

# Integrating Compost and Biochar for Improved Air Quality, Crop Yield, and Soil Health

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# List of Abbreviations

ANOVA	analysis of variance
C	carbon
°C	degrees Celsius
C:N	carbon to nitrogen ratio
cc	cubic centimeter
CEC	cation exchange capacity
CH <sub>4</sub>	methane
ChA	chicoric acid
cm	centimeter
cm <sup>3</sup>	cubic centimeter
CO	carbon monoxide
CO <sub>2</sub>	carbon dioxide
CS <sub>2</sub>	carbon disulfide
DMDS	dimethyl disulfide
DMS	dimethyl sulfide
DNPH	2,4-Dinitrophenylhydrazine
DTPA	diethylenetriaminepentaacetic acid
°F	degrees Fahrenheit
g ai ha <sup>-1</sup>	grams active ingredient per hectare
g	gram
GC-MS	gas chromatography-mass spectrometry
H <sub>2</sub> O	water
H <sub>2</sub> S	hydrogen sulfide
ha <sup>-1</sup>	per hectare
He	helium
HSD	honestly significant difference
K	potassium
kg	kilogram
L min <sup>-1</sup>	liter per minute
L	liter
LC-MS	liquid chromatography-mass spectrometry

LC-PDA-MS	liquid chromatography photodiode array mass spectrometry
m	meter
$m/z$	mass to charge ratio
$m^2$	square meter
meq	milliequivalents
MFC	mass flow controllers
$Mg\ ha^{-1}$	Megagrams per hectare
Mg	magnesium
mL	milliliter
mm	millimeter
N	nitrogen
n	sample size
$N_2$	nitrogen gas
$N_2O$	nitrous oxide
$NH_3$	ammonia
$NH_4-N$	ammonium nitrogen
NIST	National Institutes of Standards and Technology
nm	nanometer
NO	nitric oxide
$NO_3-N$	nitrate-nitrogen
$NO_x$	nitrogen oxides
$O_2$	oxygen gas
OBS	Oregon Biochar Solutions
p	probability value
PFA	perfluoroalkoxy
POM	particulate organic matter
ppbv	parts per billion by volume
ppm	parts per million
ppmv	parts per million by volume
psig	pounds per square inch, gauge
PTR-MS	proton transfer reaction mass spectrometer
$PV_{meso}\ (cm^3\ g^{-1})$	Meso pore volume
$PV_{micro}\ (cm^3\ g^{-1})$	Micro pore volume
$PV_{Total}\ (cm^3\ g^{-1})$	Total pore volume



RA	rosmarinic acid
S	sulfur
s.e.m.	standard error of the mean
Sa <sub>CO<sub>2</sub></sub> (m <sup>2</sup> g <sup>-1</sup> )	Surface area obtained by using CO <sub>2</sub> gas
Sa <sub>N<sub>2</sub></sub> (m <sup>2</sup> g <sup>-1</sup> )	Surface area obtained by using N <sub>2</sub> gas
sccm	standard cubic centimeters per minute
SLPM	standard liter per minute
TSQ	Thai Siam Queen
UHP	ultrahigh purity
US EPA	United States Environmental Protection Agency
USDA NASS	US Department of Agriculture National Agricultural Statistics Service
UV	ultraviolet
VOC	volatile organic compound
w/v	weight per 100 mL
WSU	Washington State University
wt.	weight
XIC	extracted-ion chromatogram
yd <sup>3</sup>	cubic yard
Zn	zinc
μg	microgram
μm	micrometer
μmol x•m <sup>-2</sup> •s <sup>-1</sup>	micromole per second per square meter

# Abstract

Included in this 2019-2021 biennium report are new data describing additional work from projects reported on in the 2017-2019 biennium. Soil and yield data for strawberry, basil and potato field trials now include data and analysis from an additional growing season.

This report describes work that explores the integration of composting with biochar. There are indications that biochar, when added to other feedstocks at the beginning of the composting process, can reduce emission of VOCs during composting. Two field sampling experiments and two laboratory experiments therefore examined the effect of biochar on emission of VOCs from compost. Results of laboratory experiments indicate that 10% biochar can significantly reduce emissions of monoterpenes, dimethyl disulfide, and other compounds which are not yet identified. Reduced emission of these compounds would help reduce compost odor.

Meanwhile, biochar, compost, and co-compost – the product of composting traditional feedstocks with biochar – have also been identified as potential soil amendments that, after surface application and incorporation can increase crop yield and improve soil health. The greenhouse and field trials tested the effect of compost, biochar, co-compost, and compost plus biochar as soil amendments in a variety of different cropping systems and sites in Washington: sweet basil (field, Colbert, Washington), basil (greenhouse), strawberry (greenhouse), strawberry (field, Puyallup, Washington), and potato (field, Mount Vernon, Washington). Soil and yield data for strawberry, basil and potato field trials now include data and analysis from an additional growing season. This biennium's work expanded and replicated these trials and validated previous results, including finding that basil treated with different biochar-amended composts showed moderate increases in biomass production. Continued greenhouse-based experiments with strawberries indicated productivity increases were observed in some of the biochar-compost treatments but were only moderate overall.

Expanded results from additional growing seasons and analysis indicated in general, significant effects on crop yield, that varied by amendment type, crop, and soil type. In greenhouse experiments, a strawberry cultivar and two basil cultivars showed an increase in berry mass production or in biomass, respectively. In the field, a much larger increase (almost two-fold) was observed for basil produced under organic growth conditions. In field experiments, potatoes grown with co-compost also showed a significant (but smaller) increase in yield, but strawberries did not exhibit statistically significant differences. Amendments to the soil did not significantly affect the phytochemical composition of field- or greenhouse-grown sweet basil, an indication that product quality (flavor, smell) was not compromised by addition of biochar. Co-compost, compost, and the compost plus biochar amendments were typically observed to affect soil physical and chemical properties beneficially in Puyallup and Mount Vernon field trials, but it seems that this effect is somewhat dependent upon the native soil and the amendment's application rate and nutrient content. Depending on the plant species and growth conditions, treatments yielded minimal to dramatic increases in biomass.

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# 1. Background

## 1.1 Effect of biochar on compost emissions

In recent decades, composting has been widely used to convert organics waste into nutrient-rich soil amendments. The use of compost is known to enhance soil fertility, water holding capacity, and organic matter levels (Kebreab et al., 2006; Fischer and Glaser, 2012). Production of compost often causes odor and/or greenhouse gas emissions. Application of biochar, defined as “a solid material obtained from thermochemical conversion of biomass in an oxygen-limited environment” by the International Biochar Initiative (Agegnehu et al., 2017) to reduce gas emission during and after the composting process is a promising efficient low-cost solution to this problem (Godlewska et al., 2017; Sanchez-Monedero et al., 2018a).

Literature shows that mineral, organic, or biological substrates addition to compost can affect parameters controlling composting, gas emission and compost quality (Barthod et al., 2018). Among the additives available, biochar has received intensive attention for use in composting and soil remediation. Biochar is a charcoal produced from burning biomass through controlled process called pyrolysis. Due to its high water holding capacity and porosity, it can hold moisture content in compost (Wang et al., 2013).

### 1.1.1 Methane and carbon dioxide

Biochar can be used as a carbon sink in compost to reduce emissions of CH<sub>4</sub> and CO<sub>2</sub> in compost facilities (Zhang et al., 2012; Clough et al., 2013). Jia et al. (2016) compared chicken manure compost amended with 20% biochar with a control (no biochar) and observed 55% less emission of CH<sub>4</sub> from biochar but 148% higher emission of CO<sub>2</sub>, indicating that high porosity of biochar provided more O<sub>2</sub> for aerobic organisms and better absorption/adsorption of CH<sub>4</sub>. Kammann et al. (2017) observed that biochar reduced methane fluxes in flooded soils, especially if N fertilizer use was limited. However, contradicting results also have been reported in the literature about how applying biochar to paddy or flooded soil might affect CH<sub>4</sub> fluxes. Increases in CH<sub>4</sub> fluxes resulting from biochar addition have been reported (Yu et al., 2013; Zhang et al., 2012), while other studies have found reduced methane emissions (Khan et al., 2013; Lin et al., 2015; Qian et al., 2014), or and no significant impact (Xie et al., 2013). Therefore, the interconnection of biochar and methane flux are not well understood.

### 1.1.2 Nitrous oxide

Nitrous oxide is another greenhouse gas that is produced through two main processes for removal of nitrogen from wastes, nitrification and denitrification, therefore, it is emitted from biodegradation of organics in composting. Since it has high efficiency in absorbing infrared radiation and can react with single atomic oxygen in stratosphere to destruct ozone, there is a need for research to quantify N<sub>2</sub>O emission from compost facilities. He et al. (2001) conducted an experiment with a laboratory-scale composter to generate nitrous oxide from aerated composting of food stocks. Then the results of emission rates were used to find the optimum

operational condition to minimize N<sub>2</sub>O emission. It was shown that when carbon sources are available, N<sub>2</sub>O is produced mostly later time of composting (after depletion of carbon source). Another result of this experiment was related to periodic feeding of compost with fresh organic wastes that postponed N<sub>2</sub>O emission and reduced its emission by 20%. A few studies have focused on how biochar can influence N<sub>2</sub>O emission, most of them carried out an experiment in laboratory using sieved soil (Zwieten et al., 2012; Kammann et al., 2012; Cayuela et al., 2014; Schimmelpfennig et al., 2014; Deng et al., 2015; Hüppi et al., 2015) and overall confirmed that N<sub>2</sub>O emissions were reduced. However, laboratory results cannot be generalized to field observations. In the field trials, no differences in biochar treatment was recorded compared to the control (Karhu et al., 2011; Scheer et al., 2011; Castaldi et al., 2011; Schimmelpfennig et al., 2014). The reason for that might be associated with heterogenous distribution of organisms in the field leading to high variability in measured N<sub>2</sub>O fluxes (Hüppi et al., 2015).

### **1.1.3 Volatile organic compounds**

During composting several types of VOCs including alcohols, ketones, sulfur compounds, and monoterpenes can be emitted to the atmosphere which might cause environmental hazards. For instance, H<sub>2</sub>S is generated during anaerobic conditions of composting process. To our knowledge only a few studies investigated biochar impact on H<sub>2</sub>S emissions in composting. Steiner et al. (2010) reported that compost mixed with 5 and 20% biochar emitted 58% and 71% less H<sub>2</sub>S. They also observed that ammonia emission from compost amended with 20% biochar decreased by 64%. In another study, 3% biochar was mixed with composting mixture of poultry manure and barley straw. Results showed that formation of H<sub>2</sub>S happened during the peak time period of microbial activity and biochar treatment did not affect H<sub>2</sub>S emission.

Although some studies have shown that use of biochar in composting organic wastes reduce emissions of greenhouse gases, more detailed studies are required for validation and quantification of this effect. Moreover, little is known about the effect of biochar treatment on VOC emissions from composting process.

The aim of this project was to provide measured data through field and laboratory tests in order to identify emitted VOCs and odorants and to quantify their emission fluxes from composting processes.

## **1.2 Effect of biochar and co-composted biochar as a soil amendment**

The use of biochar-compost mixtures resulting from co-composting for agriculture is an emerging technology holding great promise (Agegnehu et al. 2017). A large multi-national study recently concluded that though the effects of biochar, compost, and biochar-compost blend (biochar added at the beginning or at the end of the composting process) vary depending on feedstocks, the use of compost-biochar blends led to yields comparable to those obtained with mineral fertilizer, with a cumulatively lower environmental impact (Oldfield et al., 2018). Co-composted biochar increased yields, in comparison to compost-only, for oats (Schulz et al., 2013) and barley (Agegnehu et al., 2016). Clear benefits of co-composting in comparison to

biochar added after composting (hereafter referred to as biochar plus compost) were observed in a study evaluating the biomass yield and nitrogen uptake of quinoa (Kammann et al., 2015). Our previous study indicated greater biomass yield of sweet basil grown with co-composted biochar-compost mixtures as compared to treatments where biochar was added after composting (Gang et al., 2018). At the same time, some studies delivered opposite results, suggesting that the effects of biochar-compost mixtures are highly dependent on specific application details (Gang, 2018). Benefits and drawbacks of biochar application may be related to its electrochemical properties (Chacon et al., 2017), its interactions with soil microbiota (Hol et al., 2017), and numerous other parameters (Agegnehu et al., 2017). For example, biochar can adsorb herbicides and reduce their efficacy. Bioavailability of the herbicides S-metolachlor and sulfentrazone were reduced more by high surface area biochar than low surface area biochar at the same application rate (Graber et al., 2012). Surface adsorption and reduction in herbicide efficacy was lower in a long-aged biochar (greater than 50 years) than in a new biochar, possibly due to the formation of new surface functional groups (Cheng et al., 2014). Studies are being designed and executed to help clarify the impacts of different variables and enable prediction of effects (Bonanomi et al., 2017). In addition, the properties of the agronomical environment play a crucial role in determining the effects of biochar and/or compost application. There is evidence that biochar amendment is most effective for degraded or nutrient-depleted soil (Agegnehu et al., 2017).

A study with strawberries grown in white peat with a 3% (by weight) biochar showed increased plant growth, reduced leaf and post-harvest fruit susceptibility to *Botrytis cinerea*, and a shift in rhizosphere-associated microbial community (De Tender et al., 2016a; De Tender et al., 2016b). These effects were diminished when inorganic fertilizer and lime were co-administered, suggesting higher efficacy of biochar under conditions of limited nutrient availability. In the same study, biochar had no effect on lettuce grown in sandy loam soil (De Tender et al., 2016b). A time-course experiment revealed that the effects of biochar on the microbial community were evident from week six of growth onwards (De Tender et al., 2016a). Notably, (Harel et al., 2012) demonstrated biochar-related enhancement of strawberry plant resistance to *Botrytis* infection in an entirely different experimental setting.

It has been documented for both biochar and compost that feedstock has an important effect on the quality of the resulting product (Jindo et al., 2014; Agegnehu et al., 2017; Domingues et al., 2017). Most recently, a comparison of wood- with green waste biochar for their ability to reduce tomato early blight disease highlighted the importance of the biochar feedstock (Rasool et al., 2021). In this study, we evaluate three types of biochar-compost products resulting from combinations of two types of compost and two sources of biochar. The goal of the experiment was to compare the effects of different biochar-compost products.

Three crops were used in this current study. Basil is an important high-value crop in the State of Washington, as described in our previous report (Gang et al., 2018). In addition to biomass productivity and aroma composition, an important quality parameter for a culinary herb like sweet basil, we assessed the effect of biochar-compost amendments on the accumulation of antioxidant polyphenolics that add nutritional value for the consumer (Nunes et al., 2017). The second specialty crop in this study is strawberry (*Fragaria x ananassa*). The Pacific Northwest region including Oregon, Washington, and British Columbia (Canada) produces approximately 32 million pounds annually of strawberries, which are mostly processed or sold via direct market (Samtani et al., 2019). The predominant areas of commercial strawberry production are in the Skagit and Nooksack valleys in Washington. Albion is the major day-neutral cultivar grown. The

third crop examined in this study is potato, which is a major crop grown in Washington, with 169,000 acres of potatoes harvested in the state in 2016 (USDA NASS, 2017). The Skagit Valley of Washington is a major producer of potatoes for the fresh market.

## 2. Objectives

The specific objectives of this project were:

- 1.) To provide measured data through field and laboratory tests in order to identify emitted VOCs and odorants and to quantify their emission fluxes from composting processes;
- 2.) To evaluate the impact of biochar, compost, co-composted biochar, and biochar plus compost on
  - productivity and quality of sweet basil in the greenhouse and the field;
  - productivity and quality of strawberries in the greenhouse and the field; and
  - productivity and quality of potatoes in the field.
- 3.) To evaluate the impact of biochar, compost, co-composted biochar, and biochar plus compost on soil physiochemical properties in the strawberry and potato field trials.



## 3. Methods

### 3.1 Compost generation and properties

#### 3.1.1 Compost generation

Overall, four independent compost batches were used in different parts of the investigation:

##### *Composts produced at the WSU composting facility in 2017*

Generation of these composts was previously described by (Gang et al., 2018). The biochar used was obtained from Amaron Energy (Salt Lake City, UT), and produced using a patented rotary kiln unit. This compost will be referred to as “WSU 2017 compost.”

##### *Composts produced at the WSU composting facility in 2018*

In 2018, biochar (Oregon Biochar Solutions, White City, OR) was used to produce compost at the WSU composting facility. Biochar for the project was generated in two modified biomass boilers that convert Douglas fir (*Pseudotsuga menziesii*) and pine (*Pinus* spp.) forestry residuals into biochar at 871°C in a low oxygen environment (Oregon Biochar Solutions, White City, OR).

Overall, twelve piles were made. Each compost pile contained: 4.5 yards of screened manure solids from the Washington State University (WSU) dairy, 4.5 yards of dairy bedding straw and manure, 0.5 yards of ground clean green (woody) yard trimmings, and 0.5 yards of food waste from the WSU dining commons. These components were mixed and stacked in separate 10 yard piles, on 4 and 5 June. At least one of the piles from each treatment was built on a different day to avoid “day effect.” At the initiation of the composting process, biochar was mixed: three piles at 2.5%, three at 5% and three at 10% (by volume). Hereafter, percentages referred to in biochar mixtures indicate percent by volume. Each treatment level was assigned to three piles and the remaining three piles contained no biochar (compost control). The piles were turned one day prior to sampling for air emissions (described below). The piles were monitored for temperature (maintained >57°C) and bulk density throughout the composting process. By the end of the composting period, the piles had reduced to about 5 yards each. This compost will be referred to as “WSU 2018 compost.”

##### *Composts produced at Lenz Enterprises in 2018*

Lenz Enterprises is a commercial compost operation that employs a three-stage composting process that uses municipal yard and food waste in an aerated static pile. Piles were turned once during the aeration period, then moved out of the aerated composting bay and allowed to compost further and cure. The biochar co-composted amendment was made simultaneously with the same feedstocks with the addition of 5% biochar by volume (Oregon Biochar Solutions). Both piles were composted in the same 800 yd<sup>3</sup> aerated bin and were marked to keep them separated with the biochar co-compost having a starting volume of 200 yd<sup>3</sup>. This compost will be referred to as “Lenz compost.”

Compost air emissions were sampled on days 3, 7, 10, 16, and 22 (WSU) and on days 3, 7, 11, and 31 (Lenz). Sample collection and analyses were carried out as described previously by (Gang et al., 2018).

*Composts produced at WSU Puyallup Research and Extension Center in 2019*

Compost produced in 2019 at the Puyallup Research and Extension Center is described in detail in Chapter 4, and in Stacey et al. (2021). To create the biochar amended co-composts, we first developed a base compost mixture which was made from locally procured chicken manure and wood shaving feedstocks. The biochar was purchased from Oregon Biochar Solutions (White City, OR). This compost will be referred to as “Puyallup 2019” or “Puy 2019”.

**3.1.2 Biochar and compost properties**

The properties of 3 lots of the OBS biochar were determined by the Garcia-Perez laboratory at WSU (Tables 1-3).

**Table 1: Elemental analysis of OBS biochar**

Sample	Carbon%	Hydrogen %	Nitrogen %	Sulfur %	Oxygen %*
Lot_01	85.471	0.861	0.782	0.033	6.040
Lot_02	88.216	0.776	0.759	0.025	4.568
Lot_03	88.136	0.754	0.809	0.037	3.281

\* Obtained by difference

**Table 2: Proximate analysis of OBS biochar**

Sample	Volatile Carbon %	Fixed Carbon %	Ash %	Moisture %
Lot_01	10.481	84.684	4.835	2.226
Lot_02	9.270	88.025	2.706	3.320
Lot_03	9.911	86.431	3.658	3.739

**Table 3: Surface area and pore volume of OBS biochar**

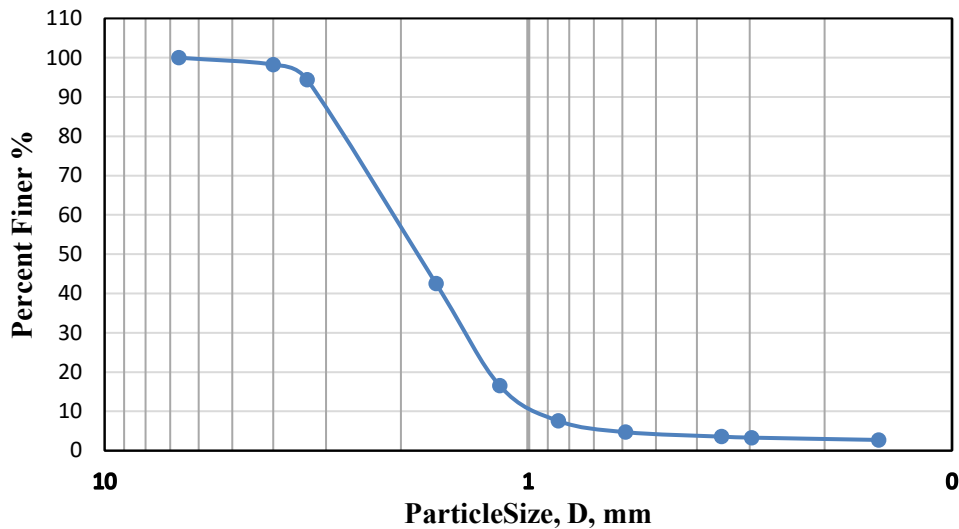
SaCO <sub>2</sub>	Sa <sub>N<sub>2</sub></sub>	V <sub>micro</sub>	V <sub>meso</sub>	V <sub>Total</sub>
m <sup>2</sup> /g	m <sup>2</sup> /g	cm <sup>3</sup> /g	cm <sup>3</sup> /g	cm <sup>3</sup> /g
549.3	520.9	0.22	0.08	0.30
580.3	547.2	0.23	0.06	0.29
592.9	531.5	0.24	0.06	0.30

$S_{N_2}(\text{m}^2 \text{g}^{-1})$ - Surface area obtained by using  $\text{N}_2$  gas  
 $S_{CO_2}(\text{m}^2 \text{g}^{-1})$ - - Surface area obtained by using  $\text{CO}_2$  gas  
 $PV_{\text{micro}} (\text{cm}^3 \text{g}^{-1})$  - Micro pore volume

Particle size distribution: Samples were placed in the first sieve and shaken for about 30 minutes. Next, the sample retained in each sieve was weighed. Table 4 shows the particle size distribution. Figure 1 shows a graphical representation of the sieve size (mm) and the percent finer (%) (Passed particles).

**Table 4: Particle size distribution of OBS biochar**

Sieve Size (mm)	6.680	4.000	3.327	1.651	1.168	0.850	0.590	0.350	0.297	0.149	Pan
Empty Sieve (g)	512.8	566.1	487.2	448.4	389.9	420.5	457.7	461.4	443.8	427.4	525.1
After 30 min shaking (g)	512.8	567.4	489.9	484.9	408.2	426.8	459.7	462.2	444.0	427.9	527.0
Mass Retained (g)	0.0	1.2	2.7	36.5	18.3	6.3	2.0	0.8	0.2	0.4	1.9
Cumulative weight (g)	0.0	1.2	3.9	40.4	58.7	65.0	67.0	67.8	68.0	68.4	70.3
Cumulative weight (%)	0.0	1.8	5.6	57.5	83.5	92.4	95.3	96.4	96.7	97.3	100.0
Passed Particles (%)	100.0	98.2	94.4	42.5	16.5	7.6	4.7	3.6	3.3	2.7	0.0



**Figure 1: Particle Size Distribution Curve (x-axis is in log scale)**

Additionally, some of the properties of the same biochar and simultaneously of the Lenz compost and co-compost were measured by the Collins team (Table 5 below).

**Table 5: Selected chemical properties for biochar (OBS), compost (Lenz), and co-compost.**

	<b>Carbon (wt. %)</b>	<b>Nitrogen (wt. %)</b>	<b>Moisture (wt. %)</b>	<b>Ash (wt. %)</b>	<b>C:N ratio</b>
<b>Amendment</b>					
Biochar	87	0.78	15.0	11.2	112
Compost	32	2.2	52.1	38.1	15
Co-compost	32	2.2	52.7	38.9	15

### 3.2 Gas emissions during composting

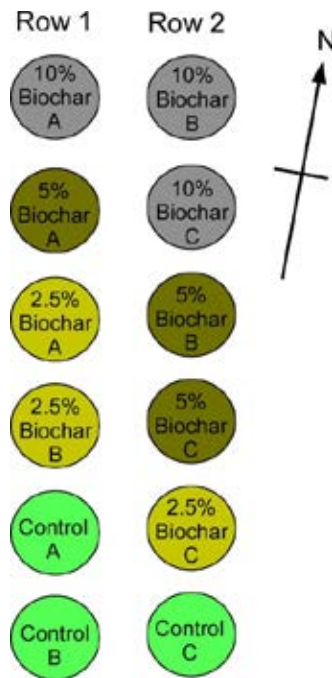
To test the effect of the same biochar (Oregon Biochar Solutions) on compost production from different components by different procedures, (co-)composting was performed at two different sites in 2018: at Lenz Enterprises in Stanwood, Washington, and at the Washington State University (WSU) composting facility in Pullman, Washington. Composting at Lenz Enterprises in Stanwood, Washington was carried out with one control and one experimental pile containing 5% biochar by volume. Composting at WSU was carried out with three different levels of biochar admixture, and three piles were made for each level. Gas emissions were monitored during the composting process.

### 3.2.1 Sampling methodology

Two large (200 yd<sup>3</sup>: approximately 22 feet wide by 17 feet long by ~ 14 feet high) aerated static piles were sampled at Lenz: a control pile and a pile with 5% biochar (Oregon Biochar Solutions). Air samples were collected on days 3, 7, 11, 20, and 30 by personnel from Washington Department of Ecology and WSU Puyallup. On each sampling day six samples were collected. Two samples were collected from the control pile from two different locations on the pile, two samples were collected from the biochar amended pile from two different locations on the pile, and a field blank was collected whereby the flux isolation chamber was placed on a clean tarp and samples collected as done with the pile samples. A duplicate sample was also collected from one of the piles whereby two canister samples for volatile organic compound analysis and two DNPH (2,4-Dinitrophenylhydrazine) cartridge samples for aldehyde analysis were collected at the same time from the flux isolation chamber. Canisters and aldehyde samples were shipped back to WSU via Fed Ex.

### 3.2.2 Composting experiments at WSU

In summer 2018, twelve static piles (10 yd<sup>3</sup>) were constructed at the WSU composting facility. Three different types of co-compost were produced using biochar from Oregon Biochar Systems. All twelve compost piles contained: 4.5 yards of screened manure solids from the Washington State University (WSU) dairy, 4.5 yards of dairy bedding straw and manure, 0.5 yards of ground clean green (woody) yard trimmings, and 0.5 yards of food waste from the WSU dining commons. These components were mixed and stacked in separate rows of six piles, each ~10 yd<sup>3</sup> in volume. Row 1 was made on 4 June and row 2 on 5 June 5, 2018, as illustrated in Figure 2. At least one of the piles from each replicate was built on a different day. Three of the piles contained biochar mixed at either 2.5%, three at 5%, and three at 10% by volume at the initiation of the composting process. Three piles contained no biochar (compost control). The piles were monitored for temperature. By the end of the composting period, the piles had reduced to about 5 yards each. Sampling of the piles was done by WSU personnel on pile age 3, 7, 10, 16, 22, and 31 days. Sampling included a field blank, and a duplicate sample, as was done at Lenz Enterprises. Sampling each row took a full working day using two flux isolation chambers (~42-L volume and 35-L volume with 0.13 m<sup>2</sup> sample area) supplied by Washington State Department of Ecology. Row 2 was sampled the day after row 1.



**Figure 2: Pile layout at WSU.**

As was done at Lenz, sampling included filling a canister for VOC analysis, the collection of a 30-L air sample on a DNPH-impregnated silica gel cartridge for aldehyde analysis, and a collection of an ammonia sample using coulometric Draeger tubes with ranges 1-30 ppmv (parts per million by volume), 5-100 ppmv, and 25-500 ppmv. Ammonia concentrations were often off-scale and so there was little usable ammonia data from this experiment. Local meteorological data (pressure, wind speed, temperature, humidity) was collected with a handheld anemometer (Kestrel). Temperatures inside the flux chambers (air temperature and pile surface temperature) and outside the chambers were measured using fine wire type K thermocouples. Interior pile temperatures were also recorded with a thermocouple that could be inserted 3 feet into the pile. Photographs of the sampling set-up on one of the piles is shown in Figure 3.



**Figure 3: Photograph of flux isolation chamber at top of the 5% biochar pile B in row 2. View is looking to the southeast.**

### 3.2.3 Flux isolation chamber

Sample collection used Washington State Department of Ecology's flux isolation chambers to collect gaseous emissions emitted from the compost pile ridge. This is a US EPA standard method for determining gaseous emission from ground surfaces (Klenbusch, 1986). Flux isolation chambers have been widely used for measurement of volatile gases from wide variety of sources including compost piles (Hellebrand and Kalk, 2001a; Sommer et al., 2004; Mukhtar et al., 2009; Parker et al., 2013). There is also a considerable number of studies focused on evaluation of performance and accuracy of this method (V. Blanes-Vidal et al., 2007; Acevedo Perez et al., 2009).

