CO-COMPOSTED BIOCHAR (COMBI) PRODUCTION AND ITS EFFECTS ON OCIMUM BASILICUM PLANTS GROWTH

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ABSTRACT: In agriculture, in recent years, more and more attention has been given to the COMBI. COMBI is a soil improver obtained from a composting process of organic material with biochar, added at the beginning of the process itself. Biochar has been used for several years and many studies have shown the benefits it brings when applied to soils, while COMBI is still little studied. This article aims to demonstrate both the effects of COMBI on the growth of *Ocimum basilicum* specie plants and to quantify the best quantity to be applied within the substrate. The experimental trials were performed in a greenhouse with controlled temperature and humidity, according to the following growth tests theses: control: soil for gardens composed mainly of peat (CTRL); thesis 1: soil mixed with COMBI at 10% v / v (COMBI10); thesis 2: soil mixed with COMBI at 20% v / v (COMBI20); thesis 3: soil mixed with COMBI at 40% v / v (COMBI40); thesis 4: soil mixed with COMBI at 80% v / v (COMBI80); thesis 5: 100% COMBI (COMBI100). The results showed good growth of basil plants in the COMBI10 and COMBI20 cases, compared to the control. COMBI80 and COMBI100 theses, on the other hand, did not lead to positive results; in particular COMBI100 did not allow either seeds germination in most cases. To compare the quality of the basil plants, the total amount of essential oil for CTRL, COMBI10, COMBI20 and COMBI 40 theses was extracted and quantified.
Keywords: biochar, agricultural residues, composting.

1 INTRODUCTION

Co-composted biochar, called COMBI, is an organic product that is formed after aerobic decomposition of organic material with the initial addition of biochar. It can be made from any organic material: organic fraction of municipal solid waste (OFMSW), pruning of trees and grass [1,2], waste from public green maintenance or from the food industry, etc. The big difference between COMBI and normal compost is that a known amount of biochar is added to the organic material at the beginning of the decomposition process. Hence the name of "co-composting process".

Biochar has been used for several years as soil conditioner and many studies have shown the benefits it brings when applied to soils. Several research have shown how biochar has water [3] and nutrients retention properties.
Therefore, if applied to soils, it allows a reduction in the amount of water and fertilizers that must be supplied to crops. It is also demonstrated that, when used as a soil improver, an increase in crop productivity is achieved [4].

COMBI, on the other hand, although in recent years it has begun to spread, it’s not well known and consequently rarely used. In literature, to date, the topic is little studied. However, some researches have shown that there are greater advantages in the use of COMBI [5] in soils as a substitute both for biochar and for mix composed of biochar and normal compost (after decomposition process).

Biochar is a coal produced through a gasification and/or pyrolysis process. Through the revaluation of agricultural waste, for example by gasifying residual biomass, thermal and electrical energy and biochar are produced [6] [7] [8] [9]. The biochar thus obtained can then be reused as soil improver as it is or to produce COMBI, which can then be re-used in the farm from which the waste comes. Biochar can also be produced through pyrolysis processes, exploiting, for example, heat from gasifier [10] that otherwise would not be recovered.

Other studies have shown how char, thanks to its high porosity, is also applicable in the industrial field as filter medium for producer gas [11] [12], instead of more common filtration methods such as baghouse filters [13], or for greenhouse gases (GHG) reduction [14]

Of all the previous uses of biochar, perhaps, the simplest is as additive in compost processes.
In fact, biochar improves the performance of composting processes in a broad sense. It reduces the density of the material making it more manageable, reducing the risk of anaerobic process; increases the temperature of the compost; reduces emissions of GHG and nitrogenous compounds; retains more water and leachate, reducing the risk of drying the biomass, and by increasing the degree of humification, produces a higher quality compost [15] [16]. Furthermore, it has been seen that the biochar used in composting processes leads faster to the stability of the final product [17], which can therefore be used as a soil improver in advance compared to normal compost.

COMBI must be mixed with the soil when used as a soil improver. Some research has been carried out to demonstrate the benefits COMBI brings when it is used both in pot and in field, for different types of crop and vegetables [18], using both biochar and hydro-char as initial additive in the co-compost process [19].

This preliminary study aims to demonstrate both the effects of COMBI, produced with waste biomass from industrial hemp farm, on the growth of *Ocimum basilicum* plants and to quantify the best quantity to be applied within the substrate. To test this, five different theses (in addition to the Control) were prepared at different percentages of soil mixed with a different amount of commercial garden soil, in volume: 10% v/v, 20% v/v, 40% v/v, 80% v/v and 100%.

2 MATERIAL AND METHODS

2.1 COMBI Production

COMBI used in this research was produced through a pilot-scale reactor for Aerated Static Pile (ASP) simulation, in order to better manage the amount of air necessary for the process, both to provide the right amount of oxygen to microorganisms and to keep temperatures in the optimal range for the composting biological reactions and bacterial proliferation, to ensure that the process did not fall into anaerobic digestion.

