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Benefits and drawbacks of combined plant and mushroom production in substrate based on biogas digestate and peat

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ABSTRACT

Production of plants and mushrooms in substrate based partly on anaerobic digestate from biogas production (30%) and peat (70%) was studied in experiments performed using oyster mushroom (*Pleurotus ostreatus*) and basil (*Ocimum basilicum*). Biogas digestate was included in order to decrease use of peat and fertilizer. In separate experiments, combined greenhouse production of mushrooms and plants in fresh substrate, mushroom production in bags of fresh substrate or spent substrate from plant production, and plant production in spent substrate from mushroom production were studied. In terms of plant yield, positive impacts of combined culture were observed, with significantly higher yield of basil when mushroom spawn was added to fresh substrate at a concentration of 2% ($p = 0.04$). Increasing the concentration to 10%, which was sufficient for fruiting body formation in parallel with plant production, did not increase basil yield compared with the control. When fresh substrate was partly replaced with spent substrate from mushroom production, significantly higher yield of basil was obtained ($p = 0.001$). Mushroom production had an impact on the nutritional composition of the substrate, resulting in changes in nitrogen dynamics, a significant decrease in phosphorus concentration by 14% ($p = 0.001$), and a change in extractable concentrations of five of 10 elements studied. In terms of mushroom yield, the impacts of combined production with plants were generally negative.

1. Introduction

In order to secure future sustainability and food security, there is interest in food production in densely populated areas. In protected horticultural production systems, high crop yields can be achieved in urban settings with low availability of arable soil (Porter, 2015; Raviv, 2017). Such production systems, often in a multilayered tower design, are now increasingly being constructed in unused urban areas such as abandoned underground stations and basements, or in small movable containers.

Production of vegetables and herbs in protected horticultural systems is dependent on consistent and assured quality of the substrate in which the plants are grown. For decades, peat has been the main potting substrate (Schmilewski, 2008). Although, peat bogs are valuable ecosystems for biological diversity, they are a non-renewable resource, resulting in demands for reduced use of peat-based substrate (Fascella, 2015). In parallel, conventional systems for degradation of organic waste through anaerobic biodigestion have developed into large-scale net energy (biogas) production technology.

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This technology is now widely used in many countries for treatment of different types of agro-industrial wastes and, besides the biogas, it generates nutrient-rich, partly degraded digestate as a by-product (de Groot and Bogdanski, 2013). Recent technological development allows energy-efficient separation of this digestate into liquid and solid fractions. The liquid fraction has potential for use in hydroponic vegetable production, e.g., in protected horticultural production systems (Lind et al., 2020; Bergstrand et al., 2020). The solid fraction, which contains a high concentration of undegraded lignocellulosic plant material and high amounts of inorganic nitrogen, potassium, and phosphorus (Nkoa, 2014), has potential for use as a component in plant potting substrates, in order to reduce peat use. This would also enable organic fertilization of the plant, based on recycled plant nutrients.

Besides decreased consumption of peat and increased use of recycled plant nutrients, future plant production systems should also include different crops, to allow diversity and decrease the risk of plant diseases. An interesting and less explored option for increased diversity is combined plant and mushroom production. Further, research on future diets suggests decreasing consumption of meat and increasing consumption of vegetables (Karlsson et al., 2017). In this context, mushrooms can be important as a future protein source, with protein levels of 20%–25% of dry weight (dwt) (Kalac, 2013). Thus, innovative systems for mushroom production are of interest.

Plants and mushroom share a need for some specific physical characteristics of the substrate. For example, the substrate needs to have a high water-holding capacity while still allowing gas exchange to avoid anaerobic conditions (Sánchez, 2010; Schmilewski, 2008). However, plants and mushrooms interact with the substrate and affect it in different ways. Fungal growth is heterotrophic, resulting in partial degradation of the substrate and release of carbon dioxide, while plant growth is autotrophic and substrate is needed to physically support the roots and to supply water and essential nutrients, such as nitrogen, phosphorus, potassium. Thus, from the perspective of plant production, inclusion of a mushroom component may add benefits such as increased substrate mineralization, and thereby increased availability of nutrients, together with release of carbon dioxide, which is beneficial for plant growth. Considering the challenge in meeting plant nutrient requirements when using only organic nutrient sources in protected horticultural systems (Bergstrand et al., 2019), it is of interest to explore these aspects further.