A standard flux isolation chamber is a 16-inch diameter stainless-steel cylinder covered with a clear Plexiglas dome with a 0.75-inch diameter opening at the top. The dome has a number of ports for attaching a supply air flow and ports for air sampling from the chamber. The chamber isolates a 0.13 m<sup>2</sup> ground area. The supply air for the chambers contained ~10% helium (He; Oxarc Specialty Gases) and the air collected from the chambers was analyzed for helium at WSU by the Analytical Chemistry Service Center. A small battery-operated fan was clipped to the inside of the flux chamber. This fan mixed the air inside of the chamber to distribute the air / He mixture uniformly. The supply air was allowed to flow at 5 L min<sup>-1</sup> for ½ hour before sampling began to allow surface emissions to reach steady state concentration inside the chamber. Sample lines for VOC sample collection consisted of ¼-inch PFA tubing and were inserted into the opening of the flux chamber to collect sample air into evacuated 6-L SUMMA canisters. The evacuated canister pulled air from the chamber at slow air flow rate so that canisters filled over the course of ~ ½ hour. This was done by slightly opening the canister valve and it was operator judgment, as the flow was not measured, on how fast the canister filled. Similarly, a Tygon tubing line was used for collection of air samples onto DNPH-impregnated silica gel tubes for aldehyde collection. Air flow rates for aldehyde sampling were set at 1 SLPM (Defender 510 portable air sampling pump) for 30 minutes, yielding a total sample volume of 30-L.

The residence time,  $\tau$ , is defined as the volume of the flux chamber divided by the supply air flow rate. Typically, after three to four residence times (~ ½ hour), concentrations inside the chamber are in steady state with respect to emissions and the supply air flow rate, and samples can then be collected. From the measured concentration in the chamber, the flux density can be determined as (EPA 1986):

$$J = \frac{Q C_{air}}{A} \times \frac{10\% He}{measured He} \quad (\text{eq. 1})$$

where  $J$  is flux is the flux density ( $\mu\text{g m}^{-2} \text{min}^{-1}$ ),  $A$  is area of emission source inside chamber (= 0.13 m<sup>2</sup>),  $Q$  is sweep air flow rate (= 0.005 m<sup>3</sup> min<sup>-1</sup>),  $C_{air}$  is the analyte concentration in the chamber ( $\mu\text{g m}^{-3}$ ). The flux density was corrected for the measured helium (He) concentration. Often low He concentrations were measured (< 2%) indicating extra dilution of the chamber. This could result from wind ventilating the chamber through the opening in the dome. We also determined that a gap between the Plexiglas dome and the stainless-steel ring allowed for unwanted air exchange. This gap was sealed for the WSU experiment using a Viton O-ring.

### 3.2.4 VOC analysis of canisters

Volatile organic compound samples were collected into evacuated 6-L SUMMA canisters provided by WSU. Canisters were cleaned using an Entech Model 3100D canister cleaning system by repeated cycles of pressuring with humidified UHP N<sub>2</sub> to 10 psig then evacuating to 1 Torr. The purging cycles were then followed a final heating and evacuation whereby the canisters were heated to 75°C and evacuated to 3 mTorr pressure for 2 hours. This cleaning procedure is the standard procedure adopted by our lab. Canisters were sent to the field evacuated with chain of custody forms to be filled out by the field sampling operator and returned with the canister samples.

Canister samples were analyzed at WSU following EPA method TO-15. A 50 mL sample was preconcentrated using an Entech 7200 preconcentrator and analyzed using gas chromatography-mass spectrometry (GC-MS; Agilent 7980/ 5997A). Separation was performed on an Agilent 624 column (60 m, 0.320 mm, film thickness 1.80 μm). The mass spectrometer was operated in scan mode from  $m/z$  33 to  $m/z$  206. The GC-MS response factors were determined from several gas standards. Monoterpene response factors were determined for seven compounds: α-pinene, β-pinene, camphene, limonene, 3-carene, α-phellandrene, terpinolene (Apel-Reimer Environmental, accuracy ±5%). Response factors for 47 C<sub>5</sub>-C<sub>12</sub> hydrocarbons (alkenes, alkanes, aromatics) were determined using a photochemical assessment monitoring mix (Linde Spectra Gases, accuracy ±10%). Response factors were also determined for 19 chlorinated volatile organic air toxic compounds from the EPA method TO-14A list (Linde Spectra Gases, accuracy ±10%). Compound identification was done with both retention time matching and mass spectrum matching. Identification of compounds not in the calibration standards was determined using the National Institutes of Standards and Technology (NIST) Mass Spectral Library. The analysis of the canisters required a daily sample blank, consisting of a canister filled with ultra-high purity nitrogen gas (N<sub>2</sub>), and a replicate run of one of the samples for quality assurance purposes.

### 3.2.5 Laboratory composting in large tanks

Emissions were also measured from composting in two large 100 gallon high density polypropylene tanks, with one compost tank having 10% biochar by volume (Oregon Biochar Solutions) and the other tank being a control. In this experiment continuous measurements were performed (1-minute time resolution) for VOCs, ammonia (NH<sub>3</sub>), methane (CH<sub>4</sub>), nitrous oxide (N<sub>2</sub>O), nitrogen oxides (NO<sub>x</sub>), and carbon dioxide (CO<sub>2</sub>) to determine if biochar-amended compost feedstocks had reduced emissions compared to feedstocks without biochar. Compost was obtained from the WSU compost facility. Two trials were done. The first trial of the composting lab test was conducted from 27 March to 8 April 2019. In the first experiment, one tank was filled with 81 gallons compost feedstocks manually mixed with 9 gallons biochar (10 vol. % biochar tank) and the other had 90 gallons compost feedstocks (control tank). This batch of feedstocks had less manure in it and no food waste compared to a typical mix at WSU. For the second trial, a more typical mixture was used that contained a mix of food waste, green waste, and manure. The second trial went from 24 April to 6 May 2019 and both tanks were filled with the same amount of total material.



Figure 4 shows a photograph of the tanks and Figure 5 shows a schematic diagram of the detailed setup for the tanks. The tanks were wrapped with an R3 reflective insulating foil. The tanks were filled to about the 90 gallon mark with compost or compost-biochar mixtures and sealed with a lid. At the bottom of the tank there was a 1-inch air gap made by a fine wire screen. Forced aeration was provided by sending compressed air through a tube to the air gap at the bottom of the tank at  $13 \text{ L min}^{-1}$ , allowing for air to diffuse through to the top of the material. In the first trial, both tanks were aerated at a rate of 20 seconds per hour for five days. For the second trial, tanks were aerated at  $13 \text{ L min}^{-1}$  for 4 minutes, 3 times per hour. This aeration rate was maintained for the first 3 days, then changed to so that air was added every 150 minutes for 4 minutes at  $13 \text{ L min}^{-1}$ . During trial 2 both compost tanks became dry and so after day 6, 12 L of distilled water was added to each tank. Compost temperature was measured by a thermocouple inserted into the middle of the material.

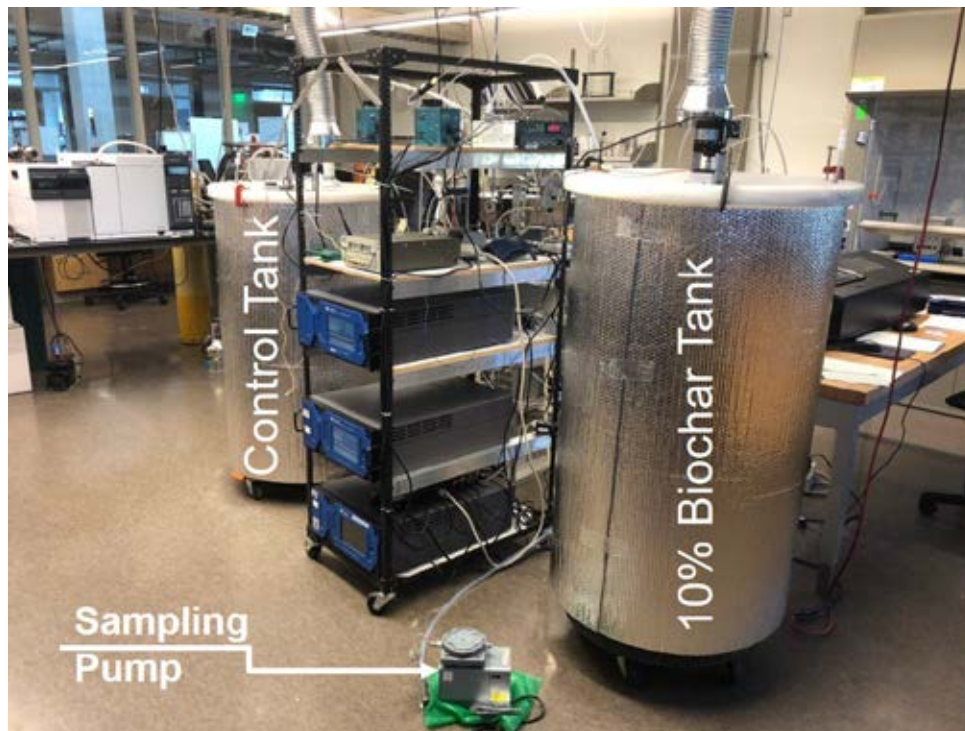
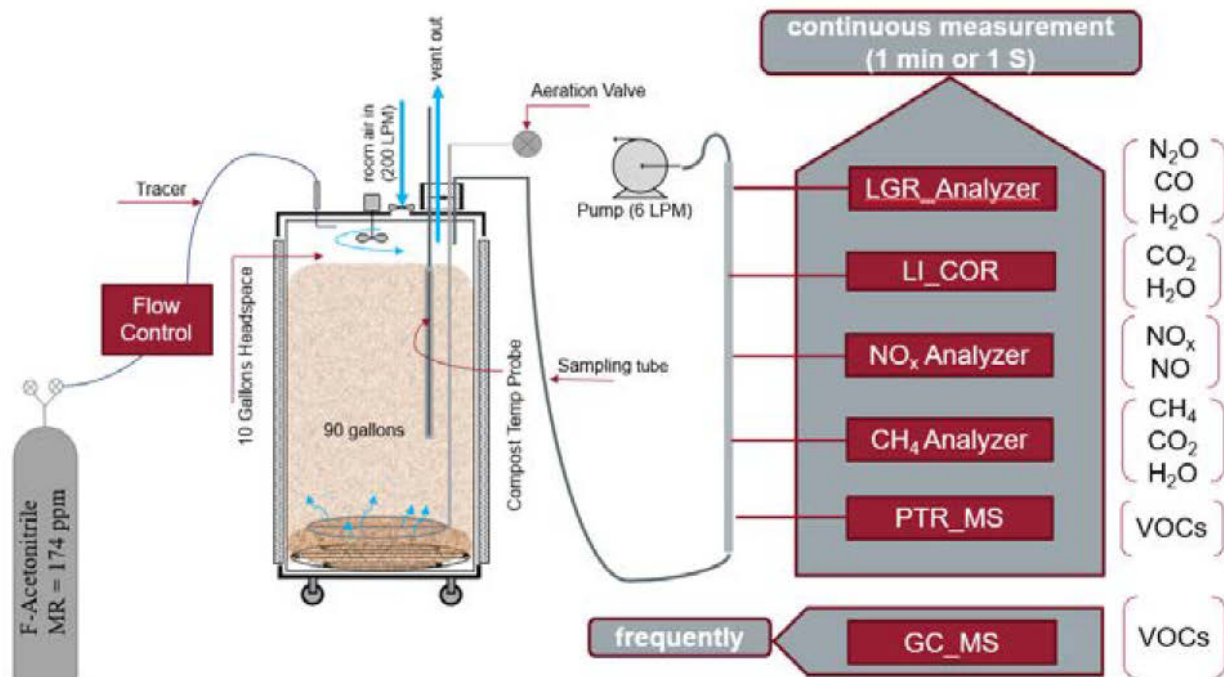


Figure 4: Photograph of compost lab test setup.



**Figure 5: Schematic diagram of one tank setup.**

Dilution air flow was provided by a small 12-volt fan attached to the tank lid. The fan blew room air into the tank at  $220 \text{ L min}^{-1}$  and the air flow was vented out a 1-inch diameter pipe attached to the lid. The vented emissions were captured by a laboratory exhaust snorkel vent that hung above this vent. A mixing fan stirred the head space of the tank ( $\sim 50\text{-L}$ ) to ensure well-mixed conditions. Given the headspace volume and dilution air flow rate, the air change rate was  $5.3 \text{ min}^{-1}$  (5.3 head space volumes exchanged per minute) compared to the much lower flux isolation chamber air change rate of  $0.14 \text{ min}^{-1}$ . The higher air change rate ensured that the tanks rapidly come to steady state and the compost emissions are well diluted. Low air exchange rate of enclosed chamber allowed buildup of the concentration of VOCs in the headspace, which resulted in a decreasing of the diffusion rate of VOCs from soil and, thus, an underestimation of surface fluxes (Sánchez et al., 2015). It was possible to reduce the dilution air flow through the tank by decreasing the supply voltage to the fan.

A tracer gas flow was added to the headspace of the chamber, as was done with flux isolation chamber sampling. The tracer used in these experiments was a mixture of flouroacetonitrile (174 ppmv) and fluorobenzene (180 ppmv) in compressed air. This mixture was added to the tanks at different flow rates using mass flow controllers (MFC), 11 standard cubic centimeters per minute (sccm) for the control tank, and 25 sccm for the biochar tank, to distinguish the data from each tank. Tracer gas concentrations were measured using a proton transfer reaction mass spectrometer (PTR-MS), the same instrument used to continuously measure VOC emissions from the tanks. By continuously measuring the trace gas concentrations, dilution air flow rate variation through the tanks could be monitored. We observed this flow rate variation to be small, on order

of 10%, likely a result of room air pressure variation. The dilution air flow rate was calculated from the measured tracer concentration:

$$\text{dilution air flow rate} = \frac{(\text{mixing ratio of tracer in cylinder}) * (\text{MFC Flow})}{(\text{mixing ratio of tracer in tank})} \quad (\text{eq. 2})$$

Then flux density for each compound was calculated as:

$$\text{flux density } (\mu\text{g m}^{-3} \text{ min}^{-1}) = \frac{\text{concentration } (\mu\text{g m}^{-3}) \times \text{tracer flow rate } (\text{m}^3 \text{ min}^{-1})}{\text{area } (0.342 \text{ m}^2)} \quad (\text{eq. 3})$$

To measure headspace concentration in the tanks, a Teflon tube inserted through the lid vent and a continuous flow of air was drawn from the headspace for the continuous emissions monitoring analyzers. Air was pulled from the tank using an external pump at 6 L min<sup>-1</sup> and analyzers subsampled from this flow from a common manifold made of PFA Teflon tubing and fittings. The gases measured were NO and NO<sub>2</sub> (Teledyne 200U NO<sub>x</sub> analyzer), N<sub>2</sub>O and CO (Los Gatos Research, DLT-100), CH<sub>4</sub> and CO<sub>2</sub> (Los Gatos Research, UPGA), CO<sub>2</sub> and H<sub>2</sub>O (Licor, 840A), and various VOCs and ammonia by PTR-MS (Ionicon Analytik). The tanks were sampled in an alternating fashion using 3-way PFA solenoid valves that automatically selected between the control tank, room, air, and the biochar tank. The sampling sequence was to measure the control tank for 1 hour, followed by room air measurements for 15 minutes, flowed by the biochar tank for 1 hour. This cycle repeated itself automatically. Gas emission data were continuously collected as 1-minute averages. GC-MS sampling was also done using a separate heated sampling line than was manually moved between the tanks.

### 3.3 Greenhouse and field trials

The sources of compost, co-compost and biochar for each of the experiments included in this study are shown in Table 6, as well as the dates during which the trials took place.

**Table 6: Summary of biochar and compost sources used in the experiments described in this report.**

Experiment	Dates(s) of trial	Biochar source	Compost source	Measurements
Emissions	Feb-Mar 2018 (Lenz) Jun-Jul 2018 (WSU)	OBS	WSU & Lenz (on-site)	Air emissions
Basil – field (Colbert)	Aug 2018	Amaron Energy	WSU 2017 Foothills	fresh plant mass phytochemical composition

Basil – greenhouse	Feb 2019 (both) Oct 2019 (Eleanora) Apr 2021 (TSQ)	Amaron Energy OBS	WSU 2017 WSU 2018 Lenz	fresh plant mass phytochemical composition
Strawberry – greenhouse	Aug-Oct 2018 Sep-Dec 2020	Amaron Energy OBS OBS	WSU 2017 WSU 2018 Puyallup 2019 Lenz	yield berry number single berry mass Trial 2020: phytochemical composition
Strawberry – field (Puyallup)	July-Oct 2018 May-Sep 2019	OBS	Lenz	yield soil properties
Potato – field (Mount Vernon)	June-Sep 2018 May-Sep 2019	OBS	Lenz	yield soil properties

### 3.3.1 Basil field trial

Sweet basil was grown in the field in this part of the project. One cultivar of sweet basil (*Ocimum basilicum* L. ‘Genovese’) was used in the field trial. The plants were grown at an organic farm, Footehills Farm in Colbert, Washington ([www.footehillsfarm.net](http://www.footehillsfarm.net)), which has been growing crops on site for many years with prior application annually of compost produced on-site. The plots used for the trial were in the middle of the field surrounded by other plants to avoid edge effects. As each basil seedling was transferred to the field, a hole of approximately 3 liters (~20 cm deep) was dug by hand with a small trowel, with approximately 500 ml of compost or co-composted biochar (of one of the various treatments) mixed in to the loose soil in the hole, and the seedling then placed in the hole. Total percent compost added to each hole was therefore approximately 17% of total volume.

Overall, control plants grown in soil without any amendment were compared to six treatments: plants grown with 1) WSU 2017 compost without biochar, 2) Footehills compost without biochar, 3) WSU 2017 compost co-composted with 2.5% biochar (total in co-compost mix by volume), 4) WSU 2017 compost co-composted with 5% biochar (total in co-compost mix by volume), 5) WSU 2017 compost mixed plus 2.5% biochar (total in co-compost mix by volume), or 6) WSU 2017 compost plus with 5% biochar (total in co-compost mix by volume). Plants were harvested on 17 August 2018, about three months after planting, when they reached the typical size harvested by the grower (Thom Foote from Footehills Farm), this was typically just as inflorescences started to develop. Each plant was cut off at the base, weighed immediately due to concerns about loss of mass due to wilting after harvest, photographed, and the two youngest fully mature leaf pairs were collected and frozen on dry ice for chemical analysis.

### 3.3.2 Basil greenhouse trial

All greenhouse experiments, with basil and strawberries, used the same soil mix recipe as in the earlier study (Gang et al., 2018), which consisted of 37.5% potting soil (Sunshine #4, Sun Gro Horticulture, Agawam, MA), 12.5% perlite, 25% field soil (collected from a field at HerbCo International’s main site in Duvall, WA), and 25% compost (with or without biochar) (all percentages by volume). For all greenhouse experiments, the pots for different treatments were placed randomly within the area used for the experiment and rotated on a regular basis to avoid edge effects.

For the basil greenhouse experiment, the same two sweet basil cultivars (Eleanora and Thai Siam Queen, abbreviated TSQ) as grown for the first study (Gang et al., 2018) were included in this investigation. The experiment took place between 28 November 2018 (seeds planted) and 4 February 2019 (harvest). Ten plants were grown for each of the 13 treatments, with three different sources of composts described above. The treatments are summarized in Table 7.

As in the earlier study (Gang et al., 2018), addition of biochar after composting was based on the initial volume of compost to mimic the proportions during co-composting.

Plants were grown as described earlier (Gang et al., 2018) except for the early stage which occurred as follows. The seeds were germinated in peat pellets (4 seedlings/pellet) and thinned after germination to 1 seedling/pellet before being transplanted into soil. Until the transplant they stayed in a growth chamber kept at 27/22°C day/night, and 12/12hour light cycle, and 80% relative humidity for 26 days. After transplant to larger pots, the plants were placed in the greenhouse and grown with supplemental lighting and under the conditions previously described for an additional 40 days of growth, at which time they were photographed and harvested on day 66 after planting the seeds. At harvest, each plant was cut off at base (about 1.5-2 cm from soil), fresh weight recorded, and leaf material collected and stored as described earlier (Gang et al., 2018).

**Table 7: Summary of treatments used in the basil greenhouse experiment.**

Treatment #	Compost	Biochar	%	Co-composted or biochar+compost
1	WSU 2017	None	0.0	n/a
2	WSU 2017	Amaron Energy	2.5	Co-compost
3	WSU 2017	Amaron Energy	5.0	Co-compost
4	WSU 2018	None	0.0	n/a
5	WSU 2018	OBS	2.5	Co-compost
6	WSU 2018	OBS	5.0	Co-compost
7	WSU 2018	OBS	10.0	Co-compost
8	WSU 2018	OBS	2.5	Biochar+compost
9	WSU 2018	OBS	5.0	Biochar+compost
10	WSU 2018	OBS	10.0	Biochar+compost

11	Lenz	None	0.0	n/a
12	Lenz	OBS	5.0	Co-compost
13	Lenz	OBS	5.0	Biochar+compost

### 3.3.3 Strawberry greenhouse trial

Bare root strawberry plants (day-neutral *Fragaria x ananassa* ‘Albion’) were purchased from Lassen Canyon nursery (Redding, CA). The plants were potted on 27 June 2018. All fruit, flowers, and runners were removed at transplant and for two weeks thereafter to promote root and leaf growth. The plants were grown in the greenhouse under ambient lighting which was supplemented by high-sodium pressure lights activating if light intensity dropped below 200  $\mu\text{mol} \times \text{m}^{-2} \times \text{s}^{-1}$ . Day temperature was kept at 21-24°C, and night temperature at 17-20°C. One half of the plants received general purpose 20-10-20 Peters Professional Liquid Feed fertilizer with chelated iron (Sprint 330), Epsom salts, and micronutrients 2-3 days per week at 200 ppm.

There was a total of 8 treatments, with 10 plants per treatment. Five treatments were based on WSU 2017 compost: 1) compost without biochar amendment, 2) co-composted with 2.5% Amaron Energy biochar, 3) co-composted with 5% Amaron Energy biochar, 4) compost plus 2.5% Amaron Energy biochar and 5) compost plus 5% Amaron Energy biochar. The other 3 treatments were based on Lenz compost: 6) compost without biochar, 7) co-composted with 5% Oregon Biochar Solutions char or 8) compost plus 10% Oregon Biochar Solutions biochar (mismatched biochar concentrations between treatments 7 and 8 are due to a miscommunication). Fruit was collected from each plant as it ripened, in total there were 22 harvests between 4 August 2018 and 31 October 2018. At each harvest, total weight of fruit and number of berries was recorded. In 2020, ten plants for each of the eleven treatments were potted on July 27, and strawberries were harvested in a total of 16 harvests between September 7 and December 14. The treatments differed from the trial in 2018 due to compost availability. Five treatments were based on WSU 2018 compost: 1) compost without biochar amendment, 2) co-composted with 5% OBS biochar, 3) co-composted with 10% OBS biochar, 4) and 5) compost plus 5% and 10% OBS biochar, respectively. Three treatments were based on Puyallup compost: treatment 6) without biochar, 7) co-composted with 40% OBS biochar, and 8) Puyallup compost plus 40% OBS biochar. Treatments 9-11 with Lenz compost were identical to treatments 6-8 in the 2018 trial. No WSU 2017 compost was available.

### 3.3.4 Basil and strawberry metabolite analysis

Extraction and analysis of non-volatile phytochemicals from sweet basil leaves (with a focus on major phenolic antioxidants) by liquid chromatography–mass spectrometry (LC-MS) was carried out as described previously (Gang et al., 2018) with the modification that methanol was used as extraction solvent instead of 85% aqueous ethanol. In addition, ultrasonication time was shortened from 30 to 15 minutes for the greenhouse grown Eleanora basil material. Those extracts were incubated on an orbital shaker for 60 minutes at room temperature after the ultrasonication instead. Extraction and analysis of sweet basil volatile aroma compounds followed the procedure described previously (Gang et al., 2018).

Metabolite analysis from strawberries largely followed procedures described by Fait et al. (2008). Fruit tissue was lyophilized for 48h and ground in a ball mill prior to extraction. Approximately 20mg of dry tissue were used for extraction of metabolites for LC-MS using 800 $\mu$ L 80% aqueous methanol supplemented with 10 $\mu$ M 6-chloro-4-hydroxycoumarin, or of primary metabolites, using 1000 $\mu$ L methanol:2-propanol:water (5:2:2, v/v/v) mixture supplemented with 10 $\mu$ L of 2mg/mL ribitol. For the analysis of total sugar content, 10 mg dry powder was suspended in a defined volume of water, vigorously shaken for 5 min, the debris removed by centrifugation for 5 min at 21,000g, and the clear supernatants analyzed using Atago 3810 PAL-1 digital hand held refractometer.

### 3.3.5 Strawberry field trials

Research plots for the potato and strawberry field trials were established at the Mt.Vernon and Puyallup Research and Extension Centers (Washington State University) in consecutive years, 2018 and 2019 to evaluate responses in soil and plant biomass following amendment and one growing season. Additionally, strawberry yields, and only strawberry yields, were collected into a second growing season (from the same plots) for each first year, 2018 and 2019.

#### *Experimental design*

Research plots were installed at Washington State University’s Puyallup Research and Extension Center (Goss Farm) and planted to strawberry (*Fragaria x ananassa* Duch. ‘Albion’).

Strawberry growers amend their soils with supplementary nitrogen in the form of fertilizer. Therefore, we included an additional component in the experimental design (i.e., fertilizer) allowing us to provide a comprehensive analysis of two management strategies on which realistic expectations and grower recommendations can be based.

Including an unamended control, five treatments: biochar alone, compost alone, co-compost and a field-mixed biochar plus compost blend, were surface amended in a randomized split-plot design which was replicated four times at each location. At both sites, the main plot factor was nitrogen fertilizer (i.e., fertilized or unfertilized) and the sub-plot factor was amendment (e.g., biochar). Amendments were applied to meet a target rate of 8.75 Mg ha<sup>-1</sup> organic carbon (C) and are listed in Table 8. However, in 2018, due to changes in biochar moisture content, we discovered that the target rate for the biochar was less than anticipated. To equalize treatments in 2018 and 2019, we amended biochar at the rate listed in table 8, instead of the original 8.75 Mg ha<sup>-1</sup> C target.

**Table 8: Application rates for each of four soil amendments applied to research plots in Puyallup and Mount Vernon, Washington in 2018 and 2019.**

<b>Amendment</b>	<b>Dry Mg ha<sup>-1</sup></b>	<b>C Mg ha<sup>-1</sup></b>	<b>m<sup>3</sup> ha<sup>-1</sup></b>	<b>C:N ratio</b>
Biochar (OBS)	8.8	7.65	40.2	112
Compost (Lenz)	27.3	8.75	54.9	15
Co-compost	27.4	8.75	54.9	15
Biochar + compost	18.7 (4.4 + 13.7)	8.21	47.6	26

### *Field trial management*

Following amendment, soils were tilled to a depth of 15 cm and then prepared for planting. Raised bed research plots were covered with black polyethylene and green-house grown bareroot plugs were planted, by hand, 10 July 2018 and 19 July 2019 in four rows per 11.1 m<sup>2</sup> plot sub-plot and immediately irrigated. Calcium nitrate (15.5-0-0) was applied by fertigation to fertilized plots in weekly applications of 2 kg N ha<sup>-1</sup>, totaling 20 kg N ha<sup>-1</sup>. All plots received an application of Malathion at 295.7 g ai ha<sup>-1</sup>. Irrigation was applied as necessary to research plots utilizing drip irrigation.

Strawberry fruit was harvested and weighed weekly from 10 August 2018 until 3 October 2018 (first establishment, first collection); from 22 May 2019 until 4 September 2019 (first establishment, second collection), and from 20 May 2020 until 3 September 2020 (second establishment, first collection). Cumulative yield is presented as the total of all harvests for a given season.

### **3.3.6 Potato field trial**

#### *Experimental design*

Research plots were installed at the Mount Vernon Northwest Washington Research and Extension Center and planted to potato (*Solanum tuberosum* L. 'Chieftain'). The experimental design and treatments were as described for strawberry field trials (above). As with the strawberry field trial, fertilizer was included as an additional component of the potato field trial.

#### *Field trial management*

Following amendment, soils were tilled to a depth of 15 cm and then prepared for planting. Using a two-row planter, potatoes were planted to a depth of 15 cm on 7 June 2018 and 31 May 2019 and hilled 2 July 2018 and 5 July 2019 in four rows per 23.6 m<sup>2</sup> sub-plot. In 2018 and 2019, all plots received herbicide applications of Outlook (dimethenamid) at 941 g ai ha<sup>-1</sup> and Tricor 75 DF (metribuzin) at 561 g ai ha<sup>-1</sup>. The following supplementary macro- and micronutrient applications were made in 2018: muriate of potash (0-0-60) at 111 kg ha<sup>-1</sup>, boron 15% granular at 5.6 kg ha<sup>-1</sup>, and procote BMZ at 111.2 kg ha<sup>-1</sup>, and in 2019: muriate of potash (0-0-60) at 246 kg ha<sup>-1</sup>, 0-0-22 at 303.7 kg ha<sup>-1</sup>, and 15% boron in solution at 4.4 kg ha<sup>-1</sup>. In 2018, unfertilized plots received 0-45-0 fertilizer at 77.34 kg ha<sup>-1</sup> and in 2019 they received 0-52-0 fertilizer at 394.5 kg ha<sup>-1</sup>. Fertilized plots were amended with the nutrients and rates listed in Table 9, which totaled 108.6 and 105.3 kg N ha<sup>-1</sup> for 2018 and 2019, respectively. In 2019, plots received an additional fungicide application of chlorothalonil at 1.72 L ha<sup>-1</sup> and mancozeb at 1.85 L ha<sup>-1</sup>. Irrigation was applied as necessary to research plots utilizing overhead irrigation. Weed cover was measured, once, on 30 July 2018.

Potatoes were harvested 10 September 2018 and 4 September 2019, and 3-plant subsamples (2 plot<sup>-1</sup>), including above and below ground biomass, were collected, separated by tuber and leaf tissue, and later weighed. Reported results are the average tuber weight per sub-plot.



