a significant increase in the ratio of CO₂ evolution, bacterial and actinobacterial communities, and soil dehydrogenase, urease, cellulose, protease, and phosphatase activities in soil amended by fly ash as previously was reported by Ram and Masto and Pandey and Singh [16,18].

Soil microbial communities are involved in critical ecosystem functions such as transformation of SOM and geochemical cycling, and strongly influence the soil physical characteristics as well as plant health and nutrition [21–23]. Nutrient cycling and turnover processes of organic matter are largely determined by the composition and activity of soil microbial communities [24]. Therefore, soil microbial biomass, activity, and community structure are important aspect of soil quality and thus, changes in soil microbial community composition can be the earliest indicators of soil health in many ecosystems processes [25]. Soil management practices such as application of organic and inorganic amendments may influence the microbial biomass, activity, and structure of soil communities [26]. The present work evaluates the impact of digestate and fly ash application on: i) soil chemical properties; ii) changes on extracellular enzyme activities and different use of C source; and ii) explores alterations in soil microbial biomass and community structure in a soil treated by digestate and fly ash. These effects were evaluated in a greenhouse experiment under controlled conditions. Based on these considerations the objective of this study was to compare the effectiveness of soil amendments, digestate and fly ash, stimulating these parameters in order to identify which had the most relevant impact improving soil characteristic.

2. Material and methods

2.1. Materials

The soil used in this study was collected from an experimental field located in Suchdol (Prague, Czech Republic, $50^{\circ}7'N$, $14^{\circ}22'E$). The soil texture and type were classified as loam (2.18% clay, 71.80% silt, and 26.03% sand) and Chernozem, respectively, according to FAO [26]. The chemical characteristics found in this soil were previously described by García-Sánchez et al. [27]. The nutrient contents of P, Mg, K, and Ca were determined according to the method described by Mehlich [28], and the values were as follows: 91 mg kg⁻¹, 240 mg kg⁻¹, 230 mg kg⁻¹, and 9000 mg kg⁻¹, respectively. Samples were randomly collected from the Ap horizon (20 cm depth) and subsequently were sieved (5 mm mesh) and mixed. The soil was stored for 3 days in thin mesh plastic bags at room temperature until the experiment was initiated.

The samples of digestate used in this experiment originated from a biogas station (1732 kW/h) (Červený Újez, Prague), where the digested material consisted of sugar beet pulp (50%), fruit marc (42%), and maize silage (8%). Digestate was previously treated by air-drying at 60 °C for 72 h in order to increase its agronomical properties before its use, as previously shown by Abdullahi et al. [29]. Meanwhile, the fly ash to be used as an amendment was derived from the combustion of wood in two reactors (1.8 MW and 0.6 MW). The chemical composition of digestate and fly ash was shown in Table 1.

2.2. Soil amendment

The experiment was set up using pots containing 300 g of soil. Digestate and fly ash were added manually and mixed thoroughly with the soil. The ratio of digestate application was of 10 g (dry weight basis) per 100 g soil (equivalent to a field application of 100 Mg ha⁻¹). This dose of application was selected to avoid low inputs of organic-C to soil and for keeping the N addition in optimal rate. Meanwhile, fly ash was added in a dose of 1.5 g (dry weight basis) per 100 g soil (equivalent to a field application of 15 Mg ha⁻¹) in order to maintain the pH soil requirements. Non-amended soil was also prepared and used as a control. One 15-day-old wheat

Table 1				
Chemical	characterization	of digestate	and fl	y ash.

	Digestate	Fly ash
рН	10	11.8
EC (ms/cm)	13.8	11
C (%)	26.9	n.d
N (%)	3.3	n.d
C/N ratio	8.53	n.d
P (%)	1.2 ± 0.01	1.3 ± 0.01
K (%)	2.1 ± 0.01	7.7 ± 0.02
Mg (%)	0.5 ± 0.02	1.4 ± 0.02
Ca (%)	3.1 ± 0.01	13.4 ± 0.1
S (%)	0.6 ± 0.01	4.1 ± 0.01
Cu (ppm)	0.004 ± 0.001	0.02 ± 0.001
Fe (ppm)	0.2 ± 0.01	2.8 ± 0.01
Mn (ppm)	0.02 ± 0.00	1.3 ± 0.01
Zn (ppm)	0.03 ± 0.00	3.6 ± 0.08
Σ16 PAHs (μ g kg ⁻¹)	n.d.	2.1
Σ2-4 rings compounds (µg kg ⁻¹)	n.d.	2.1

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