In the present study, different scenarios for integrated production of basil (*Ocimum basilicum*) and oyster mushroom (*Pleurotus ostreatus*) using a substrate based on peat and the solid fraction of biogas digestate were investigated under greenhouse conditions. These crops differ evidently; being autotrophic and heterotrophic, respectively. However, both basil and oyster mushroom are high-value crops with a fast production cycle and the aim was to explore potential synergism in the production. The scenarios were: combined production of mushrooms and plants in fresh substrate, mushroom production in fresh substrate and in spent substrate from plant production, and plant production in spent substrate from mushroom production.

2. Materials and methods

2.1. Substrate

A substrate composed of 70% (v/v) unfertilized peat (Naturtorv Solmull, Hasselfors garden AB, Örebro, Sweden) and the solid digestate from biogas production (30% v/v), limed with 3 g of lime per L, was used in all experiments. This composition (70% peat and 30% solid digestate) was chosen as initial trials reveled slightly hampered growth of both basil and fungal mycelium when digestate was added in concentration exceeding 30% (data not shown). The digestate was obtained from a commercial biogas reactor (Gasum AB, Örebro, Sweden), fed mainly with crop residues amended with pig manure. The final substrate had bulk density 355 $\pm$ 5 g/L and porosity 68%, and the pH was 6.0–6.5 after liming.

2.2. Fungal strain and plant material

Grain spawn of the fungal strain *Pleurotus ostreatus* M2191 (oyster mushroom) was obtained from Mycelia BVBA, Deinze, Belgium. Seeds of basil (*Ocimum basilicum* L. cv. Sweet) were obtained from Wexthuset AB, Enhörna, Sweden.

2.3. Conditions for plant cultivation

Seeds of basil were sown directly in pots and thinned after emergence, as described in detail in Section 2.5. The pots were placed in a greenhouse compartment and artificial lighting (400 W high-pressure sodium lamps; Philips, Eindhoven, the Netherlands) at an intensity of 129 $\pm$ 10 μmol m^{-2} s^{-1} was supplied for 16 h day^{-1} after seed germination (day 6). The temperature set-point in the chamber was 22 °C and the ventilation set-point was 24 °C. The plants were irrigated with deionized water and no fertilizer was added during the growing period. All pots were given the same amount of water when the experiment started. Irrigation supplied during the experiment was based on the water-holding capacity of the substrate. No water was lost by drainage during or after irrigation. External radiation was logged using a greenhouse climate computer (Priva, de Lier, The Netherlands)

Table 1
Treatments in experiments 1–3. All experiments were performed in substrate based on 70% peat-30% solid biodigestate. Experiments 1 and 3 were performed in a greenhouse, experiment 2 in a climate chamber for mushroom production, using fresh substrate or spent substrate from plant production (SPS). In experiment 3 spent substrate from mushroom production (SMS) was included. Mushroom yield (biological efficiency, BE) when mushroom spawn was added is shown. Plant yield (fresh weight) is presented for treatments including plants.

Experiment	Conditions	Mushroom spawn (% dwt)	Basil	BE (%)	Plant yield (g/pot)
1	Greenhouse, fresh substrate	–	+	not applicable (na)	11.0 ± 1.9 (control)
		2	–	0	na
		2	+	0	16.1 ± 2.6*
		10	–	18.1 ± 6.5	na
		10	+	15.6 ± 3.8	6.4 ± 3.3
2	Climate chamber, fresh substrate	10	–	50.9 ± 6.1	na
	Climate chamber, SPS	10	–	28.7 ± 2.5	na
3	Greenhouse, 100% fresh substrate	–	+	na	27.1 ± 0.7 (control)
	Greenhouse, 25% SMS	–	+	na	29.5 ± 0.6*
	Greenhouse, 50% SMS	–	+	na	31.6 ± 0.5*
	Greenhouse, 75% SMS	–	+	na	30.2 ± 0.7*

*Significant difference ($p \leq 0.05$) compared with the control.