**Table 9: Product, analysis, and rate for fertilized potato plots in Mount Vernon, Washington for 2018 and 2019.**

<b>Product and analysis</b>	<b>Rate (kg ha<sup>-1</sup>)</b>	<b>Year</b>
Ammonium sulfate (20.5-0-0)	89.67	2018
Ammonium sulfate (20-0-0)	49.31	2019
Monoammonium phosphate (11-52-0)	67.25, 394.4	2018, 2019
Urea (46-0-0)	196, 98.6	2018, 2019
Ammonium phosphate (10-34-60)	67.25	2018, 2019

### **3.4 Soil physicochemical properties**

From research plots in Puyallup and Mount Vernon and following amendment, soils were sampled at three separate times: 14 days and 44 days post-amendment, and at the end of the growing season. Soil samples collected at each time period were analyzed for various soil physical and chemical properties listed below.

#### **3.4.1 Bulk density**

To assess the effects of amendment on soil physical properties, two bulk density cores (136.4 cm<sup>3</sup> each) were collected from each plot at 14 days post-amendment in 2018 and 2019, before either crop had considerably altered soil conditions. Bulk density cores were weighed and recorded, and then a 25g subsample was placed in pre-weighed tins, weighed again, and dried at 80°C in a Thermo Fisher Scientific Isotemp 650G oven (Waltham, Massachusetts) for 48 hours. Dried subsamples were re-weighed, which provided an estimate of soil moisture, and bulk density was calculated as the weight of dried soil (g) divided by soil volume (cm<sup>3</sup>).

#### **3.4.2 Total carbon and total nitrogen in whole soil and particulate organic matter fractions**

At the same 14 day interval in 2018 and 2019, additional soil cores (10-12 plot<sup>-1</sup>, 2.2 cm diameter) were collected to a depth of 15 cm from unfertilized plots and were used to evaluate the effects of amendment on soil total C and N in two different soil size fractions, whole soil (total) and particulate organic matter (POM). All soil samples were homogenized and sieved to 2mm from which a 10g subsample was collected. For whole soils, the 10g subsamples were bar ground for 24 hours using Qorpak 60 ml glass vials (Chicago, Illinois) and a Wheaton benchtop roller (Millville, New Jersey). Particulate organic matter was determined following a fractionation procedure described in Marriot and Wander (2006). Briefly, 10g soil subsamples were placed into 30 ml Nalgene plastic bottles (Waltham, Massachusetts) to which 30 ml of a 10% (w/v) solution of sodium hexametaphosphate was added. The top of the Nalgene screw tops had been cut and removed so that a 53 µm screen could be placed between the bottle and screw top allowing restricted flow into and out of the bottle. Eight of the Nalgene bottles were placed into a 1-quart Ziploc freezer bag with an additional 400 ml of the sodium hexametaphosphate solution and allowed to rest overnight. The following day, the overnight solution was drained from the Ziploc bag, replaced with 400 ml distilled water and agitated on an Eberbach fixed speed shaker (Belleville, Michigan) for 1 hour. The bag was drained, refilled with 400 ml clean

distilled water and agitated for an additional 10 minutes, and this procedure was repeated 10 times or until the Ziploc bag solution was clear. The Nalgene bottles were removed from the bag and each top was carefully unscrewed, making sure not to lose any material. To collect the fine particles and using distilled water, the filter, lid and bottle were backwashed into stainless steel bowls. Samples were then placed in an oven, dried at 50°C for 48 hours, weighed, ground, and both the whole soil and POM fraction were analyzed for total C and N via the combustion method. This entire process was repeated on an additional set of soil samples collected at the end of the growing season from each cropping system in 2018 and 2019.

### **3.4.3 Mid-season inorganic nitrogen**

At 44 days in 2018 and 2019, soil cores (10-12 plot<sup>-1</sup>, 2.2 cm diameter) were collected to a depth of 30 cm from unfertilized and fertilized plots and used to assess mid-season available inorganic N as affected by amendment and fertilizer. Inorganic N was determined spectrophotometrically following a 1 normal potassium chloride extraction and cadmium reduction.

### **3.4.4 Post-season soil chemical properties and soil macro- and micronutrients**

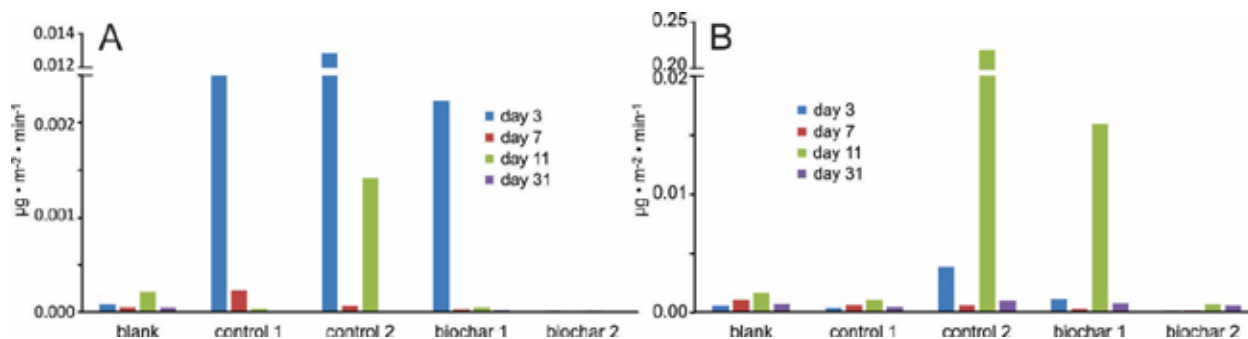
At the end of the growing season for 2018 and 2019, soil cores (10-12 plot<sup>-1</sup>, 2.2 cm diameter) were collected from unfertilized soils to a depth of 15 cm to understand the effects of amendment on certain soil chemical properties and soil macro- and micronutrients. Soil samples were air dried, ground, sieved to 2 mm, and analyzed for cation exchange capacity (CEC), nitrate-nitrogen (NO<sub>3</sub><sup>-</sup>-N), potassium (K), organic matter (OM %), calcium (Ca) magnesium (Mg), sodium (Na) sulfur (S), and zinc (Zn). Soil chemical properties and soil macro- and micronutrients were determined as follows: CEC values were calculated by ammonium replacement; OM was determined by the Walkley-Black method; NO<sub>3</sub>-N and NH<sub>4</sub>-N was calculated following 1 normal potassium chloride extraction and cadmium reduction; K, Ca, Mg, and Na by ammonium acetate extraction; S by calcium phosphate extraction and turbidimetric measurement; and Zn by diethylenetriaminepentaacetic acid (DTPA) extraction.

## 4. Results and discussion

### 4.1 Gas emissions during composting

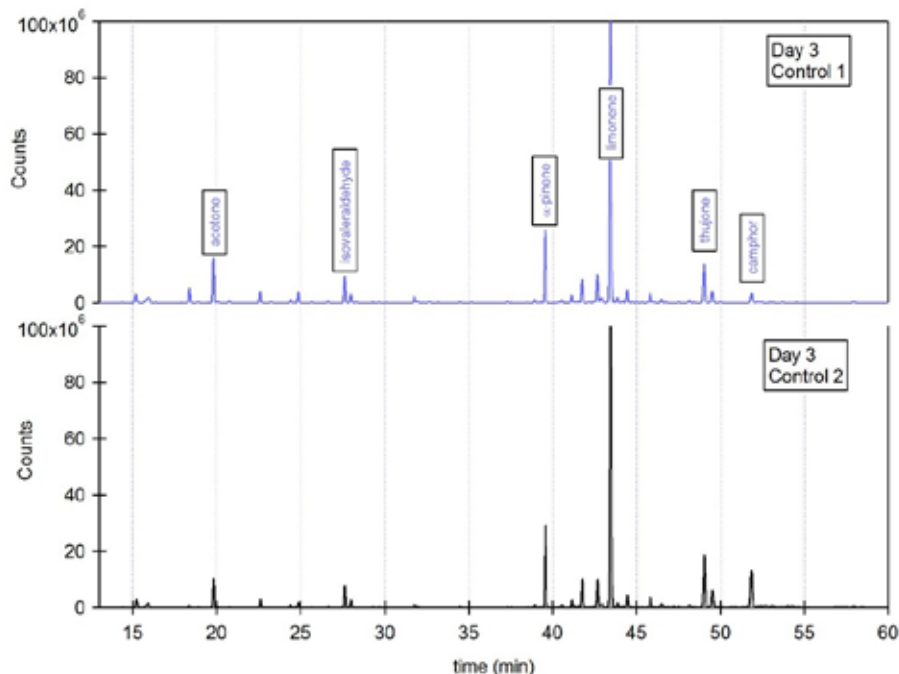
#### 4.1.1 Lenz composting experiment

Of the six carbonyl-group containing volatile compounds that were monitored, the most abundant ones were acetaldehyde and acetone. At Lenz Enterprises, two samples designated “1” and “2” were taken from both control (no biochar) and co-composted (5% biochar) pile on each sampling day. The emission of both carbonyl compounds was higher in the early (less than 14 days) composting stage. On several sampling days, the results from the two different samples varied strongly (Figure 6). Values for acetone were very disparate at the 3<sup>rd</sup> sampling time point, while those for acetaldehyde did so at the 1<sup>st</sup> sampling on day 3. Overall, the emission of both acetaldehyde and acetone was reduced by the addition of biochar, as observed in our previous study (Gang et al., 2018). The greater emission of volatiles at the early stage of composting is in agreement with previously published literature (Eitzer, 1995; He et al., 2012; Maulini-Duran et al., 2014; Sanchez-Monedero et al., 2018b).



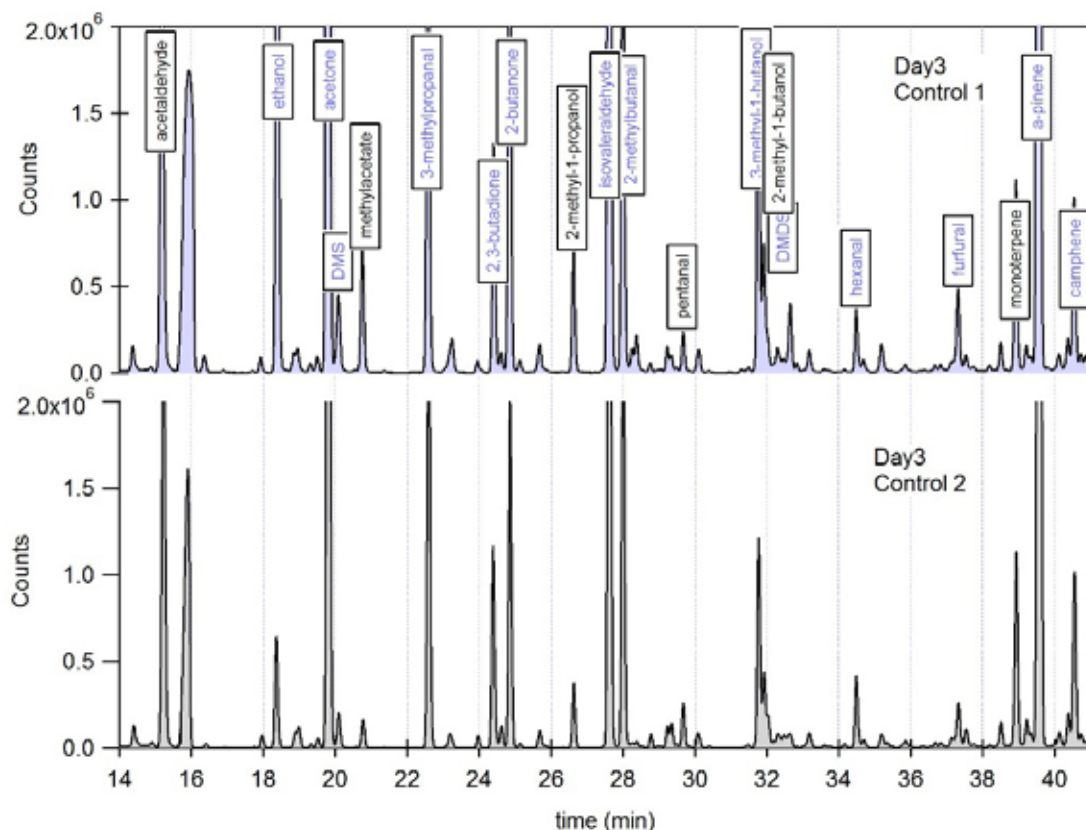
**Figure 6: Volatile organics emissions at Lenz Enterprises. A: acetaldehyde and B: acetone were collected on indicated days after piles were built. Two samples each were taken from control and biochar-amended pile on each sampling date. To demonstrate the data variance, all samples are plotted individually.**

Figure 7 shows example chromatograms from the two control piles samples taken on Day 3. Evident from the figure is that monoterpene compounds were the most abundant in these samples. This was generally true for all the samples collected at Lenz. For some samples, the 50-cc sample size was too large, and the most abundant monoterpenes produced a response that saturated the mass spectrometer preventing their quantitation. These samples had to be run again using a smaller sample volume.



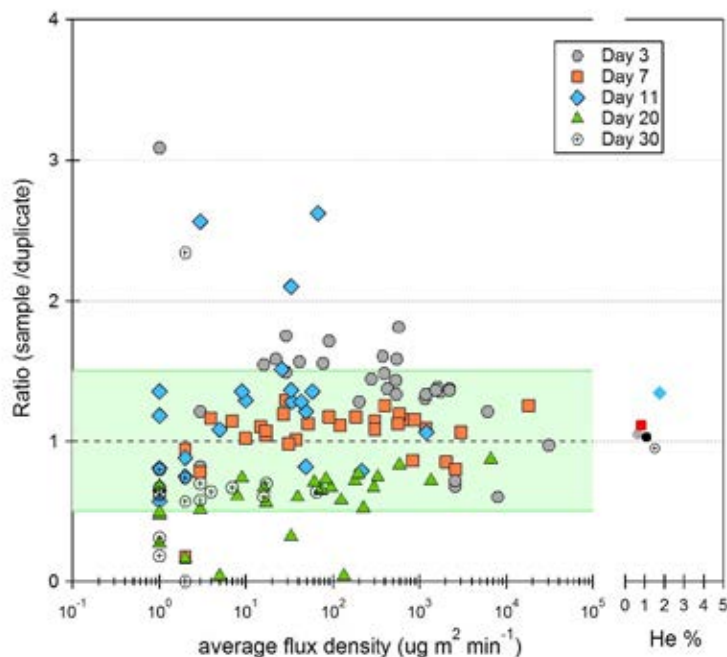
**Figure 7: Chromatogram showing total ion count rates for the two samples (Control 1 and Control 2) collected from the control pile on Day 3. Most abundant compounds are labelled in the top panel. The most abundant compound was limonene in both samples.**

**Error! Reference source not found.** Figure 8 shows the same chromatograms but the y-axis scale has been expanded by a factor of 50 to better see the numerous but lower concentration VOCs present in the samples. Over 100 peaks were identified by retention time and/or mass spectral matching. Many of these components were oxygenated compounds, principally aldehydes and ketones. For many of these compounds, we did not have calibration standards to derive response factors. For monoterpene type compounds (including camphor [C<sub>10</sub>H<sub>16</sub>O] and two related compounds identified as  $\alpha$ -thujone and  $\beta$ -thujone [C<sub>10</sub>H<sub>16</sub>O]), response factors were derived from response factors for the monoterpenes in our gas standard. For the abundant oxygenated compounds, and the sulfur compounds dimethyl sulfide (DMS) and dimethyl disulfide (DMDS), response factors were derived from hydrocarbon response factors in the gas standard. From experience we know the derived response factors work reasonably well for monoterpenes, but less well for the oxygenated compounds and sulfur compounds. The abundances of these compounds are much more uncertain (estimated factor 2 uncertainty). Acetaldehyde, though identifiable in the chromatogram was not quantified. Methanol, a compound likely emitted from compost emissions, was not detectable in the Lenz analysis because the mass spectrometer mass range was set above its molecular weight.



**Figure 8: Chromatogram showing total ion count rates for the two samples (Control 1 and Control 2) collected from the control pile on Day 3. Y-axis scale has changed by a factor of 50 from figure 1 to zoom in on the numerous compounds present at low concentrations in the first 40 minutes of the chromatogram.**

The level of agreement of the duplicate samples is illustrated in Figure 9. Shown is the ratio of the compound flux density determined from the two canisters samples versus their average flux density. Also shown are the results for the helium analysis on a separate scale. The level of agreement for VOCs was typically better than 50% over a wide range of flux densities. For some compounds in some samples, the agreement was much worse than 50% and could be a result of compound losses to canister surfaces. Some of the canisters contained liquid water in them as a result of the high humidity conditions inside the flux chambers. Liquid water in the canisters is a problem for sample storage for water soluble polar compounds like alcohols and aldehydes. Overall, we conclude that a level of agreement of 50% between duplicates, attributed to sampling reproducibility and GC-MS analysis reproducibility, is what can be expected, so differences much greater than this are required to demonstrate impact of biochar.



**Figure 9: Level of agreement of the duplicate sample versus the compounds flux density. Each data point is a particular compound. Shading shows  $\pm 50\%$  about the 1:1 line, Level of agreement of the duplicate sample for most compounds was in general within 50% of the primary sample. Flux densities varied over 4 orders of magnitude. Also shown is the level of agreement of the helium analysis versus helium concentration measured in the sample canister. Expected level of helium is 10%.**

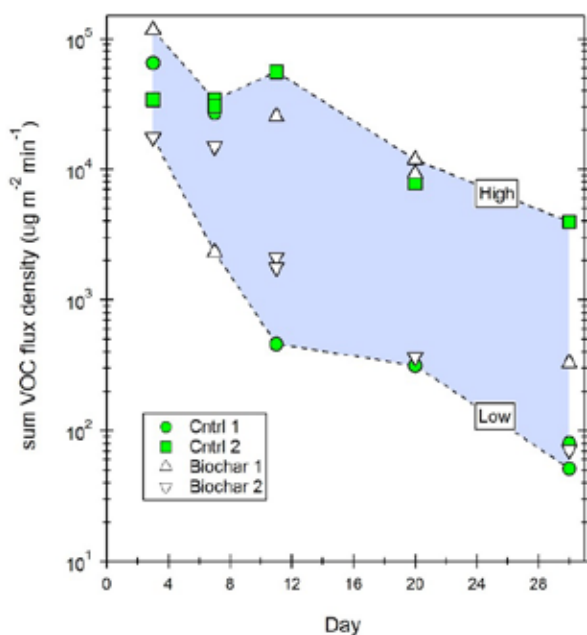
The sum of the flux densities for the 30 most abundant VOCs in the collected samples are shown in Table 10. The table also shows the fraction of the VOC flux density attributed to monoterpene compounds; this was often greater than 90%. Of the monoterpenes,  $\alpha$ -pinene and limonene were typically the most abundant. The largest flux densities were measured on days 3 and 7 and dropped off as the pile aged. The table also shows the He concentration (in %) measured in the canisters, and these were often much less than the expected 10% concentration of the supply air flow. These low values illustrate large extraneous dilutions of the chamber had occurred, perhaps caused by the positive and negative air flows created by the mechanical ventilation system of the aerated piles. An inspection of the data in Table 10 reveals that large flux density differences were observed between the two samples collected from the control pile and the 10% biochar pile. For example, on day 3, the VOC flux densities for the biochar samples differed by a factor of 10 (biochar 1 sample =  $116,493 \mu\text{g m}^{-2} \text{min}^{-1}$  compared to biochar 2 sample =  $17,571 \mu\text{g m}^{-2} \text{min}^{-1}$ ) while the He concentrations were similar. These large flux density differences could be caused by heterogeneity of surface emissions. It has been shown that hot spots exist on compost windrows and surface flux densities can differ by a factor of 30 (CARB, 2007). Another example is the factor of 120 difference measured for the control pile samples collected on day 11. Very low emission values may be the result of negative aeration flow, whereby air is being pulled into the pile, reduced diffusive emissions of VOCs from the pile surface.

**Table 10: Samples collected at Lenz with measured helium concentration, sum of reported VOC flux density, and percentage of flux density from monoterpenes.**

<b>Date</b>	<b>Sample Name</b>	<b>He (%)</b>	<b>VOC flux <math>\mu\text{g m}^{-2} \text{min}^{-1}</math></b>	<b>% monoterpenes</b>
Feb 26	Control 1	0.635	65,183	88.9
	Control 1 Duplicate	0.610	65,548	92.4
	Control 2	1.319	33,976	93.6
	<b>Day 3</b> Biochar 1	1.081	116,493	83.3
	Biochar 2	1.606	17,571	97.3
	Equipment Blank	4.910	6	95.6
Mar 2	Control 1	1.228	27,090	96.5
	Control 2	0.818	34,161	96.2
	Control 2 Duplicate	0.735	30,226	96.1
	<b>Day 7</b> Biochar 1	1.584	2,325	96.3
	Biochar 2	1.495	14,937	96.5
	Equipment Blank	5.910	12	90.7
Mar 6	Control 1	1.954	457	89.1
	Control 2	0.629	55,763	97.2
	Biochar 1	1.685	25,341	99.2
	<b>Day 11</b> Biochar 2	1.741	2,115	88.4
	Biochar 2 Duplicate	1.296	1,780	82.8
	Equipment Blank	1.493	615	79.2
Mar 15	Control 1	2.160	315	91.8
	Control 2	1.010	7,845	96.0
	Biochar 1	1.095	9,295	96.4
	<b>Day 20</b> Biochar 1 Duplicate	1.066	11,803	94.6
	Biochar 2	1.097	365	80.9
	Equipment Blank	5.115	6	95.6
Mar 26	Control 1	1.515	51	61.2
	Control 1 Duplicate	1.599	80	67.4
	Control 2	0.706	3,956	93.5
	<b>Day 30</b> Biochar 1	1.126	327	67.4
	Biochar 2	1.250	71	49.0
	Equipment Blank	3.813	1	59.8

The sum of the flux densities versus pile age are shown in Figure 10. The figure illustrates that overall flux densities decreased with pile age but there was a large amount of variability amongst the control and biochar samples. The biochar samples sometimes had the highest flux densities on a particular day and sometimes the lowest. The flux data are too variable to discern an impact of biochar. This variability may in part be due to the forced air flow through the piles by the

aeration system. The system either forced air through the pile (positive flow) or pulled air through the pile (negative flow) depending on measured temperature gradients in the pile. Positive air flow occurs roughly 50% of the time (Edward Wheeler pers. communication). Positive air flow may have impacted measured flux densities in two ways. Firstly, having additional air flow pushing through the compost pile into the flux chamber would cause dilution of the helium, imposing larger helium correction factors. Secondly, forcing air through the pile will sweep the pile of emissions and chamber VOC concentrations might be “artificially” larger as a result. We observed this to happen in our lab based composting experiments when we forced aeration flow through the compost. Thus, under positive flow the chamber concentrations will be higher and the helium correction factor larger, leading to a much larger flux than under neutral aeration air flow conditions. Under negative flow chamber VOC concentrations might be reduced as emissions are pulled from the near surface into the pile. This aeration flow variability may have contributed to the large variability in flux densities for a particular compost age and pile type. In Figure 10, the same proportion of samples define the “high” flux density boundary as the “low” flux density boundary, consistent with the roughly equal amounts of time the aerated static pile is under positive and negative aeration flows.

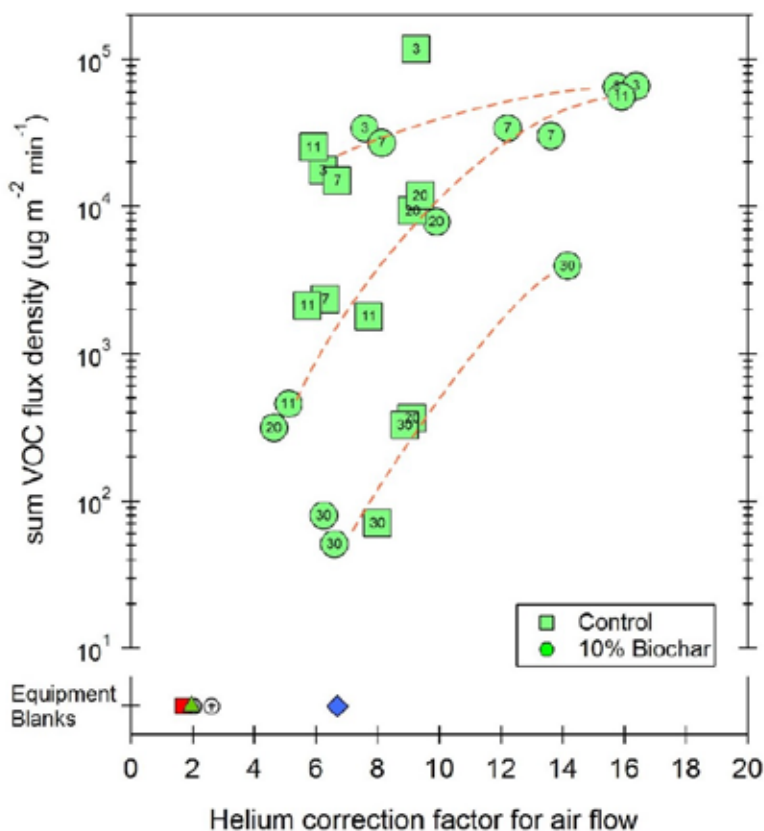


**Figure 10: Sum of the VOC flux density versus compost age for the Lenz aerated static piles. Shown are flux densities for the control pile (green colored symbols) and the 10% biochar pile (open triangles). A wide range of flux densities were measured; upper and lower bounds are defined by the shading.**

The VOC flux density did display a dependence on the helium correction factor, as illustrated in Figure 11. The six largest helium correction factors were all from the 10% biochar pile from days 3, 11, 7, and 30, and these samples had some of the largest flux densities. The largest flux densities for any particular day were associated with largest helium correction factors. For example, the day 30 biochar pile sample with the highest flux density ( $3,956 \mu\text{g m}^{-2} \text{min}^{-1}$ ) had a



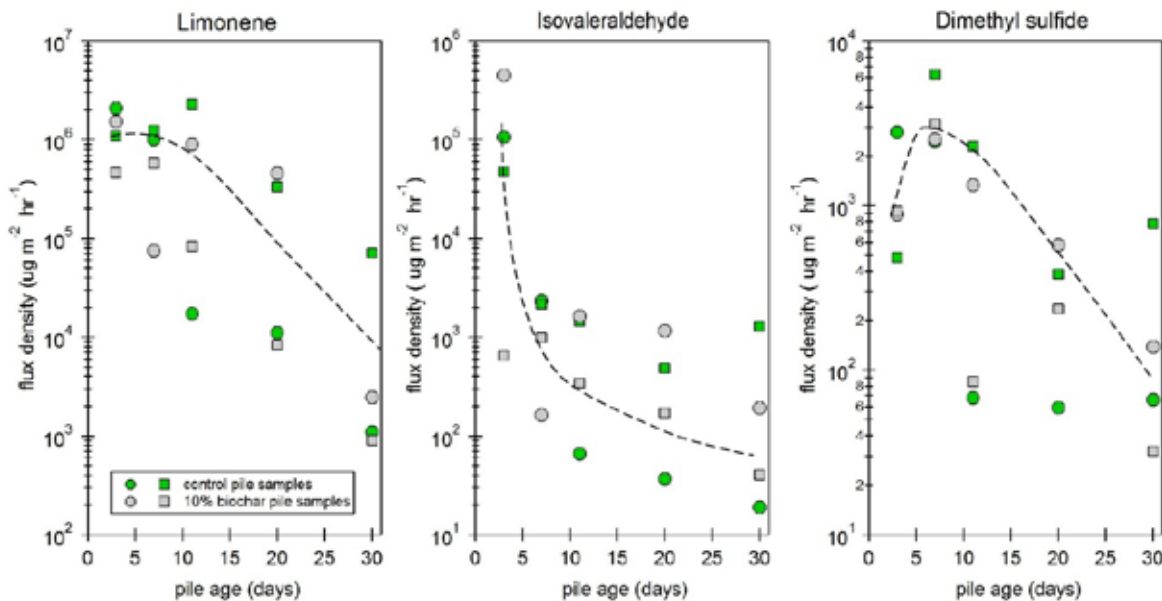
higher value than some samples collected at younger pile ages (days 7, 11, 20). This day 30 sample also had one of the highest helium correction factors, 14.2, and its flux density was more than a factor of 50 larger than the other two samples from day 30 biochar (sample + duplicate), with helium correction factors of  $\sim 6.5$ . Groups of data are apparent in the figure, as indicated by dashed lines, showing how older piles can emit at similar flux densities as younger piles, if, presumably, pile emissions are vented from the pile interior by positive mechanical air flow through the pile. Determining meaningful VOC flux densities with the flux isolation chamber on piles that have forced air flow aeration systems is clearly problematic. From this experiment it was impossible to tell if biochar amended compost reduced overall VOC emissions.



**Figure 11: Sum of the VOC flux density versus helium air flow correction factor. Numeric values inside symbols indicate compost pile age. Circles indicate 10% biochar pile samples; squares are control pile samples. Equipment blank helium correction factors are shown in the lower panel. Larger He correction factors and related larger VOC flux densities are attributed to positive forced aeration flow through the pile. Groups of data are apparent as connected by red dashed lines.**

While there was significant variability between samples from the same pile, there also were differences in how the flux density changed for some compounds with pile age (Figure 12). Emissions of isovaleraldehyde are initially very large but drop rapidly (2 orders of magnitude) within the first week or so. This profile contrasts with limonene (and other monoterpenes) where emissions were high for the first week or so before rapidly dropping off. Monoterpenes are likely emitted from the hot woody plant material of the pile and there is likely a large mass reservoir of

these compounds that volatilizes from the pile over time. It thus appears to take more time to volatilize this monoterpene reservoir compared to isovaleraldehyde. Dimethyl sulfide (DMS) emissions appear to peak around day 7 before rapidly decreasing, likely reflecting microbiological activity of the pile (Haug, 1993).