Shredded leaves and flowers of industrial hemp plants (Cannabis sativa) were used to produce COMBI. Biochar was then added to this organic material in order to obtain a mix in which biochar is present at 10% of the total volume. 945 liters of leaves and flowers and 105 liters of char were used, to obtain about 1 cubic meter of material, maximum size of the compost bin. During the preparation phase of the mix, 210 liters of water were also added to obtain a moisture content of around 50%.

An automatic Arduino-based system has blown air into the compost bin for 30 days. The automatic system was able to blow air, through an air-sparger positioned on the bottom of the bin, at regular interval of time. The air flow rate was constant, while the intervals of blowing were modified according to the biomass temperature (measured through a thermocouple). The goal was to maintain the temperature in the optimal decomposition range, between 55 and 60 ° C. After this initial 30 days, another phase called "maturation stage" (always 30 days) were then required before the COMBI could be used.

Figure 1: "Co-compost" mix at the beginning of the process

2.2 Plant growth test

Five different theses were identified to be tested in addition to the control test. A commercial soil optimized for vegetable gardens, composed mainly of peat, was used as substrate for the control test. The substrate for the different theses, instead, was obtained by replacing a part of the volume of garden soil with COMBI. The substrates were mixed through a mud mixer.

The aim of this thesis is to identify the optimal quantity of COMBI to mix with the initial substrate to identify benefits from an agronomic point of view.

The thesis tested are: Control - soil for gardens (CTRL); Thesis 1: soil mixed with COMBI at 10% v/v (COMBI10); Thesis 2: soil mixed with COMBI at 20% v/v (COMBI20); Thesis 3: soil mixed with COMBI at 40% v/v (COMBI40); Thesis 4: soil mixed with COMBI at 80% v/v (COMBI80); Thesis 5: 100% COMBI (COMBI100). A scheme showing the theses is presented in Figure 2.

Figure 2: Scheme of the 6 different substrates which have been tested during the experimental campaign

These different substrates were used to prepare 8 pots for each thesis in which 6 *Ocimum basilicum* seeds were planted. After germination took place, only three plants were kept and those in excess were eliminated. The pots were placed in a greenhouse at controlled temperature and humidity, illuminated by lights for plant growth (Fig. 3a). Photosynthetically Active Radiation (PAR) values measured at the top of the pots varies between 200 and 230 µmol m^{-2} s^{-1}.

Figure 3: Growth test pots in the greenhouse.

2.2 Analysis

The evaluation of *Ocimum basilicum* growth on different COMBI percentages has been performed through the analysis of seed germinability rate (SGR), fresh weight and dry weight biomass (FW and DW), plants high and essential oil production. Dry weight has obtained through slow drying process at 35°C lasted 7 days.

High and "diameter/high" ratio of each plant has been measured with caliber. SRG has been calculated using formula (1).

$$SGR\ (\%)\ = \frac{number\ of\ germinated\ seeds}{number\ of\ total\ seeds}\ x\ 100 \qquad (1)$$

The weight of all plants grown in each pot were measured using a scientific grade scale; the precision of the instrument is 0.0001 g. Essential oil extraction was performed using Clevenger apparatus for hydro-distillation. Fresh biomass was used to extract essential oils, which have been collected using a defined amount of xylene. At the end of 40 minutes of hydro-distillation process for each sample, xylene and total amount of oils was measured and the correct amount of these has been calculated for difference.

The results were processed with analysis of variance (ANOVA) to get representative and comparable indicators among the different thesis and control.

3 RESULTS

3.1 Biomass growth

55 days after plantation, basilicum plants were harvested and growth test parameters were measured in laboratory. As shown in Figure 4, CTRL, COMBI10, COMBI20 have averagely a similar growth. In contrast, high percentage of COMBI (in particular 80% and 100%) as substrate have an inhibitory effect on seed germination and plant growth. SGR have been calculated and reported in Table 1. The addition of low percentage of COMBI slightly improves SGR compared to control substrate.

However, these are preliminary results, which need to be confirmed with more repeated test.

Figure 4: Basilicum plants from all investigated thesis (final time).

Seed Germinability Rate, reported in Table 1, is calculated on 6 seed planted in 8 repeated pots for each thesis.