2.4. Conditions for mushroom production

Before inoculation with *P. ostreatus* spawn, the peat-digestate substrate was pasteurized at 65 °C for 8 h. When the substrate had cooled down, the spawn was mixed in as described in detail in Section 2.5. For induction of fructification, the colonized substrate was incubated at 10 °C for 3 days, and then at 22–24 °C with humidity 85% until harvest of the fruiting bodies.

2.5. Experimental set-up

2.5.1. Experiment 1: Combined basil and oyster mushroom production under greenhouse conditions

The different treatments included in the experiments are presented in Table 1. In experiment 1 a volume of 1.7 L (corresponding to 600 g wet weight) of substrate, inoculated with mushroom spawn in a concentration of 2% or 10% (dwt/dwt), was added to each pot. The pots were then topped up with 0.4 L (125 g wet weight) of substrate without any mushroom spawn, making an approximately 1.5 cm layer. The control treatment was prepared similarly but spawn was not included in the substrate. Into the top layer of substrate, 30 seeds of basil were sown initially and thinned to 25 seedlings per pot on day 6. The pot had a diameter of 13 cm and the seedlings were evenly distributed in the pot. Plant cultivation was performed as described in Section 2.3 and the basil was harvested on day 34. The mushroom fruiting bodies that emerged during the basil cultivation period were collected and used for calculation of biological efficiency (BE) of the substrate as described in Section 2.6.

2.5.2. Experiment 2: Oyster mushroom production in fresh and spent substrate

Oyster mushrooms were cultivated in either fresh unused substrate or in similar substrate which had been used previously for basil cultivation (spent potting substrate) (Table 1). Remaining roots in the spent substrate were chopped into small pieces, to a particle size after mixing of <0.5 cm. Both substrates were rewetted with distilled water to a moisture content of 80% and packed into bags suitable for mushroom production (Sac O2, Nevele, Belgium). A total weight of 1.5 kg of substrate (wet weight) was packed into each bag. All bags were pasteurized, spawn of *P. ostreatus* was added in a concentration of 10% (dwt/dwt), and the bags were incubated in a climate chamber at 22 °C and humidity 65%. When the substrate was fully colonized with mycelium, induction of fructification was performed as described in Section 2.4.

2.5.3. Experiment 3: Basil production in spent mushroom substrate

Spent mushroom substrate, collected after harvest of the first mushroom flush, was mixed and limed to reach similar pH as the fresh substrate (6.5 ± 0.1). Different proportions (25, 50, 75%) of the spent mushroom substrate were mixed with fresh substrate. The control treatment was composed of fresh substrate only (Table 1). Basil was sown in the pots and they were placed under greenhouse conditions as described in Section 2.3.

2.6. Analyses

2.6.1. Mushroom and plant analysis

Fresh and dry weight of the harvested oyster mushrooms (first flush only) and basil plants were recorded. Dry weight was determined by drying at 65 °C to constant weight. Mushroom yield (fresh weight) was related to the amount of substrate (dwt), in order to determine the biological efficiency (BE) of the substrate, calculated as:

$$BE = (\text{Fresh weight of mushroom/Dry weight of substrate}) \times 100.$$

For determination of elemental composition, basil and mushroom tissue were wet-combusted in HNO_3 (65%) using a microwave technique (CEN Mars 5) and analyzed by inductively coupled plasma optical emission spectrometry (ICP-OES). Total amount of carbon and nitrogen was analyzed using a Vario Max CN Element Analyzer.