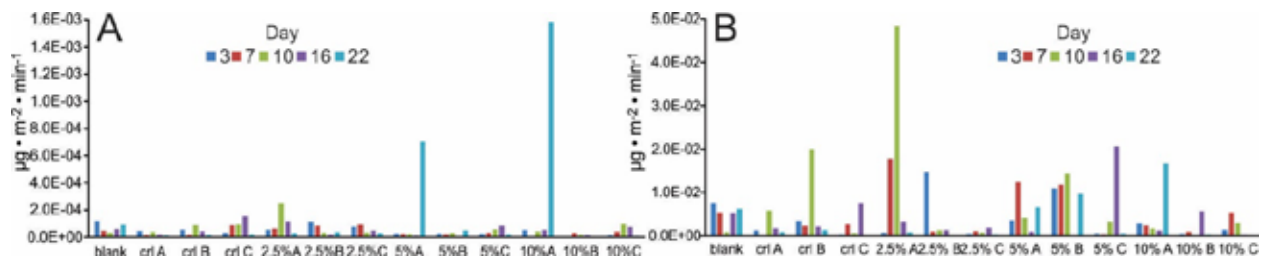


**Figure 12: Flux densities versus pile age for 3 compounds that displayed different emission profiles. Dashed lines are a rough approximation of overall emissions trends.**

Table A.1 in the Appendix lists the flux densities measured for day 3 samples to illustrate the types of compounds emitted at Lenz. The table shows 30 compounds with the highest flux densities listed by compound with the largest values for the Control 1 sample.

#### 4.1.2 WSU composting experiment

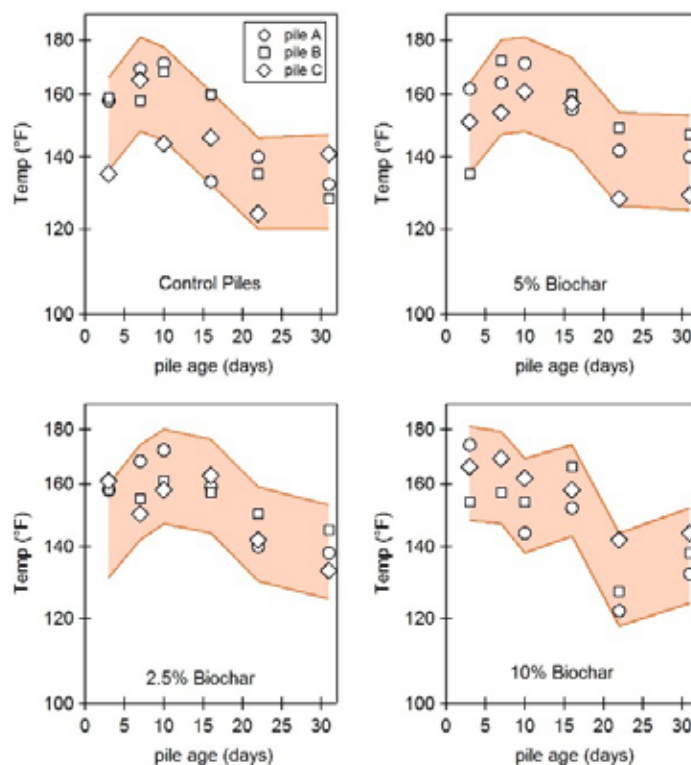
At the WSU composting facility, three replicate piles (A-C) were built for each of the three biochar levels and the control. As observed with compost produced at Lenz Enterprises, there was strong variability between the three replicate piles (Figure 13). Particularly evident are the very high values for acetaldehyde from piles A with both 5% and 10% biochar on day 22. Acetone emissions from pile B with 5% biochar and pile A with 2.5% biochar differed dramatically from the respective other two piles with the same amendment level.



**Figure 13: Volatile organics emissions at WSU Composting Facility. A: acetaldehyde and B: acetone were collected on indicated days after piles were built. Three replicate piles were built for each level of biochar-compost mixture (2.5%, 5%, or 10%, v/v) and the control (crl). Replicates are plotted individually to demonstrate the data variability.**

The cause for the high level of divergence between replicates is not entirely clear, yet likely related to adverse environmental impacts. These observations suggest that an experiment with tightly controlled environmental conditions is needed to reach reliable conclusions regarding the effect of biochar amendment on the composting process. Notably, while the reduction of composting-related nitrogenous gas emissions by biochar admixture is well documented by multiple studies (Cayuela et al., 2014; Godlewska et al., 2017), studies related to VOCs release are scarce (Sanchez-Monedero et al., 2018a).

Figure 14 illustrates how the interior pile temperatures, measured from a thermocouple probe inserted about 3 feet into the pile (average of 3 readings), varied between the piles and over the pile age. At day 3 most pile temperatures were above 140°F (60°C) and reached a maximum temperature at day 10 of ~170°F, and then slowly cooled. No systematic differences in pile temperatures between the pile types were discerned, although the 10% biochar piles did not display a noticeable peak temperature at day 10. For a given pile type, such as the 5% biochar piles, differences in temperature between the three piles (A, B, C) were more than 20°F on some days. These differences were not systematic; that is there were no consistently cold piles or hot piles. The variation in temperature differences between the piles with time was likely caused by pile turning and resulting changes in the composting process.



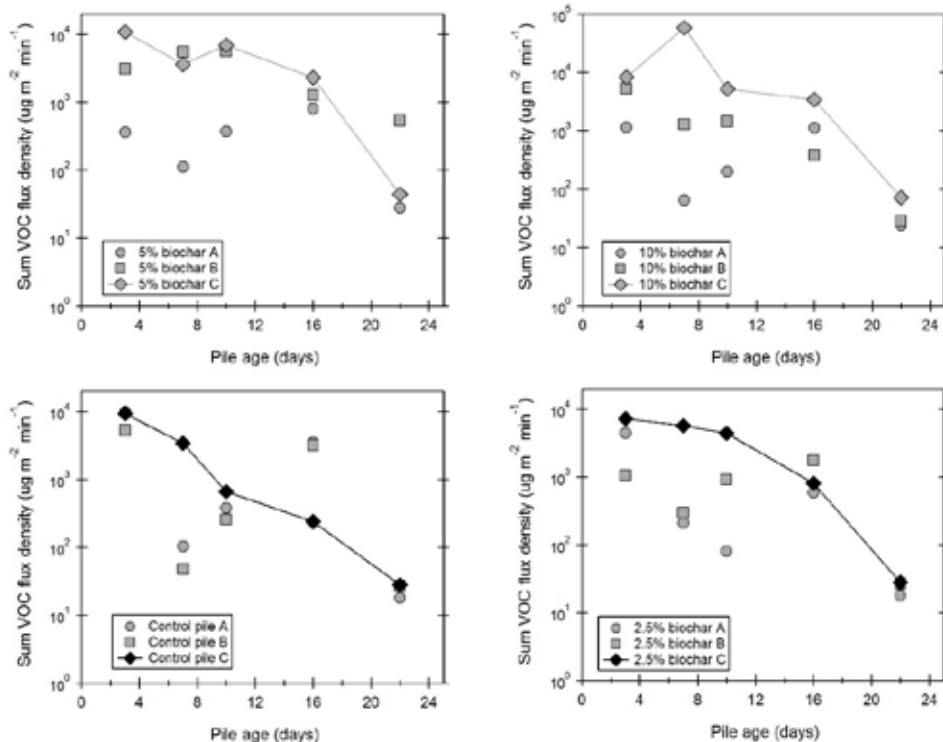
**Figure 14: Pile interior temperatures showing variation of pile temperature between the 3 piles of similar types. Shading is  $\pm 10\%$  about the average pile temperature.**

For the WSU piles, monoterpenes and sulfur compounds typically dominated the emissions. Compared to Lenz, there was a much higher abundance of sulfur compounds, such as dimethyl disulfide (DMDS), and nitrogen-containing compounds such as pyridine. This is likely due to the much higher concentration of animal manure in the WSU material mix. As was observed at Lenz, a dozen or so compounds made up the majority of the overall emissions. Table 11 contrasts the highest emitting control pile (pile A) with the highest emitting 10% biochar pile (pile C) on day 3, when overall VOC flux densities were largest. The table illustrates the types of compounds emitted by the piles and their relative flux densities. Monoterpenes compounds, principally  $\alpha$ -pinene and an identified monoterpene (perhaps ocimene) and dimethyl sulfide (DMS) and dimethyl disulfide dominated flux densities. Sulfur compounds dominated the control pile emissions, whereas monoterpenes dominated the 10% biochar emissions. The compounds listed for the control pile A in Table 11 comprised 97% of the measured VOC flux density.

**Table 11: Day 3 flux densities (ug m<sup>-2</sup> min<sup>-1</sup>) contrasting the highest emitting Control pile with the highest emitting 10% Biochar pile.**

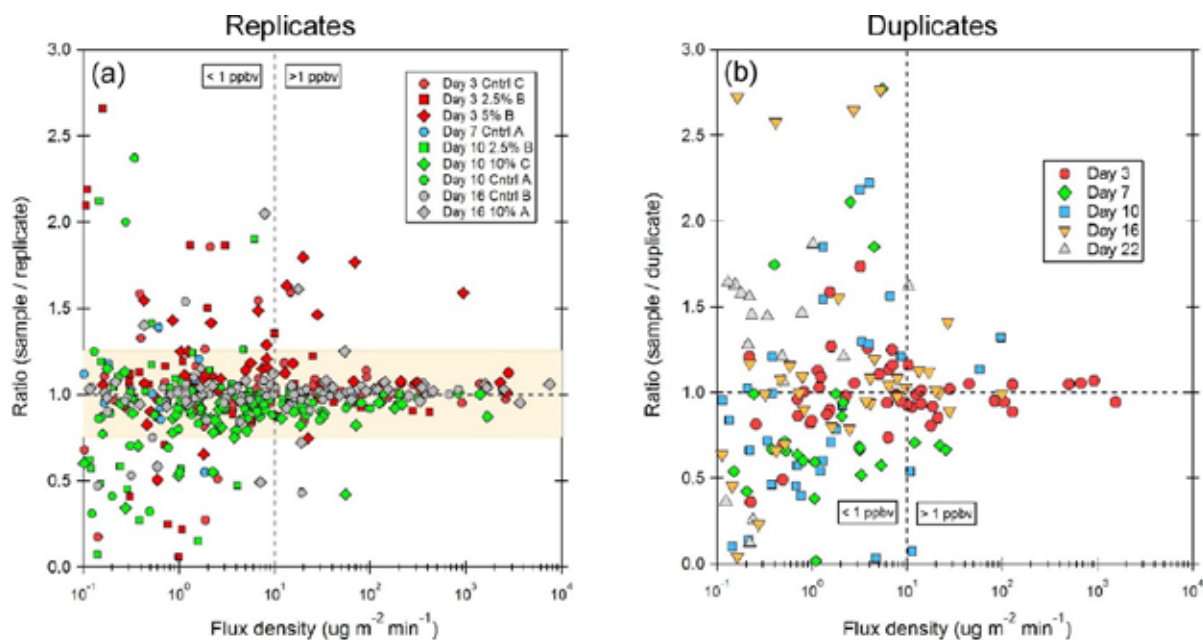
<b>Compound</b>	<b>Control pile A</b>	<b>10% Biochar Pile C</b>
<b>Sum measured VOCs</b>	10,043	8,243
<b>dimethyl disulfide (DMDS)</b>	5,803	1,154
<b>α-pinene</b>	1,761	2,403
<b>unknown monoterpene</b>	715	2,061
<b>dimethyl sulfide (DMS)</b>	352	1,479
<b>D-limonene</b>	339	200
<b>β-pinene</b>	130	154
<b>α-phellandrene</b>	109	70
<b>3-Carene</b>	108	70
<b>pyridine</b>	108	15
<b>1-Butene</b>	107	175
<b>Isoprene</b>	60	44
<b>camphor</b>	56	20
<b>terpinolene</b>	39	10
<b>styrene</b>	35	14
<b>longifolene</b>	31	3

Overall, VOC concentrations in the WSU samples were lower than at Lenz, with the sum of VOC emission flux density about a factor of 10 lower. The sum of the VOC flux densities for the different pile types are shown in Figure 15. As was observed at Lenz, there were large differences in measured flux density between the 3 piles (pile A, B, C) of identical type, suggesting the compost emission process is hard to replicate. The overall lower flux densities at WSU are likely due to the much smaller mass of material below the flux chamber compared to Lenz. Again, the pile to pile variability within a pile type precluded identifying VOC flux density differences between the biochar piles and the control piles. This was also true when individual compound flux densities were examined. The general trend was for flux densities to decrease with pile age. No flux density calculations were done for day 32 as these canisters were cleaned, inadvertently, before the helium analysis was done.



**Figure 15: Comparison of the sum of VOC flux densities versus pile age for the 4 pile types constructed at WSU.**

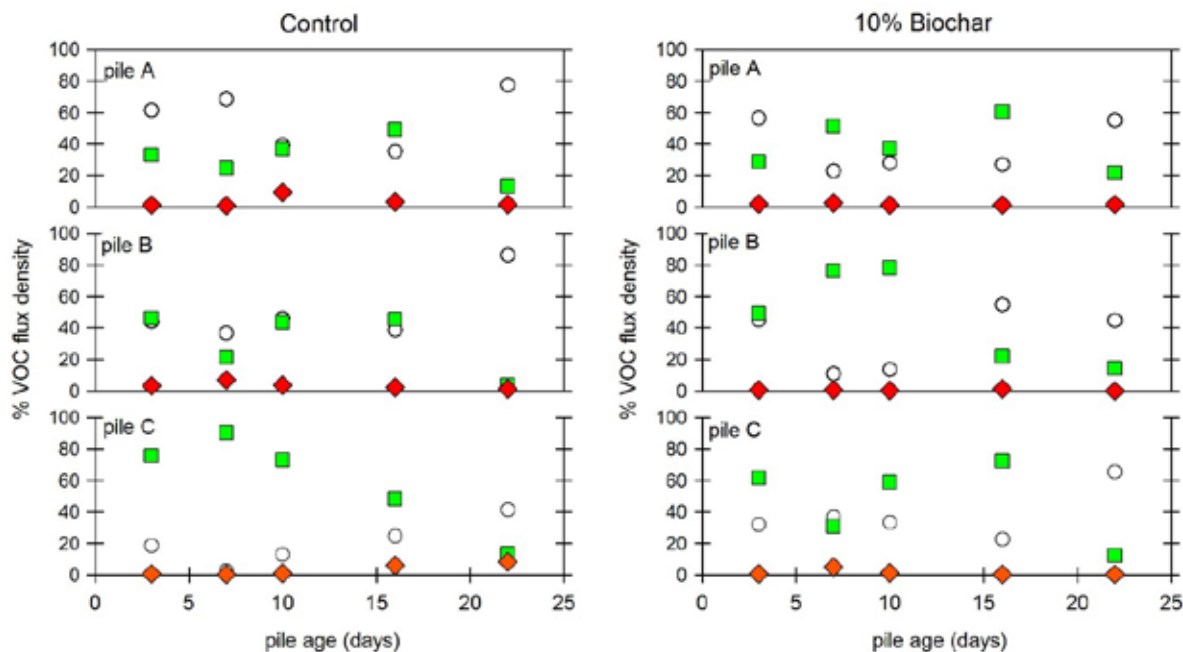
The flux density variability was attributed to differences in pile behavior and not sampling. Figure 16 shows the results from replicate sample analysis (panel a) and duplicate sample analysis (panel b). In the replicate analysis the sample canister was analyzed twice, and the ratio of the reported concentrations shown as a function of the calculated flux density. Replicate sampling was sometimes done within hours of the first analysis and sometimes days later. The variability is in part determined by the precision of the analysis and by sample storage issues, a particular concern for sulfur and nitrogen compounds. The replicate analysis plot illustrates that analysis precision was typically better than 25% for most compounds at concentrations that spanned a flux density that varied over 5 orders of magnitude. The analysis precision was better for compounds at concentration above 1 ppbv (part per billion by volume). For the duplicate samples more variability was observed, especially for components less than 1 ppbv in concentration, implying additional uncertainty due to sampling from the flux chamber. However, this variability is still less than the factor of 10 to 100 differences observed between similar pile types illustrated in Figure 14.



**Figure 16: Panel (a) shows results from sample replicates (canister analyzed twice) and panel (b) the results from duplicate samples from the flux chamber. Each data point is a particular compound. A ratio of 1 indicates perfect agreement. Shading in (a) is  $\pm 25\%$  about the ratio of 1. The vertical dashed line indicates the approximate mixing ratio of 1 ppbv.**

The compositional differences in VOC emissions between the Control piles and 10% Biochar piles are shown in Figure 17. The 10% Biochar piles are used as a comparison as presumably the piles with highest biochar composition would display the biggest effect on VOC emission reductions. The figure shows the percent composition of the measured VOC flux density for 3 types of compounds: monoterpenes, S-containing compounds (DMS, DMDS, CS<sub>2</sub>) and N-containing compounds (pyridine, 3-methyl pyridine, propionitrile, isobutylnitrile, N-methylformamide). No clear differences between the Control piles and Biochar piles are evident. The total flux densities and relative composition varied between the 3 piles (A, B, C) making it difficult as ascribe a typical compound emissions profile to Control piles or the 10% Biochar piles. A general observation was that as monoterpene emissions declined with pile age, the S-containing compounds became a greater fraction of pile emissions. The control piles also typically had larger N-containing compound fluxes than the 10% Biochar piles, hinting at the potential of biochar to remove emissions of these types of compounds. For the Control piles, about half the samples (7 samples) had N-compound emissions > 2% of the total, while only 2 samples from the 10% Biochar piles had emission greater than 2%. Emissions of N-containing compounds were as large as 9% in the Control piles. For determining the emissions from the WSU compost material, analytical methods that can better measure S- and N-containing compounds, including those not measured here such as hydrogen sulfide and amines such as methylamine, are likely important for an accurate assessment of VOC emission rates, and impact of biochar on reducing emissions of odor compounds. The figure illustrates the importance of measured S-containing compounds to the overall total flux density. Analysis of these types of compounds is typically done by air sample collection into Teflon sampling bags rather than SUMMA canisters to avoid potential storage loss issues. The same concerns would apply to the N-containing compounds. For this study the canisters were analyzed the day following the

sample collection, reducing potential storage loss. In addition, the very high ammonia concentrations in the flux chamber (> 500 ppmv) may have helped in passivating the canisters and thus mitigate losses of S- and N-containing compounds to the canister surface. Future sampling from agricultural compost piles should utilize in-situ VOC measurement technology, as described in the following section with laboratory composting, to avoid losses to storage containers.



**Figure 17: The relative contribution of the overall measured VOC flux density for S-containing compounds (open circles), monoterpenes (green squares), and N-containing compounds (red diamonds). Results from the control piles and 10% biochar piles are shown. S-compounds tended to comprise a larger fraction of the emission for older pile ages.**

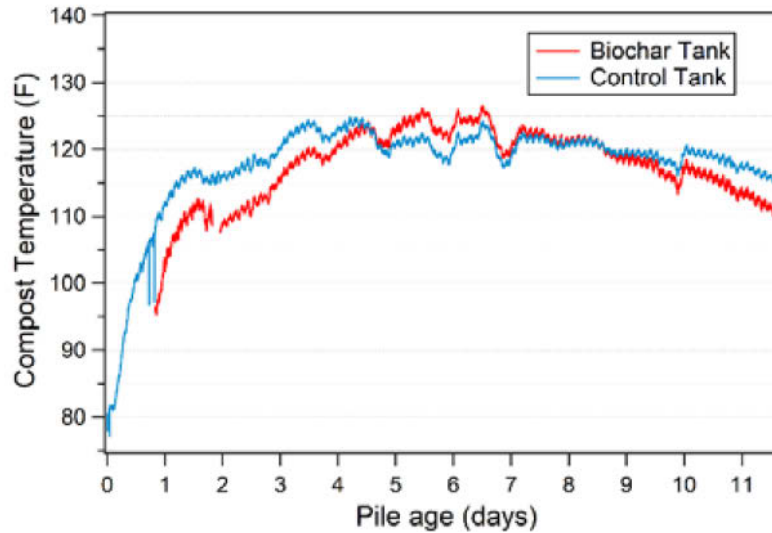
#### 4.1.3 Laboratory compost test

Composting in 100-gallon tanks was performed in two trials with WSU compost material. The first trial was conducted from 27 March to 8 April 8, 2019. In each trial continuous emission monitoring was performed and flux densities calculated to compare emission from a control tank to a tank containing 10% biochar. The mass flux emitted per tank ( $\mu\text{g min}^{-1}$ ) was compared to see if the biochar amendment reduced emissions for some compounds.

##### *Temperature*

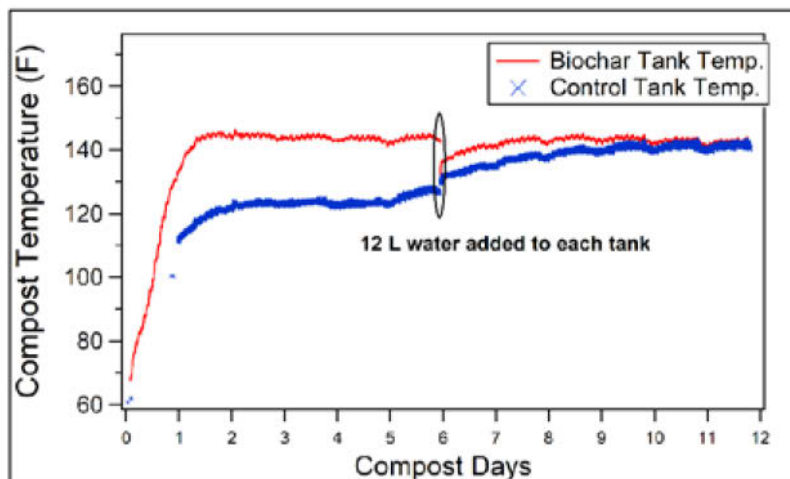
Compost temperature in the control and biochar tanks was monitored continuously. The measured temperatures for trials 1 and 2 are shown in Figure 18 and Figure 19, respectively. For the first trial, compost temperature profiles in both tanks were similar but the maximum temperature did not go above 125°F, short of the expected 140°F target (Barthod et al., 2018). The lower temperature may be a result of non-optimal conditions in the tank due to limited oxygen, deficient moisture level, and/or lack of optimum carbon to nitrogen (C:N) ratio (Mason, 2006). After the trial, the compost was turned out of the tank and we observed the top third to be dry but the bottom half was still quite moist. Hence, it was assumed that temperature did not get high enough due to lack of aeration or too high a C:N ratio in the compost.





**Figure 18: Compost temperature variation with time in the biochar tank (red trace) and the control tank (blue trace) during first trial of laboratory compost test.**

For the second trial the biochar tank quickly came up to temperature and was > 140°F after the first day. Because of initial trouble in logging compost temperature in the control tank, its temperature data was being recorded manually during the first day until the problem was fixed. The control tank temperature did not increase as quickly as the biochar tank and was about 25°F colder than the biochar tank for the first 5 days. Twelve liters of water was added to both tanks on day 6 after we noticed the surface of the tanks looked very dry. Adding water initially caused the temperature of the biochar tank to drop, while the temperature of the control tank increased slightly. Both tank temperatures slowly climbed with time after adding water and were at the same 140°F temperature by day twelve when the experiment ended.

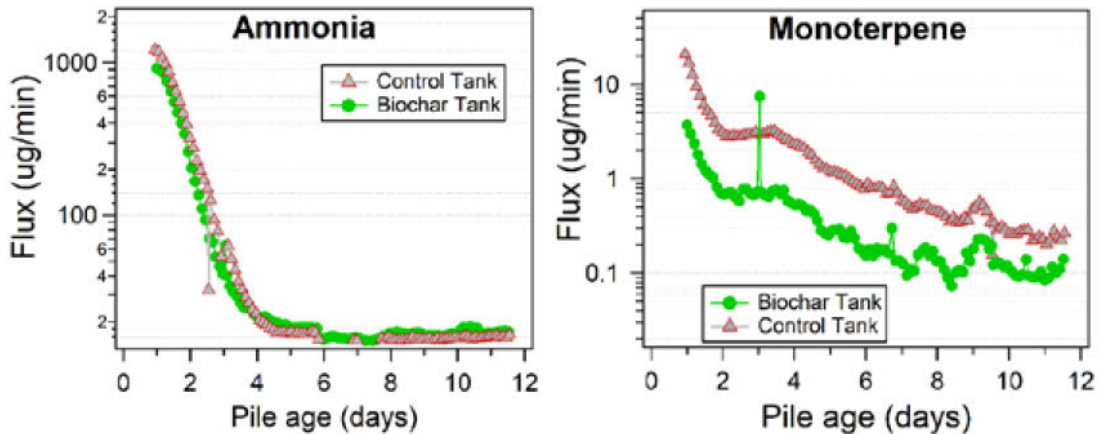


**Figure 19: Comparison of compost temperature in the biochar tank (red trace) and control tank (blue trace) during second trial of laboratory compost test.**

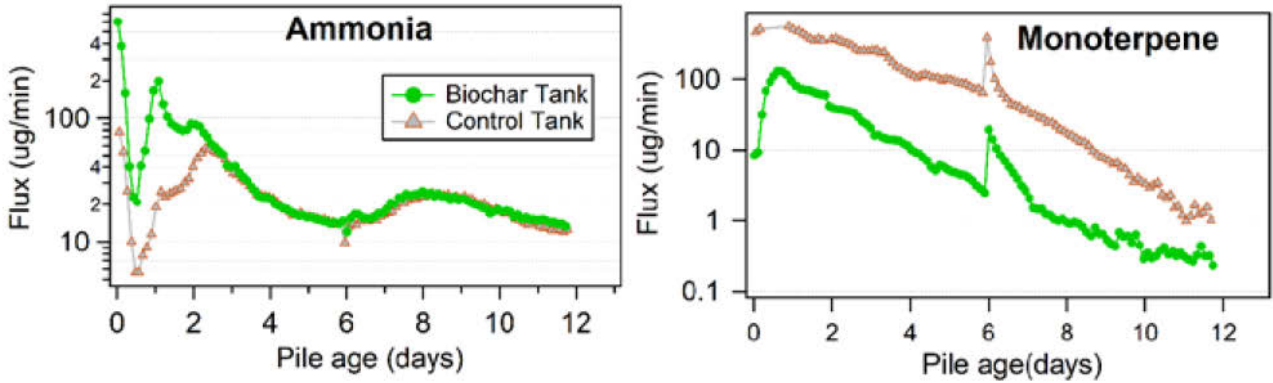
### *Emissions of ammonia and monoterpenes*

Volatile organic compounds and ammonia ( $\text{NH}_3$ ) flux values ( $\mu\text{g min}^{-1}$ ) from both tanks was averaged over 1 hour to construct time dependent emission profiles for the VOCs measured by the PTR-MS. The area under the VOC flux profile is the total mass emitted. For the first trial there is missing data for day 1 because of problems with the sampling system. The flux profiles for  $\text{NH}_3$  and monoterpenes for trials 1 and 2 are shown in Figure 20 and Figure 21, respectively. In trial 2, the monoterpene fluxes were much greater than in trial 1, likely due to the differences in compost materials: trial 2 materials had much more food waste. In both trials, monoterpene emissions from the biochar tank were significantly lower than the control tank and monoterpene emissions decreased steadily with time. Monoterpene fluxes after 12 days were about 100 times lower than the first day fluxes. The addition of water in trial 2 caused a spike in emissions of monoterpenes from both tanks.

Ammonia emissions from the biochar tank were slightly lower than from the control tank in trial 1, with ammonia emissions dropping rapidly in both tanks during the first 4 days; the change in ammonia fluxes was about a factor of 50. In trial 2, ammonia emissions were lower in the control tank for the first 2.5 days, but afterwards the ammonia flux was similar for both tanks. The initial difference in flux may be due to the much higher biochar tank temperature and therefore more rapid volatilization of  $\text{NH}_3$  from the pile. The addition of water in trial 2 did not cause a spike in ammonia emissions as observed for monoterpenes, but afterwards there was a steady increase in emissions from both tanks for about 2 days, followed by a slow decline in emissions.