Table I: Seed Germinability Rate and increase/decrease of FW biomass production: comparison of different thesis

Samples	Seed germinability rate	Fresh weigh	Increase and decrease in %
CTRL	79.2 %	67.9 g	
COMBI10	81.3 %	74.2 g	+9.4%
COMBI20	93.8 %	72.1 g	+6.3%
COMBI40	89.6 %	50.6 g	-25.5%
COMBI80	35.4 %	5.1 g	-92.6%

Plants grown in COMBI10 and COMBI20 substrates show a slight increase in plant biomass production (Figure 5): specifically, the improvement of mean FW biomass was of 9.4% and 6.3% respectively compared to CTRL, as underlined in Table 1. COMBI40 and COMBI80, instead, show a significant decrease of mean biomass produced. However, ANOVA doesn't reveal any statistically significant increase of treated samples (p-value >0.05) compared to CTRL, but statistical analysis is significant (p- value<0.05) for the decrease occurred on COMBI40 and COMBI80 growth.

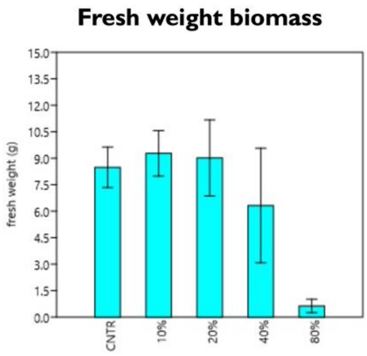

Figure 5: Fresh weight produced on different thesis and control pots. Final result is the mean of 8 pot

3.2 Other parameters

Plants high trend is reported in Figure 6, where percentiles correlated with plants high values (cm) were shown. Highest 95° percentile was achieved by COMBI10 samples (~22 cm), immediately followed by COMBI 20, which is very similar. C TRL and COMBI40 have a lower 95° percentile (~18cm). Considering diameter/high rate (Figure 6), plants grown on COMBI80 reached the highest values; however, this parameter does not mark any improvement in plants growth on COMBI80 because this substrate is not associated with a higher biomass.

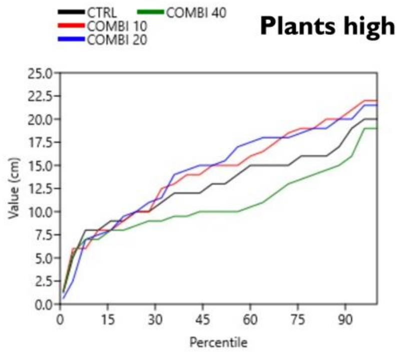

Figure 6: Percentile of plants high

3.3 Essential oil

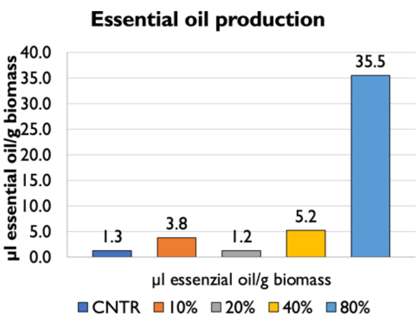

Figure 7: Essential oil

Data about essential oil production are referred to the first hydro-distillation trial run on plants biomass. Therefore, these data should be considered as preliminary and need to be confirmed. In this first report (Figure 7) we can mark how COMBI80 produces higher amount of essential oil, compared to other thesis; this phenomenon could be due to the lower growth rate of plants compared to the other thesis and following concentration of essential oil production in grown plant biomass.

3.4 Peat and COMBI

A commercial soil suitable for home gardens was used in the research. This soil, like most commercial ones, contains a high percentage of peat. Although peat does not contain nutrients, it is a highly efficient substrate because it can absorb water and nutrients and release them over time [20].

These characteristics are very similar to those of the COMBI. In addition, COMBI is naturally rich of microorganisms, useful for plant growth.

As a last point, it is recognized that the peat extraction process is not environmentally sustainable, and it is carbon positive. COMBI, instead, is carbon negative and it can also be produced through waste or residual biomass.

The results obtained in this research, did not show a significant growth of the *Ocimum Basilicum* plants planted in the COMBI compared to those in normal soil. New studies will be needed to assess whether there is a real improvement in the use of COMBI, but certainty that peat and COMBI have given the same results.

This study opens the way for further research that aims to demonstrate the possibility of replacing classic peat with COMBI, to reduce the environmental impact of commercial soil production and extraction, while maintaining high soil productivity standards.

4 CONCLUSIONS

1) Co-composting process using hemp residues and biochar gives as result COMBI, which can be used as soil amendment. Mixing low percentage (<40%) of COMBI with commercial soil did not show any damage or pathogen development during seed germinability or plant growth: it can be considered as a safe amendment.

2) *Ocimum basilicum* plants growth on substrates enriched with COMBI 10 and COMBI 20 produced a slight increase in FW biomass amount; however, the increase is not statistically significant, and it need to be confirmed.

3) Commercial soil for plants growth is constituted mainly by peat, which maximises soil composition in terms of organic matter and nutritional composition. However, peat production process is not sustainable. Our future perspective involves COMBI production and its application on depleted soils, in order to reduce peat production and its environmental impact and, at the same time, to keep high soil productivity.

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