2.6.2. Substrate analysis

Extractable elements in the substrate were determined according to standard EN 13651:2001 (CAT method). Elemental composition of the substrate was performed by wet-combustion and ICP-OES, as described in Section 2.6.1 for plant/mushroom material. In experiment 1, plant cultivation lysimeters (Prenart equipment ApS, Frederiksberg, Denmark) were installed in the pots, and samples were taken at day 2, day 8, and day 27 by applying a pressure of −50 kPa. The liquid obtained from the lysimeters was analyzed for concentrations of ammonium-nitrogen (NH_4^+-N) and nitrate-nitrogen (NO_3-N), and for pH.

2.7. Statistics

All experiments were set up with three replicates in each treatment. The data obtained were analyzed statistically using Minitab 18 for Windows. One-way ANOVA followed by Tukey's multiple comparison test was employed to test for effects of treatments and the significance level was set to $p<0.05$.

3. Results and discussion

3.1. Substrate composition

Compared with the peat only, the macronutrients analyzed (nitrogen, phosphorus, potassium, calcium, magnesium, sulfur) were present in significantly higher concentrations in the mixed peat-biodigestate substrate (Table 2). Particularly large increases were observed for potassium and phosphorus, with approximately 20-fold increases compared with peat. A significant increase in the concentrations of all micronutrients analyzed (copper, iron, manganese, molybdenum, zinc) was observed in the mixed peat compared with the peat only.

The mushroom cultivation in experiment 2 based on fresh substrate (Table 1) resulted in a weight loss of 11.0 ± 4.4% of initial dry weight of the substrate. Compared with the initial composition of the substrate, mushroom cultivation also resulted in a significant decrease in phosphorus concentration, while the concentrations of the other macronutrients were not affected (Table 2). For micronutrients, a significant decrease was observed for iron and zinc, while significant accumulation was observed for copper, manganese, and molybdenum, compared with initial substrate concentrations. Also for the CAT-extractable elements, reflecting plant availability, significant differences were observed in the substrate after mushroom cultivation (Table 3). These included significant decreases in phosphorus and iron concentrations compared with the initial concentration, and increases in concentrations of three other elements, with a major increase observed for sulfur. The latter might be important when biogas digestate is used as plant fertilizer, since the biodigestion process is known to immobilize sulfate, the preferred form for plant uptake (Fontaine et al., 2020). Overall, the nutritional analyses indicated ample amounts of nutrients for plant production in the substrates both before and after mushroom cultivation.

3.2. Nitrogen dynamics in substrate

The dominant nitrogen forms in anaerobic digestate is ammonium and organic nitrogen (Möller and Müller, 2012). Mushrooms generally prefer organic nitrogen (Deacon, 1997), while the mineralized form is the main nitrogen source for plants. However, ammonium can be toxic to plants at high concentrations and may also have secondary adverse effects on plant growth due to acidification of the root environment (Britto and Kronzucker, 2002; Gerendás et al., 1997). The concentration of nitrate in the substrate liquid in experiment 1 was 17–19 mg L^{-1} at the start, while the initial concentration of ammonium in the liquid ranged between 270 and 340 mg L^{-1} (Fig. 1). In the control treatment with basil only, a decrease in nitrate concentration was observed on day 8 compared with initial values, followed by an increase at day 27 (Fig. 1). This pattern, and the measured concentrations of ammonium and nitrate, were similar to those reported by Frerichs et al. (2020) in a study of ammonium exposure of basil when grown in an organically fertilized peat substrate. Our results suggest that naturally occurring nitrifying bacteria, converting ammonium to nitrate, had responded to the high ammonium concentration in the substrate and that nitrification had started. For the treatments receiving mushroom spawn this trend was less evident, possibly suggesting that the presence of actively growing mycelium in the substrate may have interfered with the process of nitrification in the substrate. For pH in the substrate liquid, only minor changes were observed compared with initial values (Fig. 1).

Table 2
Elemental composition of the limed 70% peat-30% solid biodigestate substrate before (fresh substrate) and after mushroom cultivation (SMS). Elemental composition of the peat used in the substrate is shown in brackets in the first column.