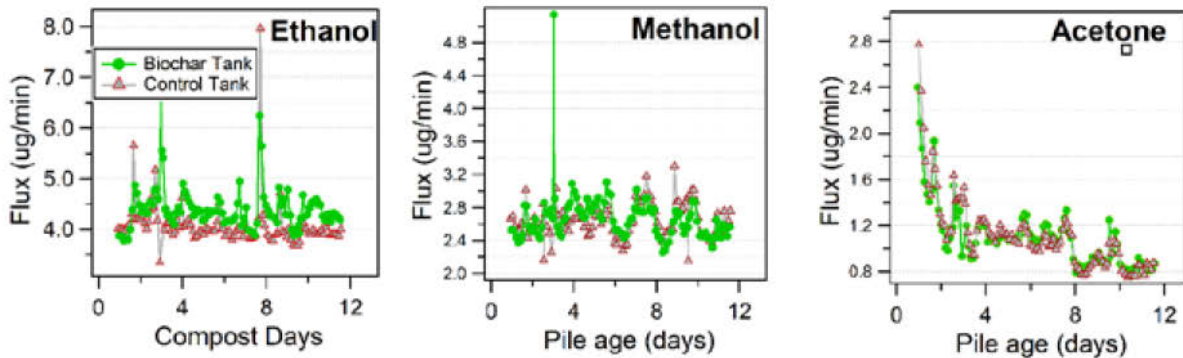
**Figure 20: Emission profiles of ammonia and monoterpene from the 10% biochar tank and control tank during the first trial.**



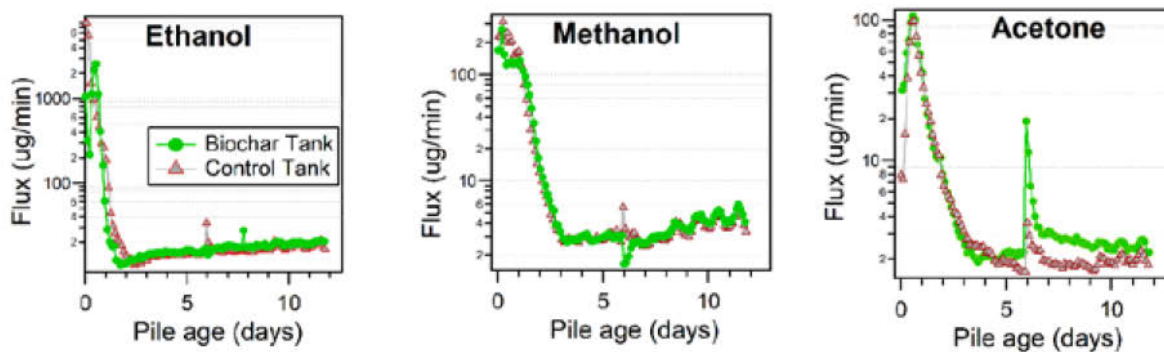
**Figure 21: Emission profiles of ammonia and monoterpene from the 10% biochar tank and control tank during the second trial.**

*Emissions of alcohols and ketones*

The PTR-MS data also provided data on the emissions of ethanol, methanol, and acetone; these fluxes are shown in Figure 22 and Figure 23 for trials 1 and 2, respectively. The second trial had much higher fluxes of these compounds than the first trial, likely attributed to the higher food waste content of trial 2 materials. In trial 2 fluxes of ethanol and methanol from both tanks decreased by about two orders of magnitude during the first couple of days. These fluxes were much higher than the monoterpene flux. For both trials the biochar and control tanks had similar fluxes and displayed similar variability. Acetone fluxes displayed a peak at about day 1 in trial 2 and fluxes dropped in both tanks by about a factor of 50 by day 3. In both trials the biochar and control tanks display similar fluxes. The addition of water in trial 2 caused acetone fluxes to sharply increase in both tanks, and afterwards the fluxes from the biochar were significantly larger for about 4 days. There was little evidence that biochar reduced emissions for these compounds.



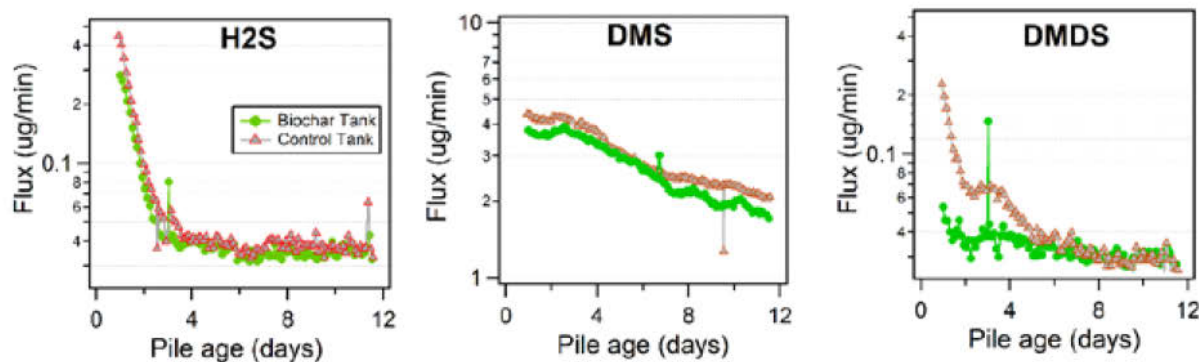
**Figure 22: Emission profiles of methanol, ethanol and acetone from the 10% biochar tank and control tank during the first trial.**



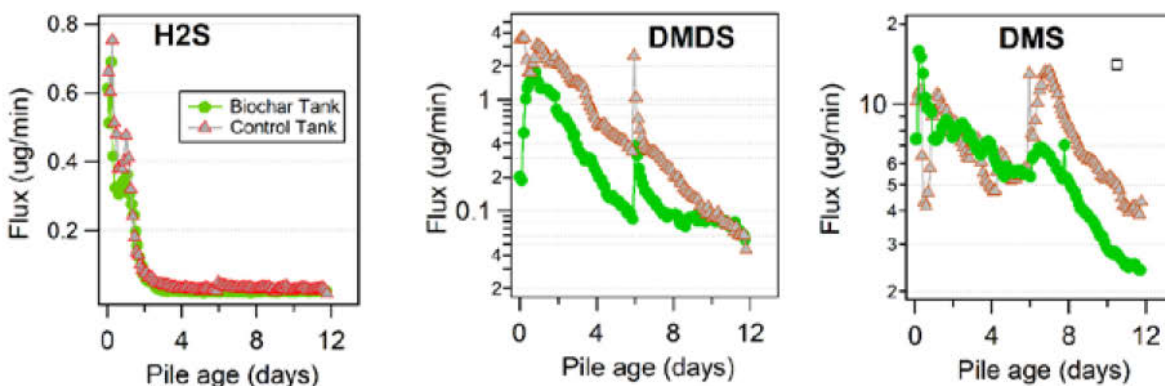
**Figure 23: Emission profiles of methanol, ethanol and acetone from the 10% biochar tank and control tank during second trial.**

### *Emissions of sulfur compounds*

Flux profiles of the sulfur compounds  $\text{H}_2\text{S}$ , DMS, and DMDS are shown in Figure 24 and Figure 25 for trials 1 and 2, respectively. Fluxes were generally higher for trial 2. For both trials the largest flux was observed for DMS. In both trials DMS fluxes from the biochar and control tanks were similar and slowly decreased with time. The addition of water in trial 2 caused increases in the DMS flux, and afterwards the control tank had higher fluxes than the biochar tank.



**Figure 24: Emission profiles of sulfur compounds from the 10% biochar tank and the control tank during first trial.**



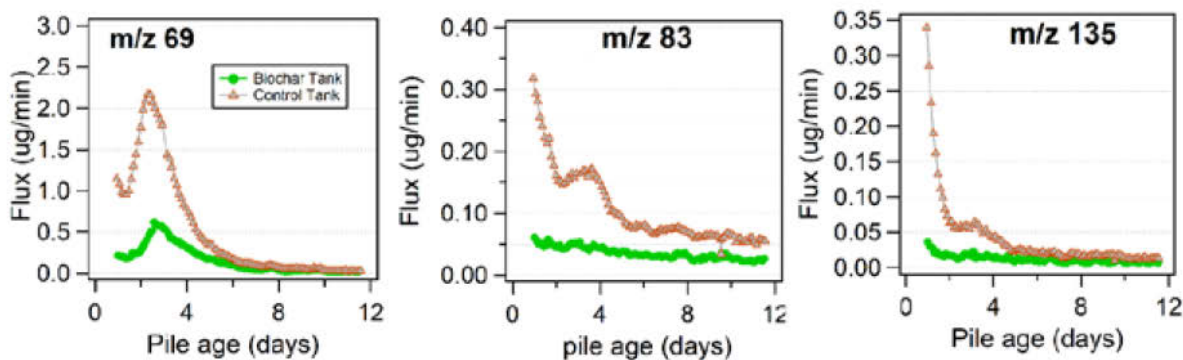
**Figure 25: Emission profiles of sulfur compounds from the 10% biochar tank and the control tank during the second trial.**

Hydrogen sulfide (H<sub>2</sub>S) fluxes for both trials were similar between the tanks. Fluxes of H<sub>2</sub>S in both trials were initially largest on day 1 and dropped rapidly to much lower values by day 4. Thereafter H<sub>2</sub>S fluxes stayed constant until the end of the experiment.

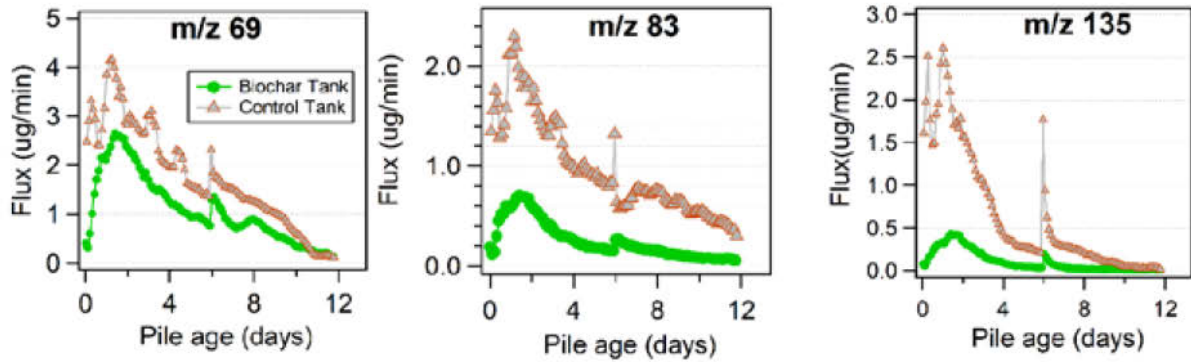
In contrast to H<sub>2</sub>S and DMS, the biochar tank had much lower fluxes of DMDS in both trials. For this compound it appears biochar has some removal capacity. Fluxes of DMDS were observed to peak in day 1 and drop steadily over the course of the experiment. Addition of water in trial 2 caused a sharp increase in emissions from both tanks.

#### *Emissions of unknown compounds*

Interesting fluxes profiles were noted for several ion signals in the PTR-MS for which a clear attribution to a particular compound or compounds could not be immediately made. This will require analysis of the GC-MS data collected. What was noteworthy with these compound(s) was that in both trials much lower fluxes were observed from the biochar tank. Fluxes of *m/z* 69 displayed a peak on day 2 (Figure 26 and Figure 27), suggesting compost temperature and microbiological activity are important factors.



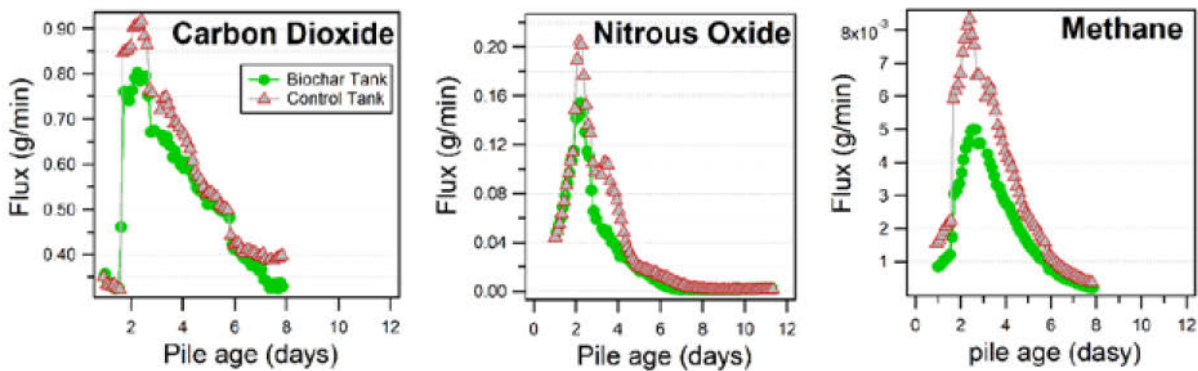
**Figure 26: Emission profiles of *m/z* 69, *m/z* 83, and *m/z* 135 compounds from the 10% biochar tank and the control tank during first trial.**



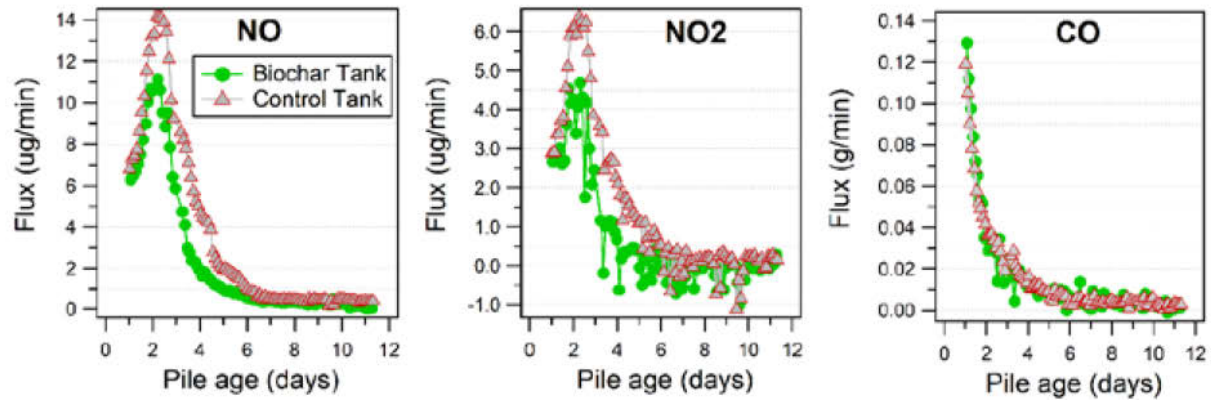
**Figure 27: Emission profiles of m/z 69, m/z 83, and m/z 135 compounds from the 10% biochar tank and the control tank during the second trial.**

*Emissions of carbon dioxide, methane and nitrous oxide and other gases*

For the first trial fluxes were determined for CO<sub>2</sub>, CH<sub>4</sub>, and N<sub>2</sub>O as well as for CO, NO, and NO<sub>2</sub>. Figure 28 shows fluxes of the fluxes of greenhouse gases (CO<sub>2</sub>, CH<sub>4</sub>, and N<sub>2</sub>O) and Figure 29 shows fluxes for CO, NO, and NO<sub>2</sub>. Fluxes of CO<sub>2</sub>, CH<sub>4</sub>, and N<sub>2</sub>O would be a general indicator of microbial activity and the decomposition process. Fluxes of CO<sub>2</sub> peaked sharply at day 2 then declined steadily till day 8 when data stopped being collected due to an instrument logging failure. Methane emissions displayed a sharp increase at the end of day 2 when the tank aeration ceased and peaked around day 3. The emissions of methane indicate that methanogenic bacteria were growing due to lack of oxygen (Kebreab et al., 2006) so that sharp increase in methane flux at that time is consistent with the reduction in oxygen being supplied to the bottom of the pile. Methane fluxes were about 100 lower than CO<sub>2</sub> fluxes and were significantly higher in the control tank.



**Figure 28: Greenhouse gases emission profiles in biochar and control tank during first trial of laboratory compost test.**



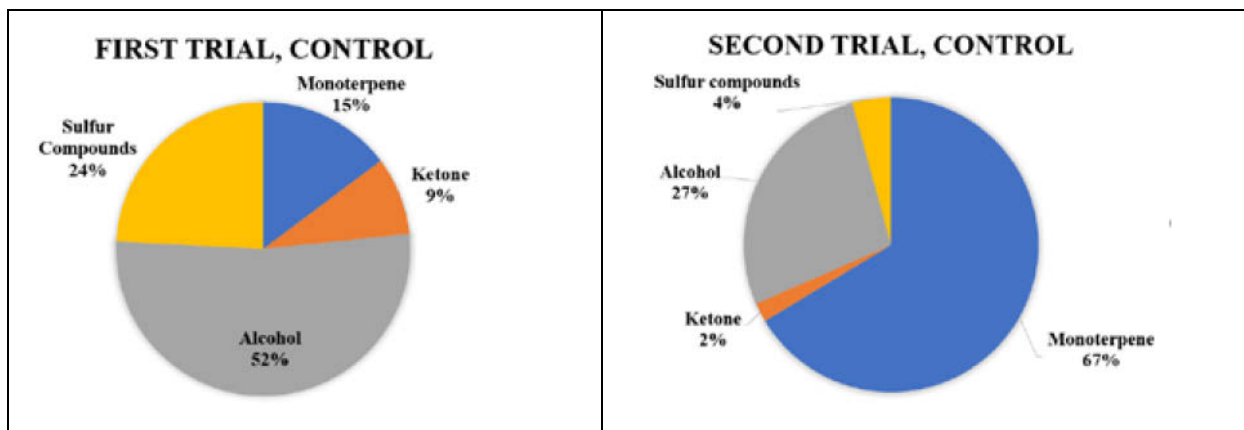
**Figure 29: NO, NO<sub>2</sub>, and CO emission profiles in biochar and control tank during first trial of laboratory compost test.**

Nitrous oxide fluxes peaked around day 3, coincident with peak fluxes of NO and NO<sub>2</sub>. Fluxes of N<sub>2</sub>O were somewhat higher in the control tank. In the control tank there appeared to be a secondary emission peak around day 4. N<sub>2</sub>O fluxes declined steadily to about zero by day 7.

Flux profiles of the gases CO, NO, and NO<sub>2</sub> are shown in Figure 29. Based on previous studies, emissions of CO occur during aerobic decomposition of organic waste and its maximum level was found at the beginning of the composting process (Hellebrand and Kalk, 2001b; Hellebrand and Schade, 2008). Measured CO levels in this trial are consistent with those reports. The difference in CO flux between the tanks was not significant and the flux was essentially zero by day 7.

#### *Comparison between trial 1 and trial 2*

Figure 30 compares the fractional emissions contribution of measured alcohols, monoterpenes, ketones, and sulfur compounds from the control tanks for the two trials, illustrating differences in emissions, presumably due to differences in material composition, temperature, and aeration. Significant differences were noted. In the first trial emissions were dominated by alcohols that displayed a constant emission rate over time, while in the second trial emissions of monoterpenes were more important. In trial 1 emissions of sulfur compounds were much more predominant than in trial 2. These compositional differences may be important in assessments of biochar's ability to reduce emissions of odorous compounds. If some types of non-odorous compounds are preferentially adsorbed by the biochar, then the effectiveness of biochar to remove odorous sulfur gases, for example, may be reduced.



**Figure 30: Comparison of VOC emission contributions of different compound types from the control tank in trial 1 and trial 2.**

The effectiveness of biochar in reducing VOC and ammonia emissions for selected compounds is shown in Table 12. The flux reduction was calculated as the difference in mass emitted between the control tank and biochar tank over the course of the experiment ( $\mu\text{g}$ ) divided by the control tank emitted mass. Since there was 10% less compostable material in the biochar tank, differences significantly greater than 10% are assumed to be meaningful reductions. Negative values mean the biochar tank had greater emissions. For trial 1, the similar temperature profiles suggest a more similar composting intensity occurred in the control and biochar tanks. The much warmer temperatures for the 10% biochar tank in trial 2, suggests differences in the composting process, and may complicate comparisons of absolute VOC fluxes between the tanks to determine biochar impacts. Nevertheless, these comparisons are made in the table to quantify the experiment outcomes.



**Table 12: Summary of VOC emissions for both trials in control and biochar tanks**

Compound	Sampled Tank	1 <sup>st</sup> Trial		2 <sup>nd</sup> Trial	
		Emitted mass (ug)	Flux reduction in Biochar Tank (%)	Emitted mass (ug)	Flux reduction in Biochar Tank (%)
Ammonia	Biochar	521,040	24	244,252	-78
	Control	681,691		137,492	
Monoterpene	Biochar	3,110	74	516,925	46
	Control	11,779		965,948	
m/z 69	Biochar	1,249	60	7,400	38
	Control	3,151		11,872	
m/z 83	Biochar	243	63	1,720	74
	Control	653		6,589	
Ethanol	Biochar	27,443	-8	162,518	48
	Control	25,416		312,779	
DMS	Biochar	17,324	7	40,203	17
	Control	18,609		48,385	
DMDS	Biochar	215	29	2,320	60
	Control	302		5,842	
H <sub>2</sub> S	Biochar	322	18	507	15
	Control	391		597	
Methanol	Biochar	16,649	-1	137,007	17
	Control	16,492		164,722	
Acetone	Biochar	6,980	-2	35,317	23
	Control	6,871		45,675	
m/z 135	Biochar	87	66	728	83
	Control	255		4,334	

Reductions of ~ 50% or more were observed in both trials for monoterpenes, the unknown compound corresponding to the PTR-MS ion signal at *m/z* 83, and the unknown compound corresponding to the PTR-MS ion signal at *m/z* 135. Significant reductions were also observed for the unknown compound corresponding to the PTR-MS ion signal at *m/z* 69, where biochar reduction was 60% for the first trial and 38% for the second trial. For these compounds the 10% biochar tank displayed consistently lower fluxes and biochar appears to reduce emissions. Monoterpenes are a major VOC emission from composts and so reducing these emissions would be potentially important for reducing odor and reducing total VOC emissions for regulatory compliance.

Significant reductions were also observed for DMDS; 29% in the first trail and 60% in the second. For DMS, which displayed much larger fluxes than DMDS, the reduction was only 7% in the first trail and 17% in the second trial. Similarly, H<sub>2</sub>S reductions were also lower than those of DMDS, 18% for the first trail and 15% for the second. The observed reductions in DMDS and H<sub>2</sub>S may be due to the difference in compost mass between the tanks, rather than the impact of biochar. For these odorous sulfur compounds, the 10% biochar loading really only had a significant impact on DMDS emissions.

For ethanol, methanol, and acetone, trial 1 showed no significant difference between the control and biochar tanks. However, there was missing data for the first day when fluxes appear to be greatest for these compounds given the data from trial 2. For trail 2 there were some reductions noted, ethanol 48%, methanol 17%, acetone 23%. Assessing emissions reduction for ethanol and methanol is somewhat uncertain because of the very large initial fluxes on day 1 and their rapid decrease over 2 days means missing data during this period will skew the results. For ethanol and methanol in both trails the emissions after day 3 were essentially the same for both tanks. Emission reductions for trial 2 for ethanol are thus determined by the first 2 days of data.

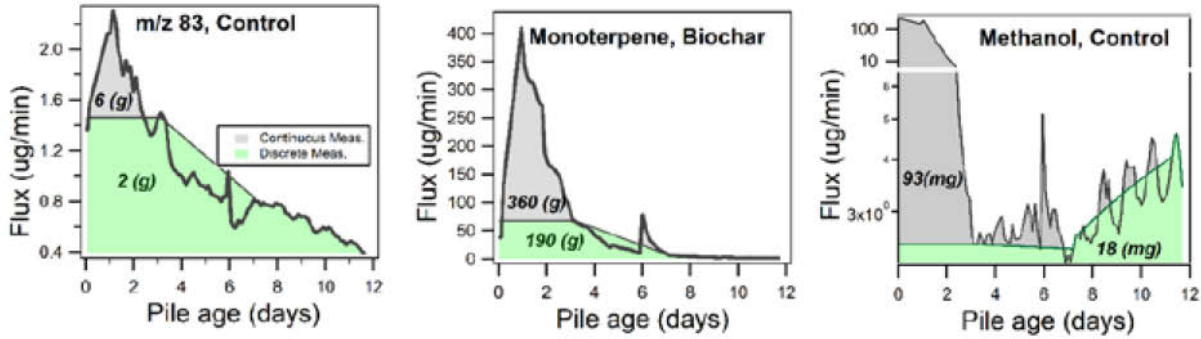
In trial 1, ammonia emissions were 24% less from the biochar tank than the control tank, hinting at potential emissions reduction potential. However, in trial 2 ammonia emissions were significantly greater from the 10% biochar tank for the first 2.5 days, causing overall NH<sub>3</sub> emissions to be 74% greater from the biochar tank. This difference could have resulted from the higher temperatures of the 10% biochar tank.

These experiments provided evidence that the addition of 10% biochar can reduce emissions of some VOCs, namely monoterpenes and DMDS, and other compounds not yet identified. The reduction of monoterpenes and DMS may help reduce odor emissions from compost. The reduction of monoterpene emissions has the potential to be useful in reducing total VOC emissions for regulatory compliance.

#### *Measurement of compost emissions: Continuous vs. discrete sampling*

The continuous emission measurements of VOCs and ammonia by the PTR-MS instrument reveals emission rates change significantly for some compounds, such as alcohols, over the first few days. Given the sampling protocol followed at Lenz and WSU to determine compost emissions, where the first sample collected was on day 3, a large fraction of the pile emissions is likely missed. If day 3 emissions are extrapolated to zero at time zero, then this discrete sampling protocol likely severely underestimates emissions for compounds like alcohols and monoterpenes. A better estimate is to assume day 3 emissions apply for the first two days. This will still severely underestimate the mass emitted for some compounds. An example is illustrated in Figure 31. The figure shows measured fluxes of 3 compounds from trial 2. Measured fluxes of methanol, monoterpenes and the compound at the ion signal  $m/z$  83 are shown (black trace) to illustrate the large initial fluxes that steeply decline to much lower values by day 3. If flux determination occurred by discrete sampling on days 3, 7, and 11 as done at WSU and Lenz, would miss the large emissions of material that occurs in the first 3 days. The green shading in the figure illustrates the estimated mass emitted as the area under the flux profile, assuming day 3 fluxes apply for the initial period. The grey shading illustrates the missing flux, and can be a significant fraction of the total mass emitted. Table 13 lists the underestimate of mass emitted by a discrete sampling protocol. For trial 2 which had the more complete data for the early phase of composting, discrete sampling underestimates the flux of monoterpenes by 47%, methanol by

81%, ethanol by 90%, ammonia by 40%, acetone by 60%, H<sub>2</sub>S by 71%, and DMDS by 18% and *m/z* 83 by 74%. Such underestimates would have a significant impact on determining total VOC emissions factors for composting as monoterpenes and alcohols typically have the largest flux densities.



**Figure 31: Comparison of emission profiles of *m/z* 83, monoterpene, and methanol estimated by continuous measurement and discrete measurements. Black trace shows measured fluxes. Area under this curve is the mass emitted. Green shading illustrates the area of the flux profile if discrete sampling at day 3, 7, and 11 was done to determine fluxes. The grey shading shows the amount of mass that is under reported by discrete sampling.**

**Table 13: Comparison between continuous measurements and discrete measurements to estimate emitted mass of VOCs**

Compound	Measurement Type	1 <sup>st</sup> Trial		2 <sup>nd</sup> Trial	
		Emitted Mass (ug)	Error (%)	Emitted Mass (ug)	Error (%)
<b>Ammonia</b>	Discrete	229,320	-66.5	144,825*	-39.5*
	Continuous	681,691		239,550*	
<b>Monoterpene</b>	Discrete	12,257	4.1	189,939*	-47*
	Continuous	11,778		360,206*	
<b>m/z 69</b>	Discrete	5258	67	11,411	4
	Continuous	3151		10,997	
<b>m/z 83</b>	Discrete	893	37	1,627.0	73.5
	Continuous	653		6,133.98	
<b>Ethanol</b>	Discrete			24,970	-90
	Continuous			241,342	
<b>DMS</b>	Discrete	-	-	-	-
	Continuous	-		-	
<b>DMDS</b>	Discrete	361	-19.5	4,122	-18
	Continuous	302		5,031	
<b>H<sub>2</sub>S</b>	Discrete	-	-	128	-71
	Continuous	-		432	
<b>Methanol</b>	Discrete	-	-	18,053	-81
	Continuous	-		93,265	
<b>Acetone</b>	Discrete	6042	-12	10,758	-60
	Continuous	6871		25,947	

## 4.2 Impact of amendment on crop productivity and quality

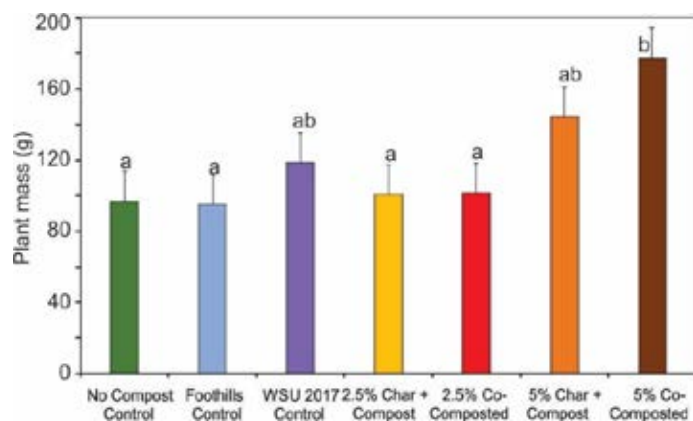
### 4.2.1 Basil field trial

The treatments did not lead to visible differences among plants (i.e., none of the treatments caused chlorosis or leaf malformation, and all plants appeared healthy; Figure 32). Both biomass production (yield) and chemical composition (herb quality) were determined.



**Figure 32: Organic Genovese basil grown at Footehills Farm (Colbert, Washington) with addition of A. WSU 2017 compost, mixed with 5% Amaron Energy biochar post-composting, B. WSU 2017 compost, co-composted with 5% Amaron Energy biochar, or C. Footehills organic compost.**

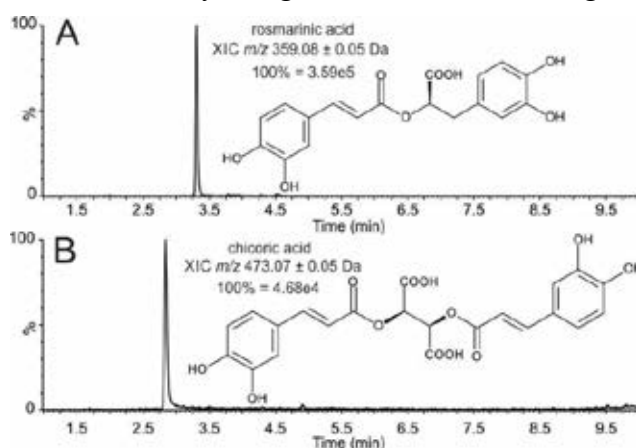
The fresh plant mass yield was very similar for four treatments: plants grown without or with Footehills compost, and plants grown with WSU 2017 compost amended with 2.5% biochar (Figure 33). The addition of WSU 2017 compost amended with 5% biochar prior to composting led to significantly higher plant mass in comparison to these four treatments (ANOVA with Tukey post-hoc HSD test,  $p < 0.05$ ). Average biomass yield was also higher when 5% non-composted biochar with WSU 2017 compost, or WSU 2017 compost by itself were added to the soil, but not significantly different in comparison to all other treatments (Figure 33).



**Figure 33: Effect of amendments on fresh mass of Genovese sweet basil grown at Foothills Farm (Colbert, Washington). Average plant mass  $\pm$  s.e.m. (n=10) are shown. Same lower case letters indicate there was no significant difference between treatments ( $p < 0.05$ ). Note that in the treatment labels, char + compost means char added post-composting, while co-composted means that char was added prior to composting.**

The outcome of this field study is in agreement with our earlier greenhouse study of basil where 5% co-composted biochar led to significant biomass increase for both cultivars (Eleanora and TSQ). Importantly, the very same compost as used in 2017 was also applied in the study presented here. In contrast, in the previous greenhouse study, amendment with co-composted biochar at a rate of 2.5% was also effective in increasing the vegetative yield of both basil cultivars significantly as compared to plants grown both without compost, or with unamended compost (Gang et al., 2018).

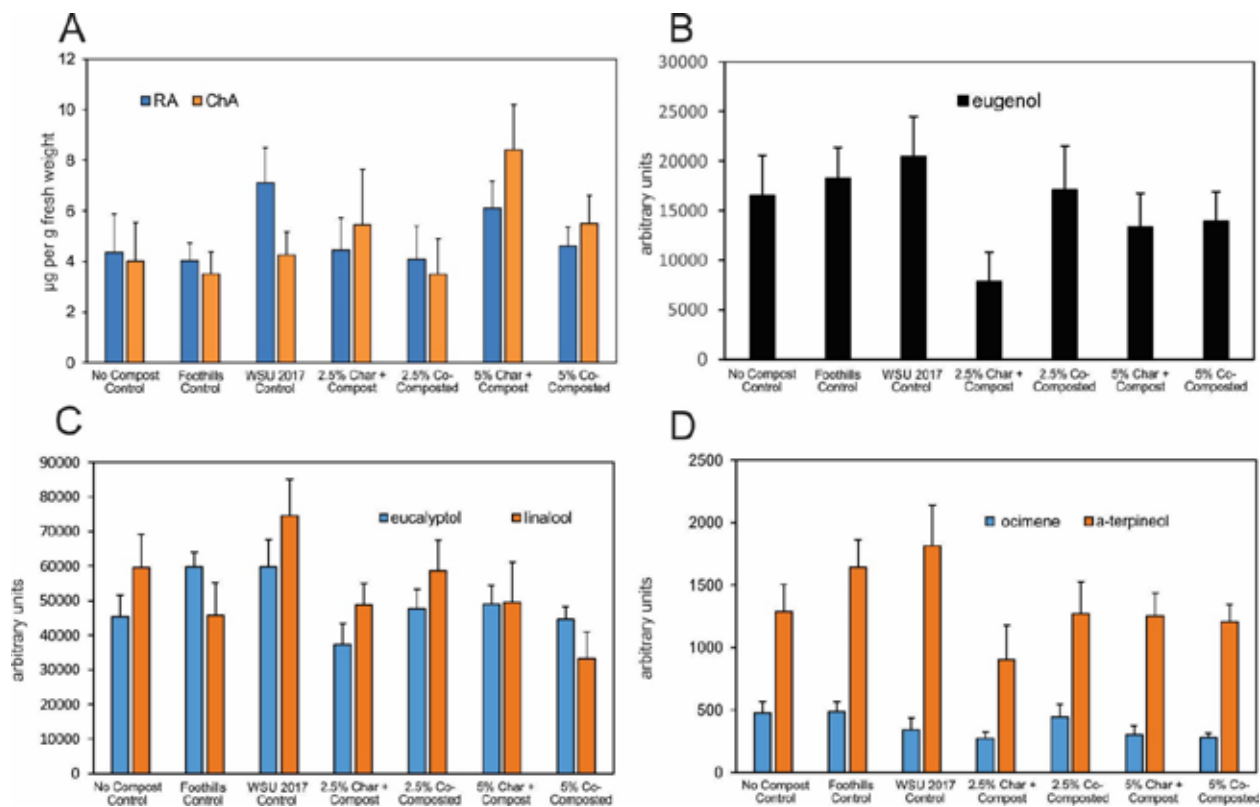
The two youngest fully expanded leaf pairs of the harvested plants were further analyzed for their content of major antioxidant phenolics rosmarinic and chicoric acid, and for the levels of aroma-defining volatile compounds. In the ‘Genovese’ variety used here, those are eugenol, linalool, and eucalyptol. The accumulation of antioxidants was low in comparison to typical contents and could be best visualized by using selected ion chromatograms (Figure 34).



**Figure 34: Analysis of non-volatile phenolics in field-grown sweet basil. Structures and representative XIC (selected ion chromatograms) showing A. rosmarinic acid and B. chicoric acid accumulation in extracts from mature basil leaves. Y-axis is scaled to strongest signal.**

The levels of the two monitored compounds differed strongly within and between treatments, as reflected by large error (Figure 35). Highest levels of rosmarinic acid were measured in WSU 2017 treatment while highest average level of chicoric acid accumulated in plants grown with WSU 2017 compost with 5% biochar added post composting. However, due to large variation mentioned above, those levels were not statistically different.

For volatile flavor components, no significant differences between treatments were found for the monitored compounds (Figure 35B-D) when the data were analyzed using one-way ANOVA ( $p < 0.05$ ). At the same time, pairwise two-tail T-tests which only compare between two selected treatments suggested some significant differences could be observed. For example, the concentration of linalool decreased significantly in plants grown with 5% co-composted biochar as compared to plants treated with unamended compost (Figure 35C). Notably, some plants exhibited a deviation from the major chemotype, accumulating methylchavicol instead of eugenol. While the largest number of plants with the methylchavicol chemotype (5 of 10) was seen in plants grown with 2.5% non-composted biochar added to WSU 2017 compost, it is more likely to be genetic and coincidental rather than an induced shift in metabolism causatively linked to the amendment.



**Figure 35: Production of non-volatile antioxidants and volatile aroma compounds in in two youngest mature leaf pairs of Genovese sweet basil grown at Foothills Farm (Colbert, Washington). Accumulation of A. major phenolics rosmarinic acid (RA) and chicoric acid (ChA), B. eugenol, C. eucalyptol and linalool, D. ocimene and  $\alpha$ -terpineol Y-axis is in  $\mu\text{g g}^{-1}$  fresh weight for A, and in arbitrary units (normalized to internal standard and tissue weight) for B-D. Note that in the treatment labels, “char + compost” indicates char added post-composting, while co-composted means that char was added prior to composting.**

Similarly to the above, our earlier greenhouse study (Gang et al., 2018) supported the view that biochar-compost mixtures (whether co-composted or added post-composting) do not significantly affect the aroma or phenolic production in sweet basil. Another earlier investigation which evaluated the effect of biochar admixture came to the same conclusion, even though no compost plus biochar or co-composted biochar treatments were tested there (Pandey et al., 2016). Our most recent field study needs to be reproduced to confirm the biomass increase observed in plants amended with 5% biochar, added before or after composting. Adding a treatment with 10% co-composted biochar or biochar added after composting would benefit our understanding of biochar's dose-dependent effect on yield.

#### **4.2.2 Basil greenhouse trials**

The experiments with greenhouse grown basil were conducted in both 2017-2019 and 2019-2021 biennia and aimed to reproduce the results obtained in our earlier study and evaluate the effects of two other compost types produced at different facilities. In 2017-2019 biennium, basil plants of Eleanora and Thai Siam Queen varieties were grown in the greenhouse under the same conditions as in March-May 2017, but at a different time of the year (late November-first days of February). The trial included thirteen treatments with three different types of compost: produced at WSU in 2017 and 2018, and produced at Lenz in 2018. Composts produced in 2018 were co-composted with the same biochar from Oregon Biochar Solutions.

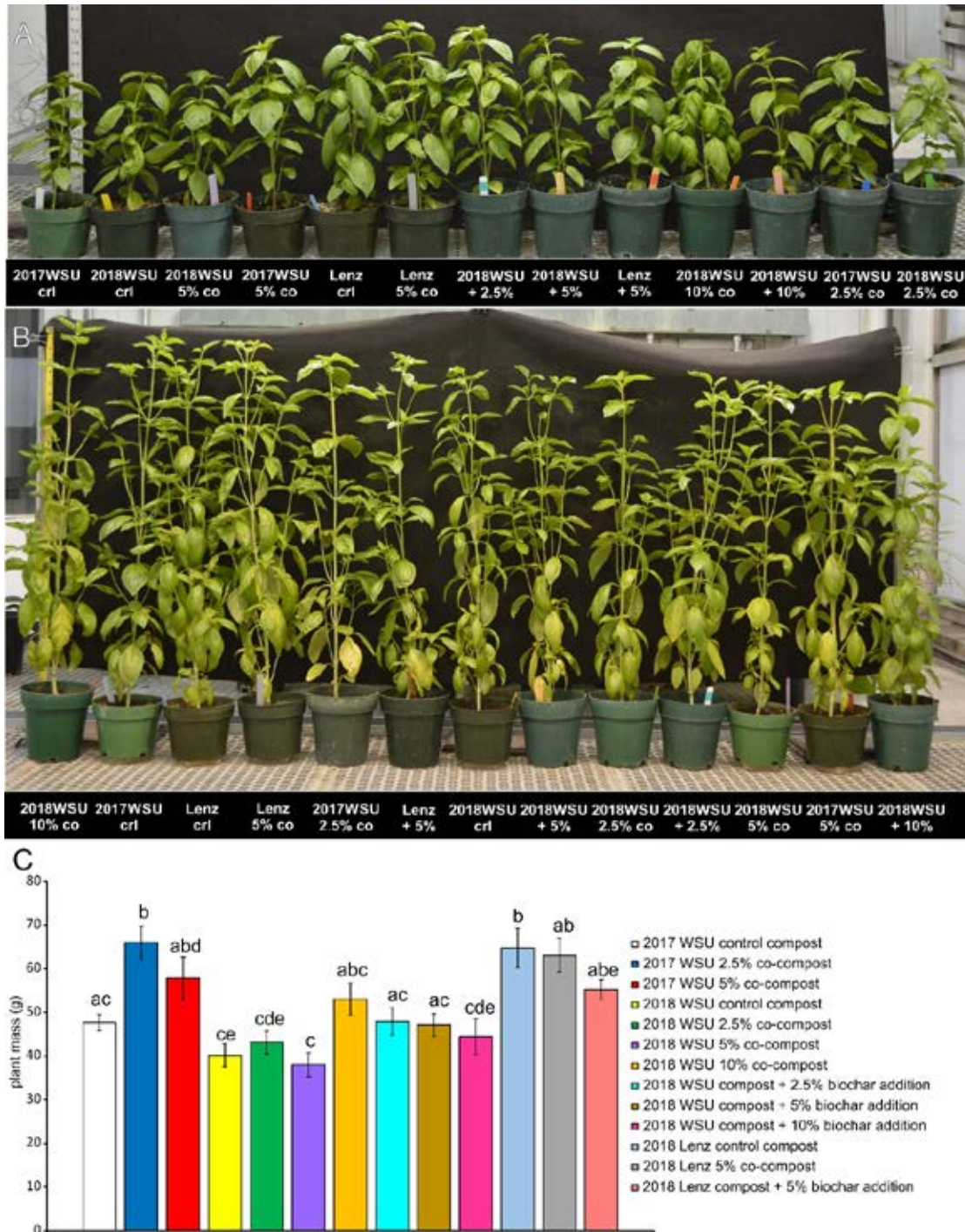
The plants of both varieties grew faster than in the earlier study (Gang et al., 2018). About 40 days after transplanting they had already reached harvest-suitable size (Figure 36A, Figure 37A). To observe their further growth dynamics, they were left to grow until 66 days after transplant when they were harvested. At that time point, Eleanora basil plants still looked healthy (Figure 36B), with the exception of the first true leaf pair which was often yellowed and sometimes wilted, while 'TSQ' plants showed clear signs of leaf damage and senescence (Figure 37B).

The amendments resulted in significant biomass yield differences across treatments. The effects of amendments differed between the two sweet basil cultivars. Application of Lenz compost, alone, co-composted with biochar or mixed with biochar post-composting, had no significant effect on biomass yield in both Eleanora and TSQ (Figure 36C, Figure 37C). WSU 2017 compost, alone or co-composted with 2.5% or 5% Amaron Energy biochar, had no significant effect on biomass yield of TSQ (Figure 37C). In Eleanora, average mass of plants grown with WSU 2017 compost, co-composted with 2.5% biochar, was significantly greater in comparison to plants grown with WSU 2017 compost alone. Co-compost with 5% biochar resulted in biomass yield that appeared at first glance to increase, however, it was not statistically significant (Figure 36C). Vegetative yield of Eleanora basil with WSU 2018 compost, alone, co-composted biochar or biochar added after composting with different amounts of biochar, was statistically unchanged. In TSQ, biomass yield from plants grown with WSU 2018 compost admixed with all three levels of biochar was statistically lower than yields after co-composting 5% or 10% of the same biochar with the same compost.

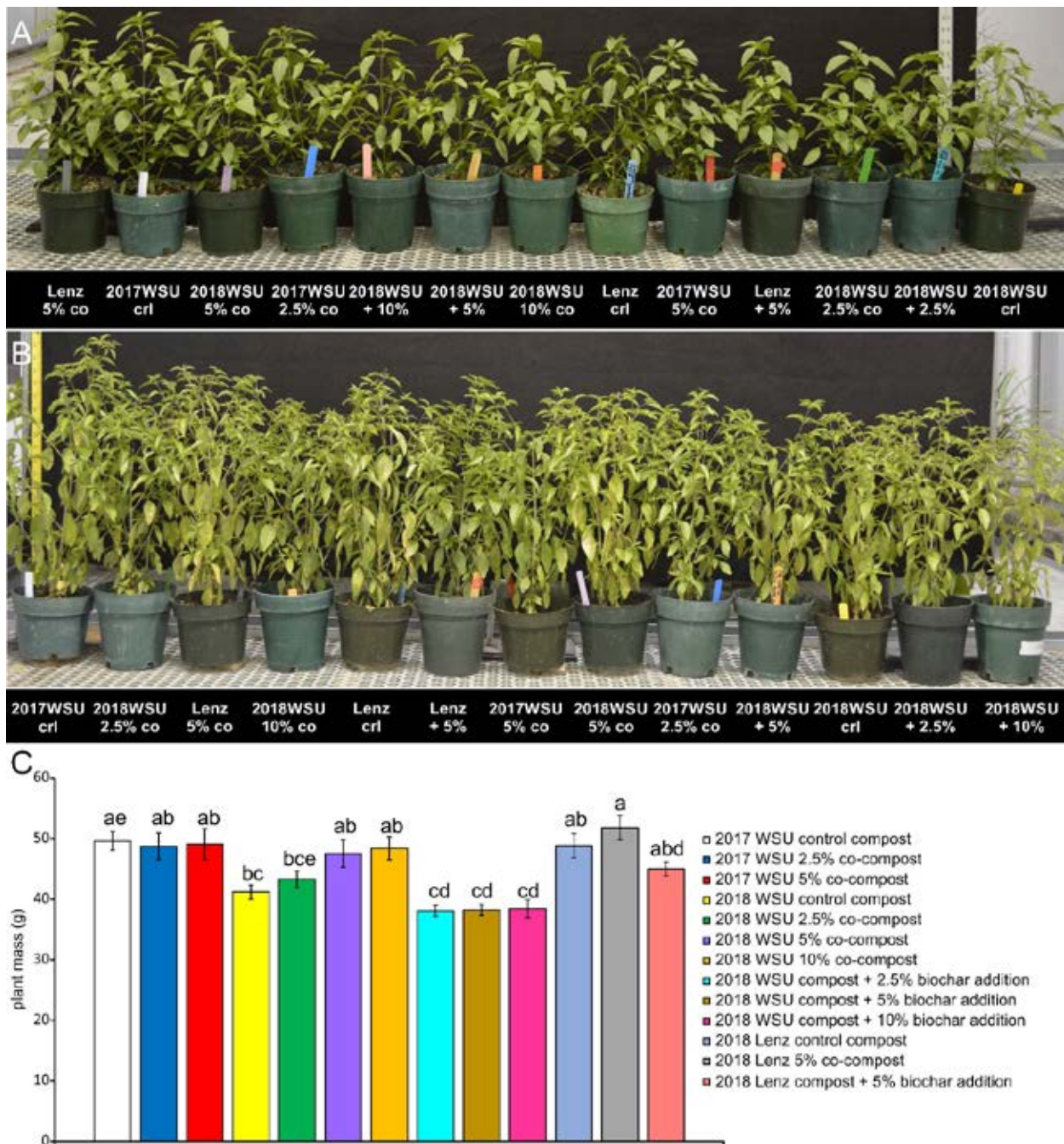
These results appear to be in contrast to the results of our 2017 study where the biomass yield of both Eleanora and TSQ plants grown with co-composted biochar (both at 5% and 2.5% level) was significantly higher in comparison to compost-only control (Gang et al., 2018). The same treatments (WSU 2017 alone, or co-composted with 2.5% or 5% Amaron Energy biochar) relied on the same growth media and seed, and would have been expected to behave similarly if not identically. One possible reason for the difference between the two experiments is the faster



growth observed in latest experiment following extended growth of the seedlings in the growth chamber compared to the first experiment. While the average plant mass for the different treatments was around 40 g or more, this value for the plants at time of harvest in the 2017 experiment was half as high, at 20 g or less. As seen on images of the tallest plants from each treatment, the order of the tallest plants, by treatment, is not the same in January as it is in February 2019 (Figure 36 and Figure 37, A and B). Although their absolute age in days was very similar, the plants were therefore developmentally older at harvest in the latest experiment. This led to the TSQ plants reaching the end of their typical lifespan in 2018/2019, leading to full inflorescence development, seed set and initiation of senescence. In the 2017, the plants were harvested prior to this developmental growth stage, at the time that they would be harvested for the fresh market. Thus, the growth rate differences caused by the different treatments that were observable in the 2017 experiment were masked in the second experiment because the plants were grown too long. The Eleanora plants had not yet reached full maturity at this same time point, and thus some differences related to treatment were still observable. The faster growth in last experiment is most likely related to the improved (trans)planting technology employed (growth in a warm and humid growth chamber until root systems were better established), which reduced the transplanting stress and led to earlier rapid growth. Replication of these experiments was definitely warranted to verify these conclusions.



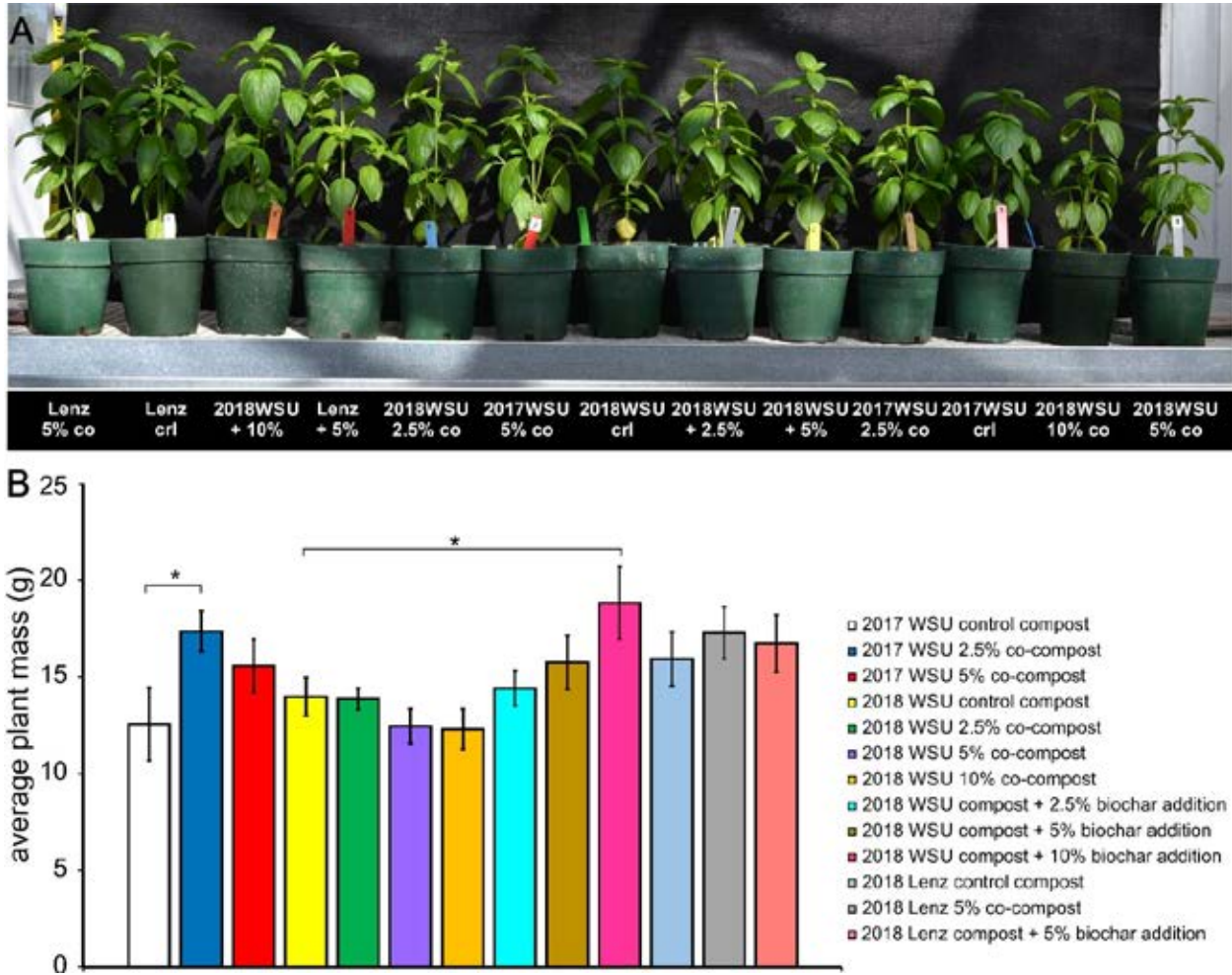
**Figure 36: The effect of biochar amendments on biomass production in greenhouse grown Eleanora sweet basil. Tallest plants from each treatment were photographed on A. 9 January 2019 and B. 4 February 2019. On 4 February, plants were lined up in the order of decreasing height C. Biomass of all plants for each treatment is shown as mean  $\pm$  s.e.m (n=10). In A and B, “+5%” indicates 5% biochar added after composting and “5% co” indicates 5% biochar co-composted (added before composting).**



**Figure 37: The effect of biochar amendments on biomass production in greenhouse grown Thai Siam Queen sweet basil. Tallest plants from each treatment were lined up in the order of decreasing height and photographed on A. January 9, 2019 and B. February 4, 2019. C. Biomass of all plants for each treatment is shown as mean  $\pm$  s.e.m (n=10). In A and B, “+5%” indicates 5% biochar added after composting and “5% co” indicates 5% biochar co-composted (added before composting).**

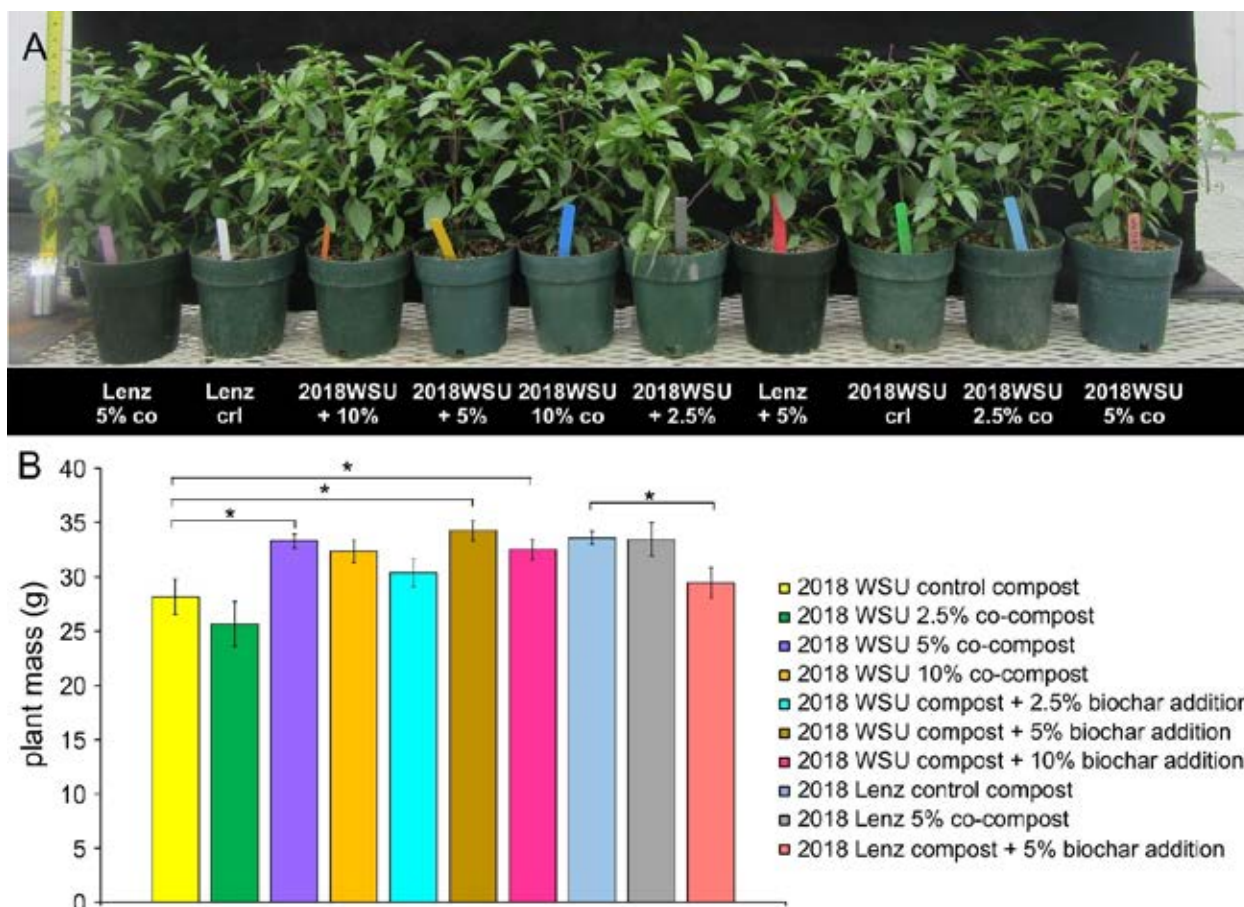
The experiments were therefore repeated with both basil cultivars in the 2019-2021 biennium, taking into account the faster growth and adjusting the harvest time to that in 2017. Eleanora basil plants were grown in September-October 2019. As in 2017, the average mass of individual plants

was less than 20g. In pairwise comparisons, the average mass of plants grown with WSU 2017 compost with 2.5 % biochar was significantly higher than that of control (Figure 38). No differences were observed for treatments with compost produced at Lenz. This is consistent with the observations in the previous trial. Of the treatments with WSU 2018 composts, the admixture of 10% biochar to mature compost resulted in significant biomass increase as compared to control. In the previous biennium, none of WSU 2018 composts significantly affected the yield.



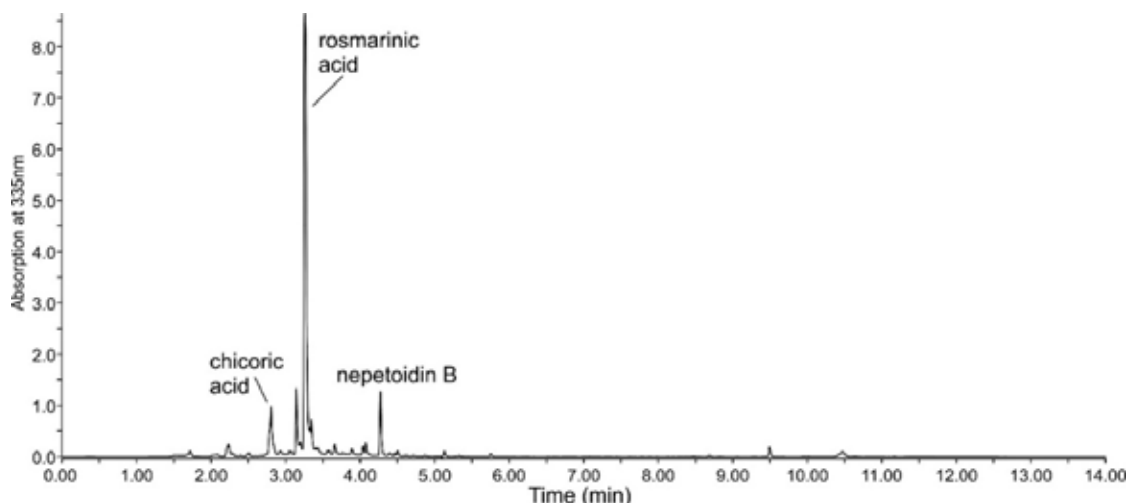
**Figure 38: The effect of biochar amendments on biomass production in greenhouse grown Eleanora sweet basil in 2019. A. Tallest plants from each treatment were lined up in the order of decreasing height and photographed on October 24, 2019. B. Biomass of all plants for each treatment is shown as mean  $\pm$  s.e.m (n=10). In A and B, “+x%” indicates x% biochar added after composting and “x% co” indicates x% biochar co-composted (added before composting).**

The greenhouse growth experiment was also repeated with Thai Siam Queen variety in 2021. Ten treatments were evaluated here due to depletion of WSU 2017 compost stock (Figure 39). Significantly higher biomass was produced by the cohorts treated with WSU 2018 compost amended with 5% biochar, both before and after composting process. Furthermore, WSU 2018 admixed with 10% biochar post-composting also yielded higher average biomass. Lenz compost admixed with 5% char resulted in significantly lower biomass accumulation.