Element	Fresh substrate	SMS
C (%)	48.1 ± 0.4a* (51.3 ± 0.6)	43.8 ± 0.1b
N (%)	1.5 ± 0.03a (0.9 ± 0.08)	1.6 ± 0.4a
Ca (mg kg^{-1}, dwt)	18861.4 ± 1753.1a (2415.4 ± 193.6)	15840.5 ± 1312.4a
Cd	0.2 ± 0.05a (0.3 ± 0.05)	0.2 ± 0.02a
Cu	8.8 ± 0.1b (1.3 ± 0.1)	10.1 ± 0.2a
Fe	4174.0 ± 52.7a (1025.3 ± 106.3)	3261.2 ± 69.2b
K	2767.8 ± 14.0a (146.8 ± 10.3)	2716.7 ± 36.6a
Mg	3551.7 ± 165.9a (1457.6 ± 25.8)	3298.2 ± 55.3a
Mn	70.2 ± 1.8b (15.2 ± 0.7)	77.2 ± 2.2a
Mo	0.6 ± 0.07b (BDL**)	0.9 ± 0.07a
P	4573.2 ± 95.6a (194.9 ± 14.8)	3932.8 ± 65.4b
S	3064.2 ± 136.7a (1389.5 ± 109.0)	3237.6 ± 108.2a
Zn	54.8 ± 1.0a (2.7 ± 0.2)	17.3 ± 0.7b

*Values within rows followed by different letters indicate significant difference ($p \leq 0.05$).
**Below detection limit.

Table 3
Extractable elements (mg L^{-1}, wet wt) in the fresh 70% peat-30% solid biodigestate substrate and in spent substrate from mushroom production (SMS).

Element	Fresh substrate	SMS
Cd	<0.007	<0.007
Cu	0.2 ± 0.04a*	0.3 ± 0.03b
Fe	70.3 ± 2.9a	48.7 ± 0.6b
K	226.7 ± 5.8a	230.0 ± 4.5a
Mg	258.0 ± 10.4a	278.7 ± 7.1b
Mn	5.7 ± 0.1a	5.7 ± 0.2a
Mo	<0.1	<0.1
P	183.3 ± 5.8a	170 ± 2.2b
S	68.0 ± 3.6a	153.3 ± 5.8b
Zn	3.4 ± 0.1a	3.5 ± 0.1a

*Values within rows followed by different letters indicate significant difference ($p \leq 0.05$).

3.3. Elemental composition of the crops produced

The elemental composition of the crops will be partly influenced by the composition of the substrate, and may vary significantly for some elements (Burducea et al., 2019). In large-scale commercial biogas production, additives of various kinds are used to ensure stability in production. A common and important additive is ferric chloride, which is mainly used to reduce emissions of hydrogen sulfide (Kutter et al., 2015). This results in high concentrations of iron in the digestate, for example in the present study the iron concentration was 352.7 ± 4.5 mg L^{-1} of substrate (wet weight), which was considerably higher than for the other microelements analyzed. However, the iron concentrations in the basil, 100.0 ± 1.1 mg kg^{-1} dwt, and oyster mushrooms, 100.7 ± 9.6 mg kg^{-1} dwt, produced in the present study were in line with published values (basil 1–109 mg kg^{-1} dwt; oyster mushrooms 33–550 mg kg^{-1} dwt) (Burducea et al., 2019; Mleczek et al., 2018). Thus, use of ferric chloride in the biogas process did not seem to compromise use of the digestate for crop production.