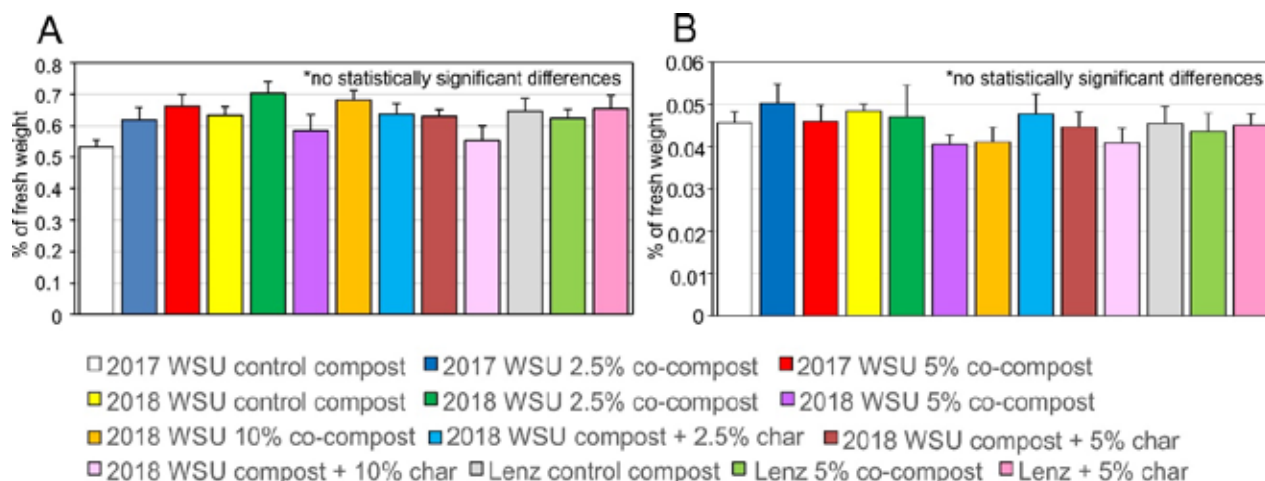


**Figure 39: A.** The effect of biochar amendments on biomass production in greenhouse grown Thai Siam Queen sweet basil in 2021. Tallest plants from each treatment were lined up in the order of decreasing height and photographed on April 30, 2021. **B.** Biomass of all plants for each treatment is shown as mean  $\pm$  s.e.m (n=10). In A and B, “+5%” indicates 5% biochar added after composting and “5% co” indicates 5% biochar co-composted (added before composting). Significant differences between the average weights are indicated by \* ( $p < 0.05$ , Student’s two-tailed t-test).

The accumulation of antioxidants was evaluated in Eleanora plants. The overall profile (Figure 40), and abundances of both major phenolics rosmarinic and chicoric acid (Figure 41) were similar to the levels measured in our earlier study (Gang et al., 2018). Using one-way ANOVA ( $p = 0.05$ ) as a statistical method, no significant differences were detected between the treatments (Figure 41), which is also in agreement with the earlier study. Compared to WSU 2017 control, accumulation of rosmarinic acid appeared to be higher with 5% co-composted biochar, while addition of Lenz compost with or without biochar resulted in slightly higher rosmarinic acid abundance. These results, now extended to three different compost types, confirm the notion that application of biochar, co-composted biochar, compost plus biochar and compost by itself have little impact on the specialized metabolism of sweet basil, but instead may have an influence on yield. These results are highly promising for the horticultural industry, suggesting that only positive benefits from biochar application may be expected for some crops, like sweet basil.



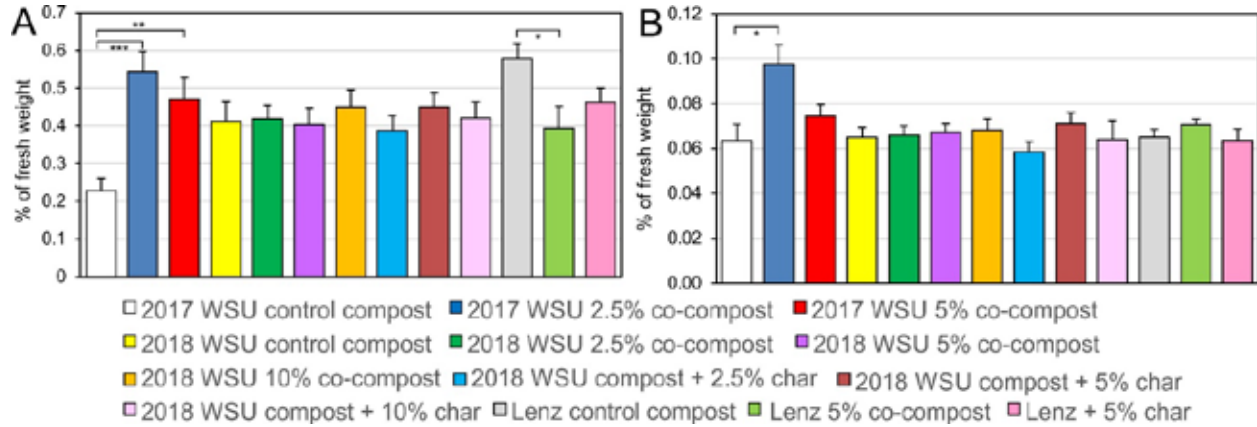
**Figure 40: Phenolics profile of Eleanora sweet basil. Methanolic extract from youngest mature leaves was analyzed using LC-PDA-MS and the profile visualized at UV 335±3 nm. Main phenolic compounds chicoric and rosmarinic acids were quantified for this report.**



**Figure 41: The effect of biochar amendments on antioxidant phenolics accumulation in Eleanora sweet basil. Contents of A. rosmarinic acid and B. chicoric acid in the two youngest mature leaves are presented as % of fresh weight. Note that “+5% char” indicates 5% biochar added after composting and “5% co-compost” indicates 5% biochar co-composted (added before composting). Shown data are means ±s.e.m. (n=10). Statistical differences were evaluated using one-way ANOVA (Tukey’s post-hoc HSD test, p<0.05).**

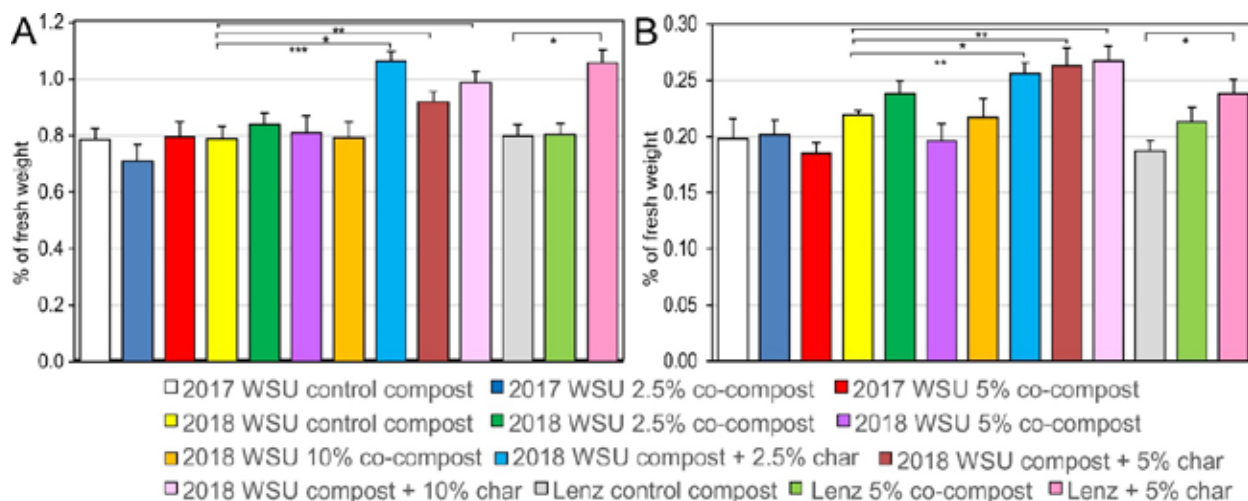
In the October 2019 replication of the study with developmentally younger Eleanora plants, the addition of WSU 2018 compost with any of the amendments did not result in significant differences between phenolics accumulation (Figure 42). However, the abundance of rosmarinic acid was elevated in plants grown with WSU 2017 compost that was co-composted with either 2.5% or 5% biochar. Notably, the levels of rosmarinic, but not chicoric acid in plants grown with WSU 2017 control compost were markedly lower than those of all other 12 treatments.

Qualitatively similar but statistically not significant reduction of rosmarinic acid accumulation in plants grown with WSU 2017 control compost was also observed in the previous trial (Figure 41A). By contrast, plants grown with Lenz control compost accumulated significantly higher levels of rosmarinic acid compared to those grown in soil with Lenz compost that was co-composted with biochar. There was no appreciable affect of any of the Lenz-based compost mixtures on chicoric acid levels (Figure 42 B). The production of chicoric acid was significantly higher in plants supplemented with WSU 2017 compost, co-composted with 2.5% biochar, as compared to its control group.



**Figure 42: The effect of biochar amendments on antioxidant phenolics accumulation in Eleanora sweet basil in Fall 2019 trial. Contents of A. rosmarinic acid and B. chicoric acid in the two youngest mature leaves are presented as % of fresh weight. Note that “+5% char” indicates 5% biochar added after composting and “5% co-compost” indicates 5% biochar co-composted (added before composting). Shown data are means  $\pm$ s.e.m. (n=10). Statistical differences between treatments and their respective compost-only controls were evaluated using pairwise two-tailed Student’s t-test (\*,  $p < 0.05$ , \*\*,  $p < 0.005$ , \*\*\*,  $p < 0.0005$ ).**

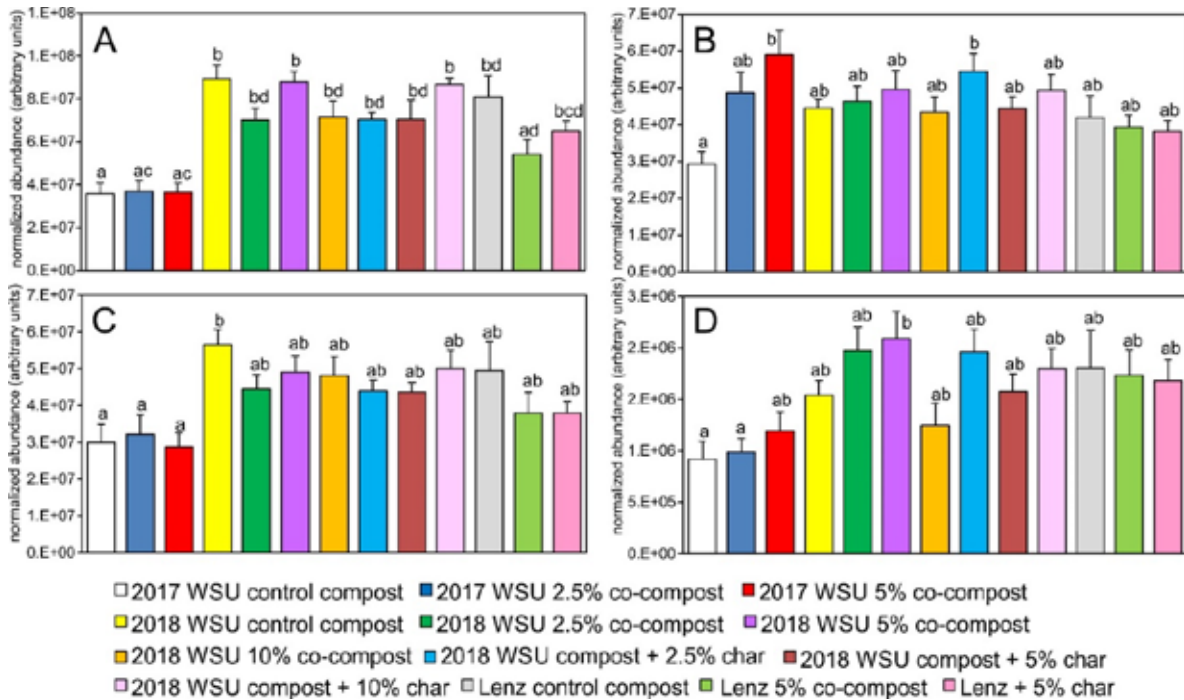
The abundance of the same major phenolics was also monitored in Thai Siam Queen basil in the 2017-2019 biennium. The results were quite different from those with Eleanora cultivar. Admixture of biochar to both WSU 2018 and Lenz composts led to significantly higher levels of both rosmarinic and chicoric acids (Figure 43). Conversely, WSU 2017 compost and its derived products did not have a significant effect on the accumulation of these phenolics.



**Figure 43: The effect of biochar amendments on antioxidant phenolics accumulation in Thai Siam Queen sweet basil in 2019 trial. Contents of A. rosmarinic acid and B. chicoric acid in the two youngest mature leaves are presented as % of fresh weight. Note that “+5% char” indicates 5% biochar added after composting and “5% co-compost” indicates 5% biochar co-composted (added before composting). Shown data are means  $\pm$ s.e.m. (n=10). Statistical differences between treatments and their respective compost-only controls were evaluated using pairwise two-tailed Student’s t-test (\*,  $p < 0.05$ , \*\*,  $p < 0.005$ , \*\*\*,  $p < 0.0005$ ).**

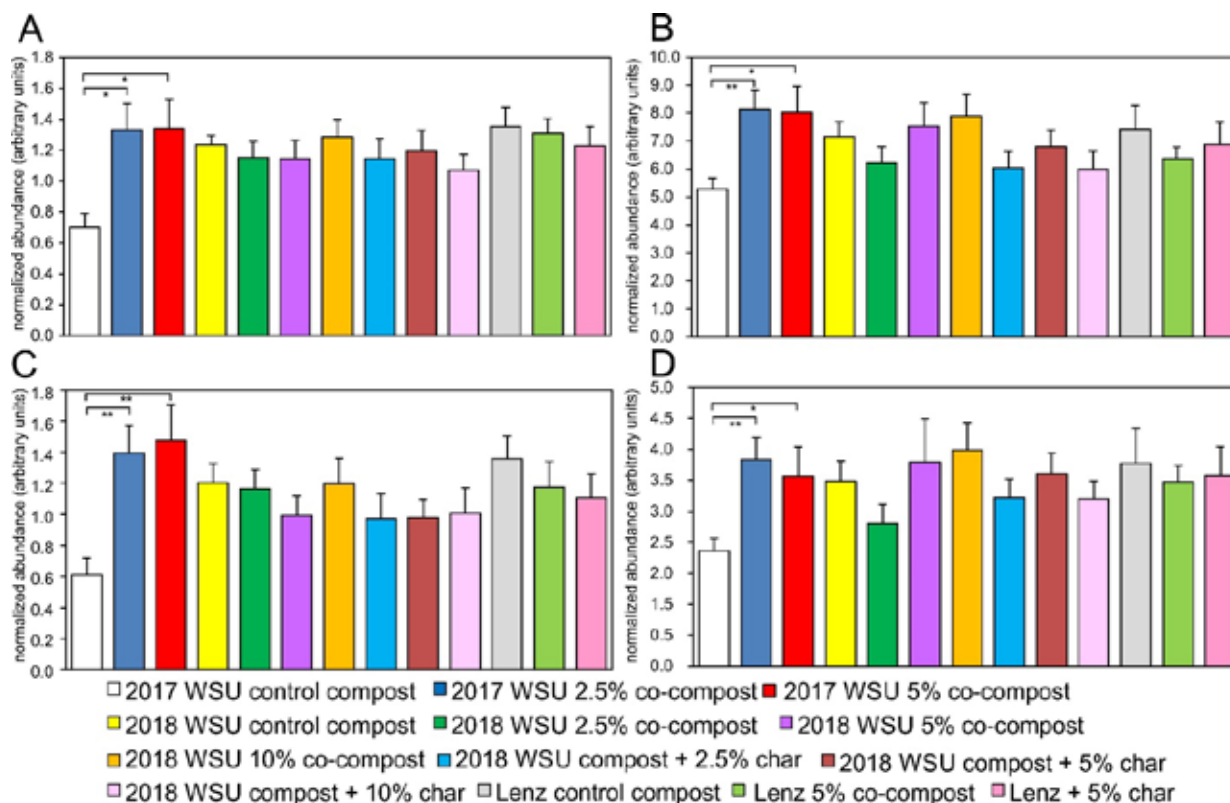
To evaluate the effect of biochar and compost treatments on the quality of Eleanora basil, volatile aroma compounds were also analyzed. The major components detected by gas chromatography mass spectrometry (GC-MS) were eucalyptol, linalool, eugenol, and ocimene. All plants exhibited similar overall profiles (not missing, and not having additional or alternative major components). Eugenol, a typical phenolic flavor component of basil leaves, and linalool, a monoterpene component of basil’s aroma, were less abundant in plants treated with amendments containing WSU 2017 compost (Figure 44 A,C). Viewed separately for each compost type (WSU 2017, or WSU 2018, or Lenz compost), the levels of these two compounds between treatments were not statistically different. As compared to WSU 2017 compost amendments, accumulation of eugenol was significantly higher in plants amended with WSU 2018 compost, WSU 2018, co-composted with 5% OBS biochar, and WSU 2018+10% OBS biochar. Linalool was significantly more abundant only in plants grown with plain WSU 2018 compost in comparison to all treatments involving WSU 2017 compost (Figure 44 C). The abundance of eucalyptol, another major monoterpene in basil leaves, was significantly higher in plants grown with WSU 2017 compost that was co-composted with 5% Amaron Energy biochar, in comparison to plants grown with the same compost without any biochar (Figure 44B). Accumulation of ocimene was significantly lower in WSU 2017 and WSU 2017, co-composted with 2.5% Amaron Energy biochar, in comparison to WSU 2018 compost, co-composted with 5% OBS biochar. The results obtained in this experiment are similar to the outcome of the study conducted in 2017, where no statistical differences were associated with different compost and biochar amendments (Gang et al., 2018). The observation of reduced eugenol, linalool, and ocimene abundance upon treatment with WSU 2017 compost and its derivatives should be reproduced for confirmation.





**Figure 44: The effect of biochar amendments on volatile aroma component accumulation in Eleanora sweet basil. Contents of A. eugenol, B. eucalyptol, C. linalool, and D. ocimene in the two youngest mature leaves are presented in arbitrary units, normalized to internal standard and fresh tissue weight. Note that “+5% char” indicates 5% biochar added after composting and “5% co-compost” indicates 5% biochar co-composted (added before composting). Shown data are means  $\pm$  s.e.m. (n=10). Statistical differences were evaluated using one-way ANOVA (Tukey’s post-hoc HSD test,  $p < 0.05$ ).**

The Eleanora plants harvested in October 2019 were also monitored for their volatile aroma accumulation. Notably, plants grown with WSU 2017 compost amended with biochar accumulated significantly higher levels of the four evaluated aroma compounds compared to respective WSU 2017 compost-only plants (Figure 45), with the levels observed in WSU 2017 control compost treated plants being moderately to significantly below average as compared to all other treatments. This is unlike the results from the harvest in February 2019 (Figure 44), but similar to the accumulation of rosmarinic acid in those treatments (Figure 43 A). It is worth remembering that the biomass of plants in WSU 2017 control compost treatment was somewhat reduced compared to average values of other treatments, yet not dramatically so. The considerable number of replicates (n=10) used in this study increases the confidence in these results. Overall, these observations might indicate the presence of some components modulating specialized metabolism in the WSU 2017 control compost, which might be inactivated or removed by the addition of biochar. The discrepancy between the two trials, plants harvested in February vs October 2019, may also have led to the experimental differences. Among details to consider is the plant development stage as well as the age of compost.



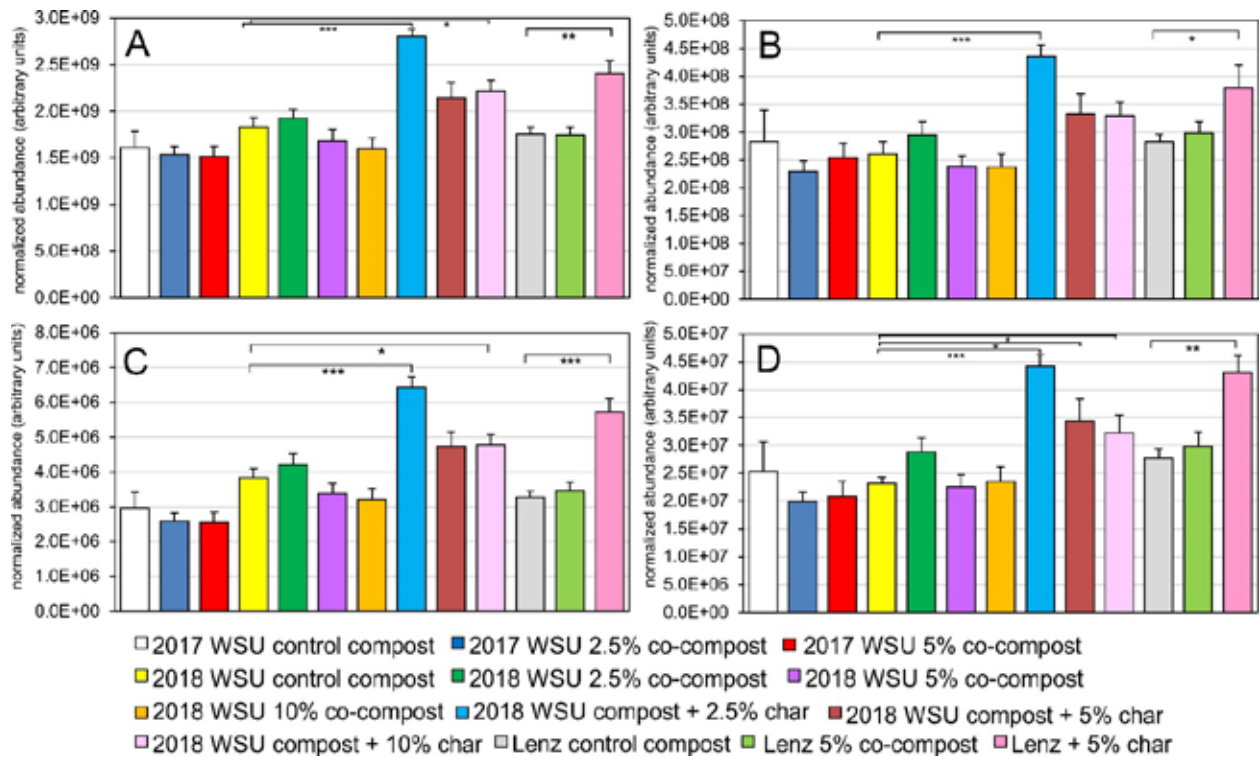
**Figure 45: The effect of biochar amendments on volatile aroma component accumulation in Eleanora sweet basil. Contents of A. eugenol, B. eucalyptol, C. linalool, and D. ocimene in the two youngest mature leaves are presented in arbitrary units, normalized to internal standard and fresh tissue weight. Note that “+5% char” indicates 5% biochar added after composting and “5% co-compost” indicates 5% biochar co-composted (added before composting). Shown data are means  $\pm$  s.e.m. (n=10). Statistical differences were evaluated using two-tailed Student’s t-test (\*, p<0.05).**

The Thai Siam Queen cultivar of sweet basil accumulates estragole, eucalyptol, methyl eugenol, and ocimene as major components of essential oil. Their production in response to compost and biochar amendments was monitored in the 2019 greenhouse trial. As seen for phenolics (Figure 45), the levels of specialized metabolites were increased in plants grown in soil amended with WSU 2018 compost with admixed biochar, where 2.5% admixture resulted in the highest accumulation increase (Figure 46). The levels of the four evaluated major aroma volatiles were also significantly higher in plants grown with Lenz compost admixed with 5% biochar, behavior that was also observed for phenolics.

### Major conclusions from the different basil greenhouse trials:

These results with basil demonstrate biochar can have a very positive impact on plant productivity when added to compost, particularly when it is co-composted. On the other hand, specific experimental conditions, such as time of year of the experiment (and associated differences in light quality and/or ambient humidity in our greenhouses), can confound the impact of the compost+biochar or co-composted biochar effects. Different composts from different facilities or generated using different methods, or generated in different years, can also

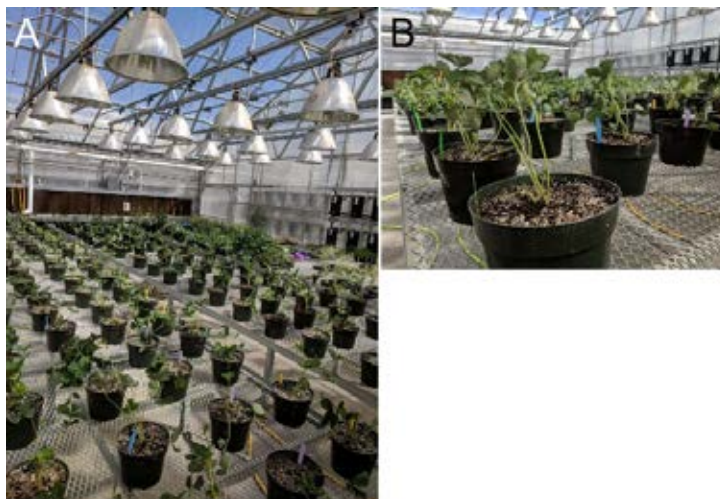
affect the results. This was clearly seen when comparing results from 2017 and 2018 composts generated at WSU and when comparing the Lenz-produced composts. These all had slightly or greatly different compositions. It is clear that biochar can have a very positive effect on plant productivity, while having a limited effect on plant quality (as observed for the chemical compositional analysis results), but it can also have a large effect, all depending on the particular compost/biochar/growth condition interaction. It is also clear that time of sampling (for the plants that grew “too long”) can have a large impact on the results. For crops where specific timing is essential to get the best quality crop, such as young sweet Eleanore basil, biochar can have a very positive impact on overall yield. When the crop is grown much longer, to the point of being overgrown, as occurred in one year of these trials, then such differences appear to disappear. Thus, biochar addition appears to have an impact on growth *rate*, not on overall growth *capacity*. And the timing of the harvest relative to that rate appears to have an impact on whether crop quality will be affected or not. Finally, these results suggest that important factors are generated when biochar is present that the plants respond to, which affect growth and other properties, which opens interesting avenues for research into the underlying mechanisms.



**Figure 46: The effect of biochar amendments on volatile aroma component accumulation in Thai Siam Queen sweet basil. Contents of A. estragole, B. eucalyptol, C. methyl eugenol, and D. ocimene in the two youngest mature leaves are presented in arbitrary units, normalized to internal standard and fresh tissue weight. Note that “+5% char” indicates 5% biochar added after composting and “5% co-compost” indicates 5% biochar co-composted (added before composting). Shown data are means  $\pm$  s.e.m. (n=10). Statistical differences were evaluated using two-tailed Student’s t-test (\*,  $p < 0.05$ , \*\*,  $p < 0.005$ , \*\*\*,  $p < 0.0005$ ).**

### 4.2.3 Strawberry greenhouse trial

Commercial ‘strawberries (Albion’) were grown in pots in the greenhouse (Figure 47). Half of plants were fertilized, and both fertilized and non-fertilized plants were subjected to 8 treatments testing WSU 2017 and Lenz composts and Oregon Biochar Solutions biochar.