To examine whether mushroom growth in the substrate in combined plant–mushroom production affected plant composition, elemental composition of the basil produced in experiment 1 was analyzed. The results showed similar elemental composition of the basil produced in the control treatment and in the treatment with inclusion of 2% spawn. In the treatment inoculated with the highest concentration of mushroom spawn (10%), four of the analyzed elements

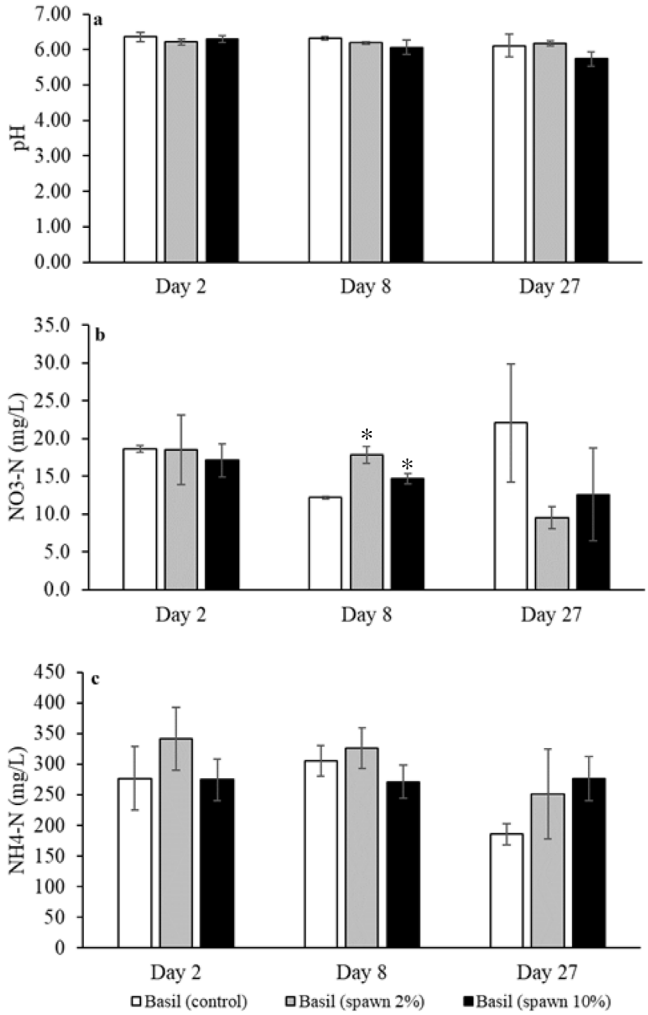

Fig. 1. (a) pH, (b) nitrate-nitrogen (NO_3^--N) and (c) ammonium-nitrogen (NH_4^+-N) concentration (mg L^{-1}) in the liquid sampled from the substrate over time in the different treatments. (* indicate significant difference ($p < 0.05$) compared to control).

(Fe, Mg, Mn and S) were significantly increased compared to the control treatment (Fig. 2). This result can possibly be explained by the fact that colonization of a substrate by white-rot fungi such as *P. ostreatus* will result in degradation of the lignocellulosic content of the substrate. The degradation is due to fungal release of extracellular degrading enzymes (Eichlerova et al., 2000) and in the partly degraded substrate there might be an increased availability of certain elements to the plant.

3.4. Plant and mushroom production in the substrate

In terms of plant production, positive impact of mushroom inclusion in the substrate, with significantly higher yield of basil, was observed when mushroom spawn was included in a concentration of 2% w/w (Table 1). No such increase in plant yield was observed in the treatment receiving 10% w/w (Table 1). The 10% spawn inclusion rate was sufficient for fruiting body formation and it is possible that intense mycelial colonization, a prerequisite for fruiting body formation (Stamets, 2000; Sánchez, 2010), may have interfered negatively with plant growth. A recent study on direct co-cultivation of mushrooms and plants also found no increase in plant yield under conditions allowing fruiting body production (Stoknes et al., 2019). However, Jasinska et al. (2016) observed a significant increase in yield of cowpea when co-cultivated under conditions allowing fruiting body production. It can be speculated that, besides direct competition between growing hyphae and roots during co-cultivation, the plants may have been exposed to certain elements at toxic levels due substrate degradation in this treatment as discussed above. However, none of the analyzed micronutrients in the basil exceeded toxicity levels in plant tissue as established by Marschner (1995).

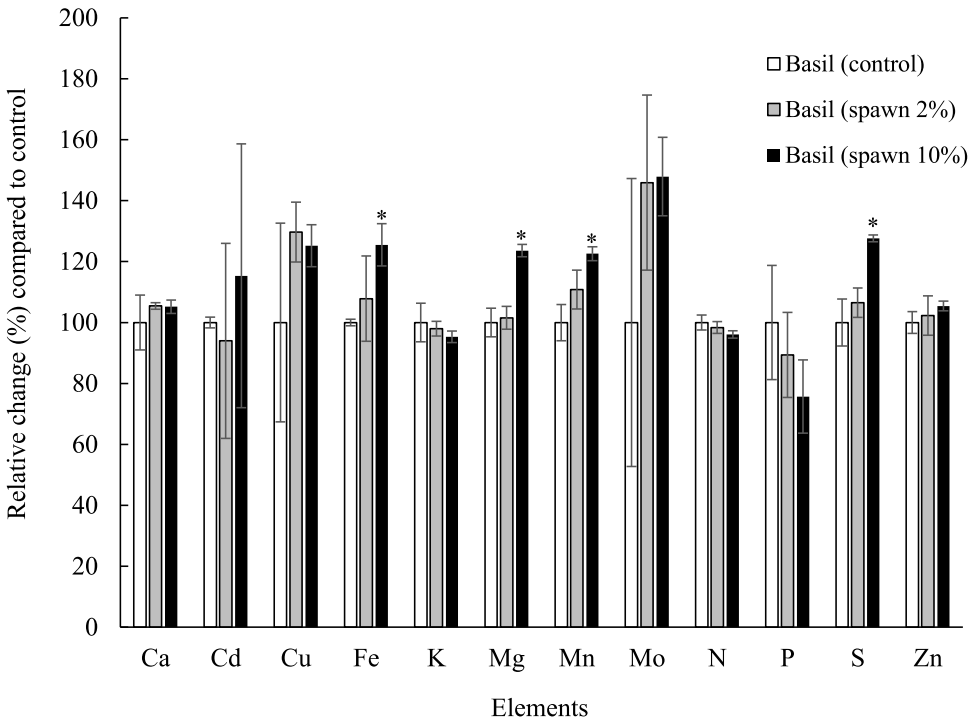

Fig. 2. Relative change (%) in the concentration of different elements in basil on inclusion of mushroom spawn (2% or 10% w/w) in the substrate. (* indicate significant difference ($p < 0.05$) compared to control).

In terms of mushroom production, inclusion of plants in the growing system was less useful. In the pots inoculated with the higher concentration (10%) of spawn in Experiment 1, mushroom growth was observed in the treatments both with basil and without basil, with no significant difference in mushroom yield. Both treatments had BE below 20% (Table 1). In Experiment 2, significantly higher BE was obtained with fresh substrate compared with spent substrate from plant production (Table 1). Thus, neither combined cultivation with plants or use of spent substrate from plant production was beneficial for mushroom production. The BE value of the substrate is an important parameter in mushroom production. In commercial production of *P. ostreatus*, the BE value of the substrate can be expected to exceed 100% (Stamets, 2000). The results obtained in the present study suggest that the used substrate was not optimal for production of oyster mushroom. The highest BE value observed was 50.9 ± 6.1% and this value was obtained in experiment 2 using fresh substrate. The higher BE values observed in experiment 2 compared to experiment 1 were expected, as experiment 2 was performed under conditions optimized for fungal growth, i.e., cultivation in enclosed bags under controlled high humidity (Stamets, 2000; Sánchez, 2010), while experiment 1 was performed under greenhouse conditions best suited for basil production.

In experiment 3, the use of spent mushroom substrate for production of basil was examined. The spent mushroom substrate was mixed into the fresh substrate in different proportions (25, 50 and 75%) and resulting in some differences in elemental composition and availability between the different treatments (Table 2, Table 3). Higher yield of basil was observed for all the treatments with spent mushroom substrate compared to the control treatment with fresh substrate only (Table 1). There was a significant difference in yield of basil related to the inclusion level of spent mushroom substrate and the substrate containing 50% of spent mushroom substrate had a higher yield compared to the inclusion level of 25%. In line with the result of the present study, previous studies on inclusion of spent mushroom substrate in potting substrate have reported beneficial effects (Meng et al., 2018; Medina et al., 2009). However, very variable proportions of spent substrate with different pretreatments have been applied. It should be pointed out that the characteristics of spent mushroom substrate depend largely on the initial composition of the fresh substrate, which needs to be tailored to the selected fungal species. In the study by Medina et al. (2009), spent mushroom substrate from production of white button mushrooms (*Agaricus bisporus*) and from production of oyster mushrooms (*P. ostreatus*) was observed to vary widely in terms of important parameters such as salinity, water-holding capacity, and content of macronutrients. Thus, straightforward comparison between studies is difficult and detailed studies in different production systems are needed. Overall, however, it can be concluded that inclusion of spent mushroom substrate in plant potting substrate is promising for crop productivity and also for circularity, resource efficiency, and waste reduction.

It should also be pointed out that experiments 1 and 3 involved plant production using a similar set-up in the greenhouse. The generally higher plant biomass production in experiment 3, compared with experiment 1 (Table 1), is probably attributable to higher solar radiation, as experiment 3 was performed in June and experiment 1 was performed in February. External radiation was on average 7900 J m^{-2} $week^{-1}$ in experiment 1 and 16700 J m^{-2} $week^{-1}$ in experiment

3. The conventional rule of thumb for greenhouse production is that a 1% increase in radiation will generate 0.5%–1% more yield (Marcelis et al., 2006). Therefore it seems logical to attribute the roughly 100% higher yield in experiment 3 to solar radiation, which was around 100% higher than in experiment 1.

3.5. Practical implications of the study

When developing new systems for sustainable and circular crop production, inclusion of mushroom production would add another dimension to agriculture diversity. The results from this study suggest that substrate based partly on biodigestate can be used first for production of oyster mushrooms and then reused for plant production, with benefits in mushroom production and increased plant yield. However, for greenhouse growers it is possible that the economic benefits of including mushroom production would not outweigh the costs and labor involved in developing new skills and techniques for mushroom production. The need for controlled high humidity during mushroom production imposes different demands on climate control and equipment than used in greenhouse plant production.

On the other hand, the increased yield of basil when including a low concentration (2%) of mushroom spawn in the plant substrate has direct practical implications for greenhouse growers. Spawn is easily available at a reasonable cost from specialist producers and has a shelf-life of approximately 2 months when stored cold (Stamets, 2000). Considering the substantial increase in plant biomass observed on inclusion of mushroom spawn in this study, it can be of interest to develop this system further from an applied perspective.

4. Conclusions

This study investigated integrated plant and mushroom production in substrate based partly on digestate from biogas production. In terms of plant production, positive impacts of mushroom inclusion were observed, both when applying a low amount of fungal spawn to the substrate and when replacing part of the fresh substrate with spent substrate from mushroom production. Thus, plant yield can be increased by adding a mushroom component to the production system. Growing mushrooms in the substrate evidently affected the nutritional composition of the substrate considering both elemental composition and nitrogen dynamics. From the perspective of mushroom production, inclusion of plants in the production system was less useful. Innovative and sustainable systems for food production in densely populated areas are in demand, and combined plant–mushroom production is a future possibility. In the immediate future, the substantial increase in plant biomass production observed here on including low amounts of mushroom spawn in the substrate is of significance.

CRediT authorship contribution statement

Malin Hultberg: Conceptualization, Methodology, Formal analysis, Data curation, Writing – original draft. **Camilla Oskarsson:** Investigation, Methodology, Writing – review & editing. **Karl-Johan Bergstrand:** Conceptualization, Formal analysis, Writing – review & editing. **Håkan Asp:** Methodology, Formal analysis, Writing – review & editing, Funding acquisition.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Ethical approval

This work did not involve any studies with human participants or animals performed by any of the authors.

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