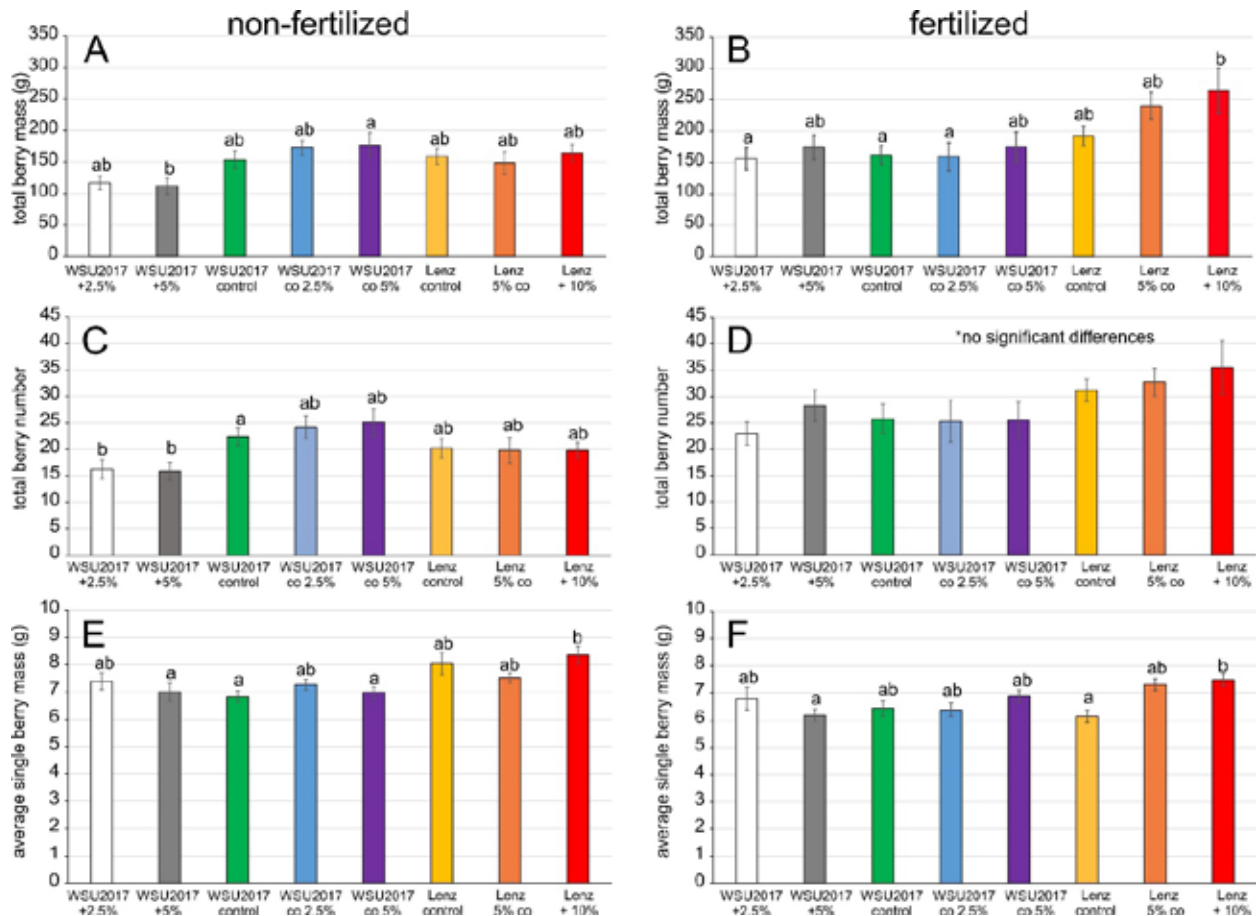


**Figure 47: ‘Albion’ strawberries were grown potted in the greenhouse. Half of the plants were fertilized. All plants were subjected to overall 8 biochar-compost treatments with WSU 2017 and Lenz composts. A. overview. B. close up of a plant.**

The results obtained from the two cohorts (fertilized and non-fertilized) were not identical (Figure 48). In the non-fertilized group, the difference was only significant between two treatments (Figure 48A). Cumulative berry mass collected from plants grown with WSU 2017 compost that was co-composted with 5% biochar was greater than when the same amount of biochar was added after composting. However, neither of these two treatments differed significantly from the corresponding compost-only yields. None of the treatments with Lenz compost had statistically significant effects on cumulative berry mass yield. Total berry number from plants grown with WSU 2017 compost plus biochar (both 2.5% and 5%) was significantly lower in comparison to the compost-only control (Figure 48C). In the fertilized group, supplementation with Lenz compost plus 10% biochar resulted in significantly higher total berry mass as compared to three other treatments (WSU 2017 compost alone or with 2.5% co-composted or biochar added after composting) (Figure 48B). The difference was not significant in comparison to other amendments with compost generated at Lenz Enterprises. The cumulative berry number did not vary significantly between treatments (Figure 48D). For both fertilized and non-fertilized groups, the average single berry mass from plants grown with Lenz compost plus 10% biochar was significantly higher in comparison to several other treatments (Figure 48E,F).

The interaction between additional fertilizer/plant nutrition and amendment with biochar and compost are in agreement with the study by De Tender et al. (2016b) who reported that inorganic fertilizer and lime added to growth substrate (white peat) attenuated the beneficial effects of compost-biochar mix. In other studies, the benefits of biochar were more pronounced in nutrient-poor soils when co-administered with fertilizer, in agreement with the role of biochar as moisture and nutrient retention agent (Yamato et al., 2006; Van Zwieten et al., 2010; Schulz et al., 2013;

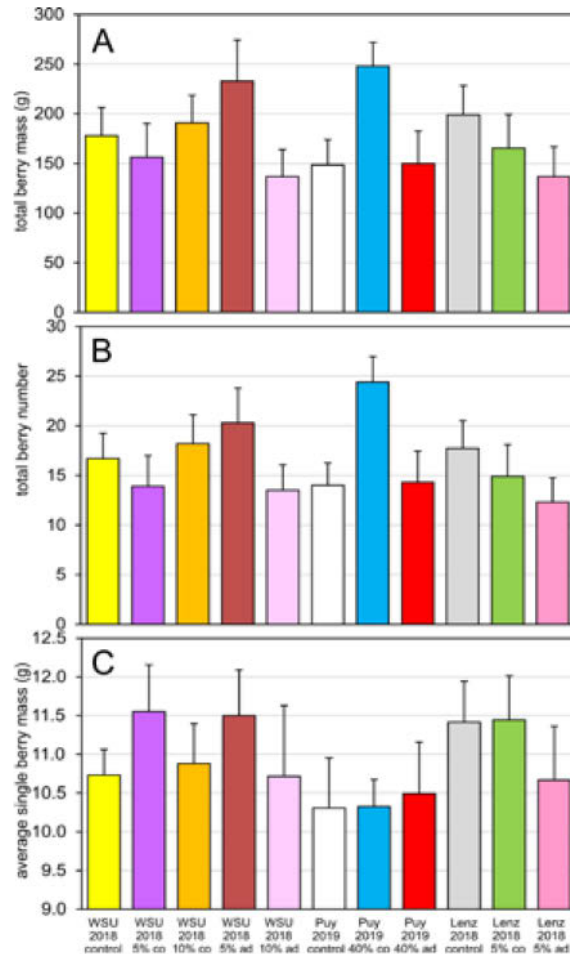
Agegnehu et al., 2015). Bird et al. (2012) concluded that biochar addition was more efficient in nutrient-poor as compared to relatively rich soils.



**Figure 48: Effect of compost and biochar amendments on productivity of Albion strawberries. Two cohorts of plants (fertilized and non-fertilized) were subjected to 8 treatments. Fruit was harvested as it ripened over 22 harvests from each plant. Total berry mass (A and B) and berry number (C and D) per plant were recorded. Average single berry mass (E and F) was calculated from the totals. In these figures, “+5%” indicates 5% biochar added after composting and “5% co” indicates 5% biochar co-composted (added before composting). Results are means  $\pm$  s.e.m. (n=10). Different letters indicate significant difference between means, calculated by one-way ANOVA with Tukey post-hoc tests ( $p < 0.05$ ).**

In 2019-2021 biennium, a greenhouse trial with the same strawberry cultivar was reproduced with a modified set of composts which included two “Puy” composts from Dr. Doug Collins (WSU Puyallup): a control and a co-compost with 40% OBS biochar, described in section 3.3 of this report. An admixture with 40% OBS char was prepared by the IBC greenhouse staff for comparison. This time, all plants were fertilized as this is a common practice in commercial farming. Harvest time was September-December 2020. In contrast to harvest conducted in 2018, yields from individual plants varied strongly, resulting in large variation of average fruit number and total yield per plant in all treatments. Several treatments included plants that produced no fruit over the harvest period, and in other treatments the difference in total strawberry number over the harvest period ranged from 18 to 34 (average, 27.7 fruit difference between least and

most productive plants). As a consequence, none of the observed differences of the average numbers is statistically significant (Figure 49).

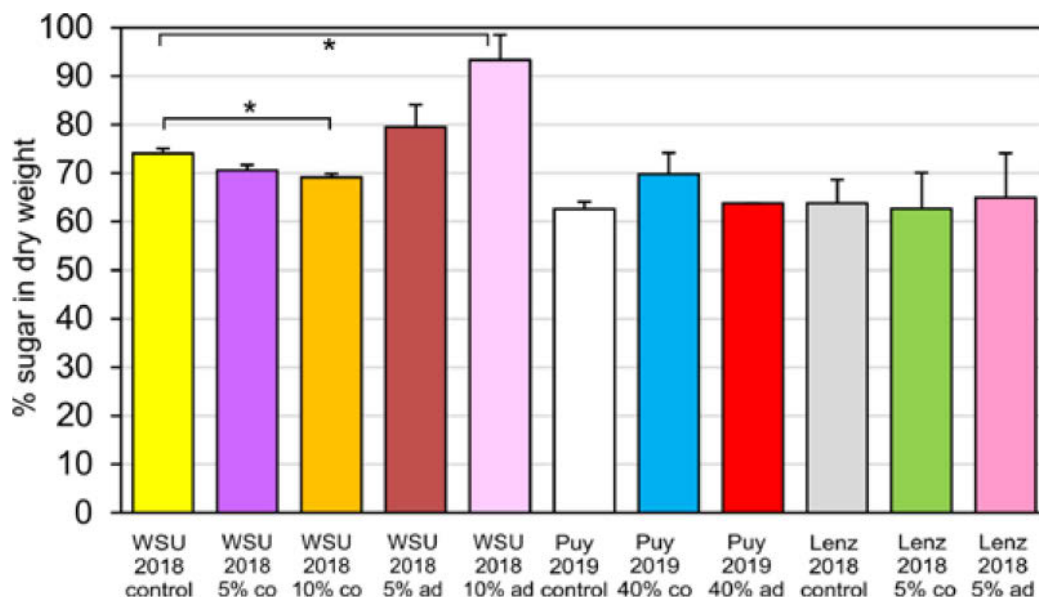


**Figure 49: Effect of compost and biochar amendments on productivity of Albion strawberries. Plants were subjected to 11 treatments. Fruit was harvested as it ripened over 16 harvests from each plant. Total berry mass (A) and berry number (B) per plant were recorded. Average single berry mass (C) was calculated from the totals. In these figures, “+5%” indicates 5% biochar added after composting and “5% co” indicates 5% biochar co-composted (added before composting). Results are means  $\pm$  s.e.m. (n=10).**

To further evaluate the differences potentially caused by the treatments, we analyzed selected chemical properties of the fruit. Fruit was freeze-dried and ground using a ball mill, and the powder extracted for analysis by LC- and GC-MS as well as sugar content analysis via refractometry. Due to processing complications, only 4-6 replicates per treatment were successfully analyzed, while treatment “Puy 40% ad” was reduced to one sample and was therefore not amenable to statistical analysis.

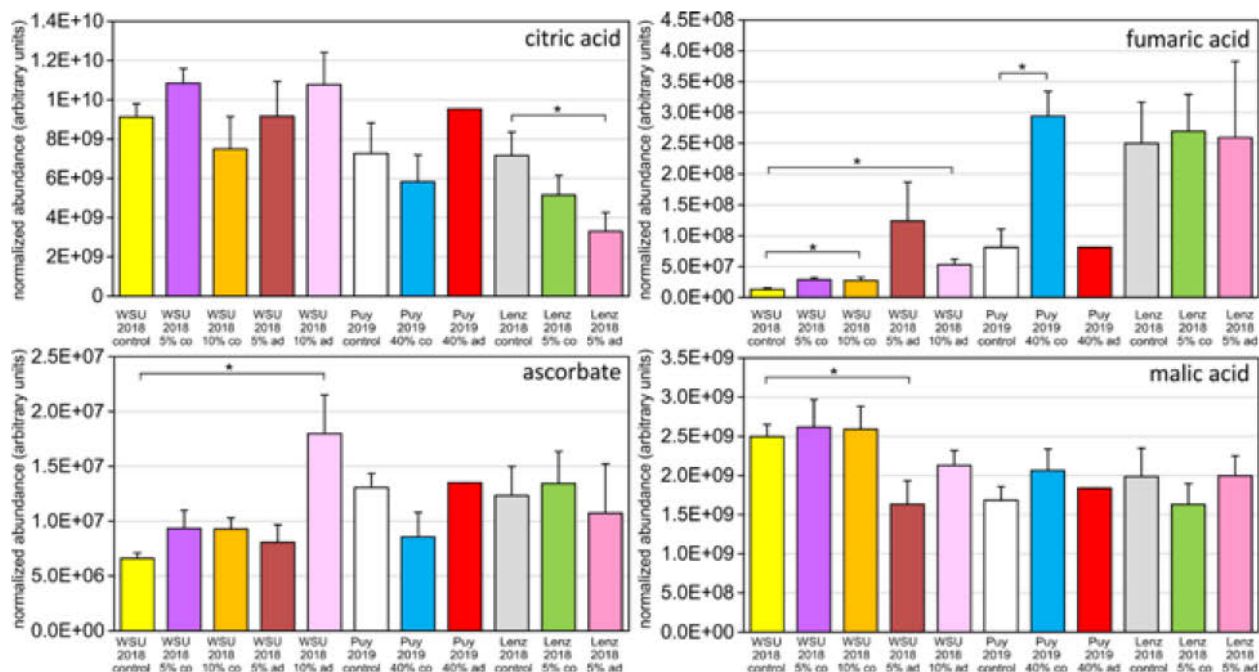
The total sugar content evaluation suggested that fruit from plants grown with admixture of WSU 2018 compost with 10% biochar had higher levels of sugars (Figure 50). While not a large difference in absolute values, sugar concentration in plants treated with WSU 2018 compost co-

composted with 10% of the same biochar was significantly lower. An important caveat is that strawberries collected for this analysis differed in size and therefore weight proportion of the achenes vs receptacle. Furthermore, the degree of ripeness of individual fruit could have had a significant impact on the results. A more careful selection of fruit for chemical analysis may produce more consistent results.



**Figure 50: Sugar content of freeze-dried and powdered strawberries, as determined in an aqueous extract using a handheld refractometer. Values shown are means  $\pm$  s.e.m. (n=4-6, treatment “Puy 2019 40% ad, n=1 due to tissue processing problem). \* indicates significant difference between the means of two treatments, as determined by two-tail Student’s t-test (\*,  $p < 0.05$ ).**

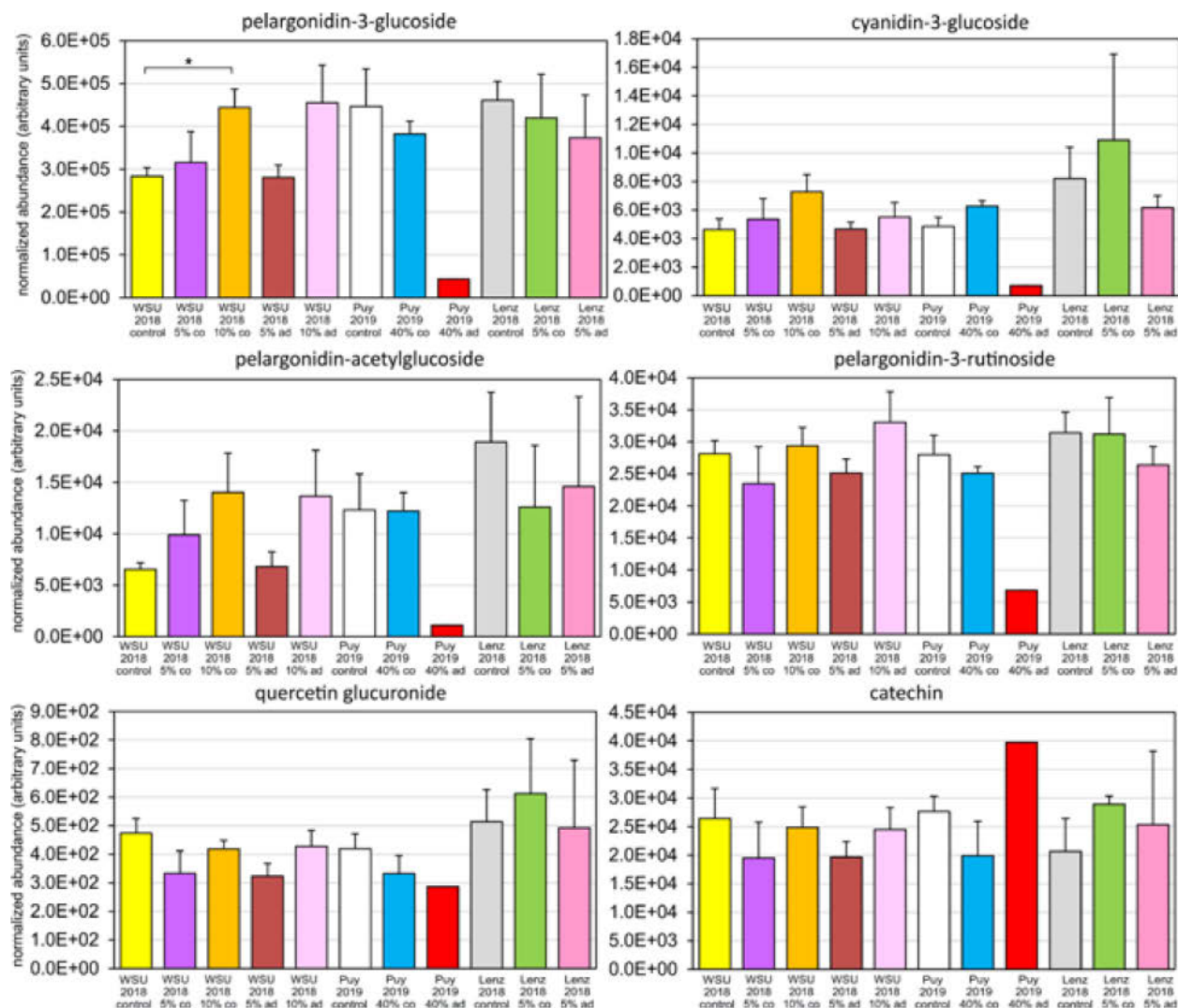
Aside from the sugar content, strawberry flavor is modulated by the small organic acids, a blend of flavor compounds, and a fine balance between all of the contributing components. An evaluation of several abundant acidic compounds suggested that WSU 2018 compost with 10% admixed, or, to a lower extent, co-composted biochar resulted in increased levels of fumaric acid (Figure 51). Fumaric acid abundance was also increased in plants grown with Puyallup 2019 compost, co-composted with 40% biochar. An increased accumulation of ascorbate was recorded in plants grown with WSU 2018 compost, admixed with 10% biochar. Decreased abundances of citric and malic acid were found in plants amended with Lenz 2018 compost, admixed with 5% char, and in those amended with WSU 2018 compost, admixed with 5% char, respectively.



**Figure 51: Effect of compost and biochar amendments on small organic acid accumulation of Albion strawberries. Plants were subjected to 11 treatments. Fruit for analysis was harvested at the end of the season (late November – early December 2020). In these figures, “x% ad” indicates x% biochar added after composting and “x% co” indicates x% biochar co-composted (added before composting). Results are means  $\pm$  s.e.m. (n=4-6, treatment “Puy 2019 40% ad, n=1 due to tissue processing problem). Significant difference between the means is indicated by \* (p<0.05, two-tailed Student’s t-test).**

Further metabolites that are relevant for strawberry quality are anthocyanins, which contribute to the coloration of the fruit. Candidate signals for the four most abundant anthocyanins as reported by Lopes-da-Silva et al. (2002) as well as most abundant related flavonoids, catechin and quercetin-3-glucuronide (Fait et al., 2008) were identified in extracts from dried strawberries. A comparison across treatments revealed the levels of pelargonidin-3-glucoside, the most abundant anthocyanin in the analyzed tissue, were significantly higher in plants grown with WSU 2018 compost, co-composted with 10% biochar (Figure 52). While there was variation between the abundances of other evaluated flavonoids, none of them were statistically significant. It is worth mentioning that, while the abundance of anthocyanidins in “Puy 2019 40% ad” treatment is dramatically lower, the treatment was represented by a single sample whose analysis may have been compromised. A repetition with a larger number of replicates is necessary for conclusions.





**Figure 52: Effect of compost and biochar amendments on flavonoid accumulation of Albion strawberries. Plants were subjected to 11 treatments. Fruit for analysis was harvested at the end of the season (late November – early December 2020). In these figures, “x% ad” indicates x% biochar added after composting and “x% co” indicates x% biochar co-composted (added before composting). Results are means  $\pm$  s.e.m. (n=4-6, treatment “Puy 2019 40% ad, n=1 due to tissue processing problem). Significant difference between the means is indicated by \* ( $p < 0.05$ , two-tailed Student’s t-test).**

#### 4.2.4 Strawberry field trial

Strawberry yields in 2018, measured as cumulative production over the course of one season ( $\text{kg ha}^{-1}$ ), did not increase in response to fertilizer applications, nor were they affected by any of the surface applied soil amendments (Table 14; 2018, Collection 1). When cumulative yield was measured from the same research plots after a second growing season, however, comparisons among amendments revealed significant differences between treatments which were observed between plots receiving the highest rates of amendment (compost and co-compost) and those receiving biochar (Table 14; 2018, Collection 2). Unfortunately, for the 2019 established planting, yield data were inconclusive; the first collection was damaged by geese and the second

collection was adversely affected by conditions resulting from the COVID-19 pandemic. Means from the unfertilized portion of the 2019 collection 2 are presented in Table 14 for illustrative purposes only.

Strawberry yield responses vary due to fertilization (Bottoms et al., 2013), and organic amendment (Jay et al., 2015; Vestberg et al., 2008), and therefore the insignificant responses in collection 1 (2018 planting) are not completely unexpected. That year, our cumulative yield data were collected from newly established plantings and as Latrou et al. (2013) observed, for fertilization to have a notable effect on cumulative strawberry yield, two full growing seasons may be necessary. In addition, in 2018, strawberry bare root plugs were planted in July, when soil and air temperatures are elevated. Considering temperature plays an important role in flower development, and thus fruit production (Manakasem and Goodwin, 2001), our observations may reflect the effects of this environmental condition rather than the non-effects of fertilizer or soil amendment. For collection 2 (2018 planting), the significant differences between our two compost treatments and the biochar amended plots likely reflects differences in N mineralization. N mineralization is the mechanism that provides plant available soil N, and organic materials, including composts, mineralize at variable rates dependent upon soil moisture, temperature and the physicochemical properties of the starting material (e.g., C/N ratio). The high C/N ratio of the biochar material (e.g., 115) indicates that less soil N will be available for plant uptake, while the low C/N ratio of the compost materials (e.g., 15) indicates that plant available soil N will be available. The timing of that release (mineralization), however, depends on soil temperature and moisture, among others, and the delayed response (into the second year) we observed here is not surprising.

**Table 14: Mean values for cumulative strawberry yield from two consecutive collections in each of two establishment years, 2018 and 2019. Note that the lack of data for all collections is due to geese damage, and COVID-19 influenced challenges.**

	2018 established planting				2019 established planting	
	Collection 1 <sup>‡</sup>		Collection 2 <sup>‡</sup>		Collection 2 <sup>‡</sup>	
Treatment*	Fert	Unfert	Fert <sup>†</sup>	Unfert	Fert	Unfert
Bio	-**	-	18391b	-	-	11568
Comp	-	-	21053a	-	-	13975
Co-C	-	-	21185a	-	-	12106
C+B	-	-	19956ab	-	-	12343
NoA	-	-	19684ab	-	-	11474

\*Treatment labels are as follows: Bio is Biochar; Comp is Compost; Co-C is Co-Compost; C+B is Compost + Biochar; NoA is No Amendment.

\*\*Dashes indicate no significant difference.

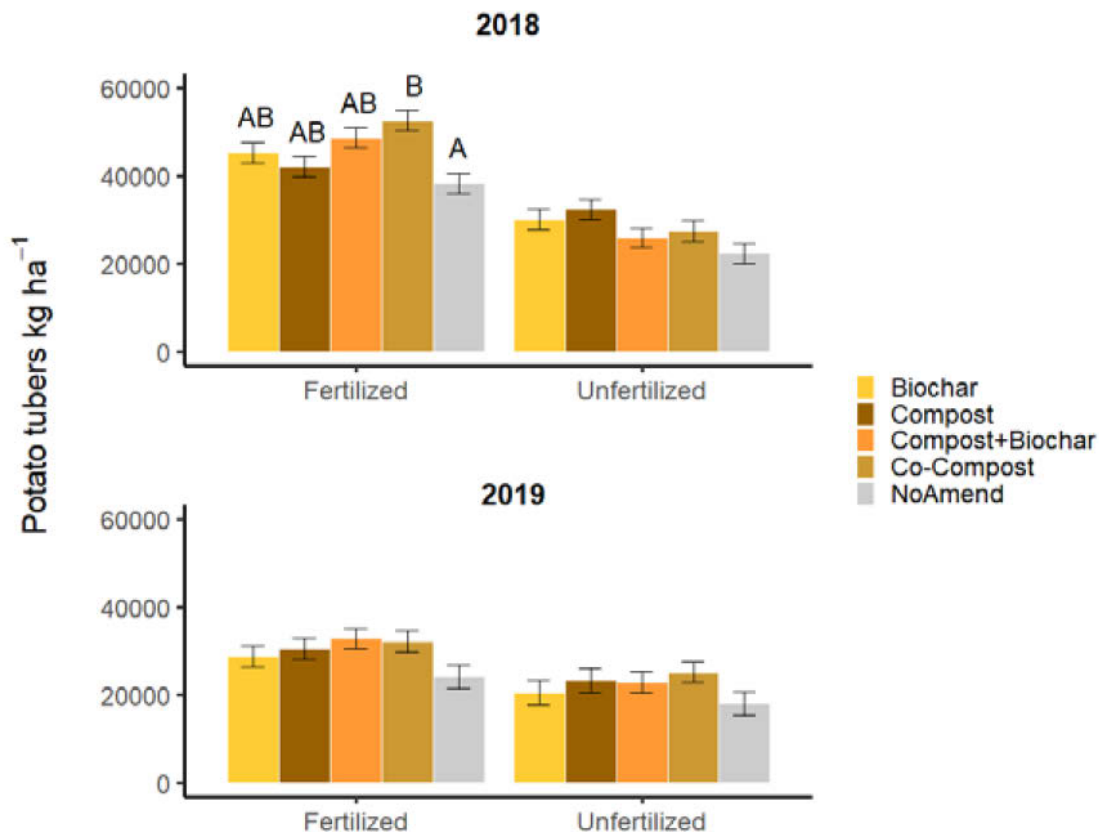
<sup>†</sup>Mean values represent main effects of amendment, those followed by different letters are significantly different according to Tukey's HSD test  $p < 0.05$ .

<sup>‡</sup>Collection 1 yield was affected by animal damage, means are presented for illustrative purposes. Collection 2 was adversely affected by the COVID-19 pandemic.

#### 4.2.5 Potato field trial

Potato yields, in contrast to strawberry yields, were significantly affected by fertilizer treatment in 2018 and 2019 (Figure 53), and average yields in fertilized plots increased 113% in comparisons with the unfertilized split-plots. Recent research has indicated tuber yield (across several varieties) responds quadratically to increasing rates of supplementary N additions, where the upper limits of tuber response plateau between 350 and 400 kg N ha<sup>-1</sup> (Sun et al., 2019). Research plots in this study received 108.6 kg N ha<sup>-1</sup> in total, well below the suggested upper limit, and therefore tuber yield increases following N additions are consistent with these observations.

Significant differences within the amendment factor were detected in 2018 but not in 2019, and these differences were only observed in comparisons between the unamended and co-compost amended, fertilized plots (Figure 53, top). No clear trends were revealed in potato tuber weights and following amendment for 2018 or 2019. Crop yield observations following co-compost amendment, like the one used here, are highly variable. For example, Shulz et al. (2013) observed improved oat yields (*Avena sativa* L.) in two different soils at varying rates of composted biochar, while Wang et al. (2017) reported significantly diminished yields in rapeseed (*Brassica napus* L.) and lettuce (*Lactuca sativa* L. 'longifolia'), which were grown in pots with and without a composted biochar amendment. Where crop yields are improved, the addition of a composted biochar is thought to influence soil nutrient availability directly, as a source of macro- and micronutrients (Shulz et al., 2013), or indirectly, by altering the native soil physicochemical properties (Hagemann et al., 2017). It is unclear from our observations which process is dominant here, but it is possible that both, additional nutrients and changes in soil chemical cycling, are influencing nutrient availability and thus, crop yields.



**Figure 53: Potato yield (tuber weight) at Mount Vernon as affected by fertilizer and amendment in 2018 (top) and 2019 (bottom). Bars capped by different letters are significantly different according to Tukey's HSD test  $p < 0.05$ .**

Weed cover was measured in the potato trial to determine whether the biochar amendment would impact herbicide efficacy. However, there were no differences in the treatments overall, with excellent weed control in all the amended treatments (2018 only).

## 4.3 Impact of amendment on soil physicochemical properties

### 4.3.1 Bulk density

In Mount Vernon soils, significant bulk density reductions were observed in plots receiving the largest quantity of material (Table 15). Plots receiving compost or co-composted amended soils had bulk density reductions of 7.5% and 5.7%, respectively, when compared to unamended plots (Table 15). In Puyallup soils, no significant effects were observed following amendment in either year, 2018 or 2019 (Table 15). While our observations at Mount Vernon revealed small decreases in bulk density, severe *increases* in bulk density, often the result of mechanical compaction (Hamza et al., 2011), are correlated with reductions in crop yield (Batey and McKenzie, 2006). In addition, native soil organic matter is known to influence bulk density

measurements (Arvidsson, 1998), because Puyallup soils are greater in organic matter (data not shown), the rates at which we surface amended these soils may not have resulted in observable differences between amendment types or between amended and unamended research plots at Puyallup.

**Table 15: Mean bulk density measurements (g cm<sup>-3</sup>) following amendment in Puyallup and Mount Vernon soils for 2018 and 2019. Within a location (e.g., Mt. Vernon) and year, values followed by different letters are significantly different according to HSD test, p<0.05.**

Treatment	Puyallup		Mt. Vernon	
	2018	2019	2018	2019
Biochar	1.11	1.24	1.18ab	1.15
Compost	1.12	1.21	1.13b	1.12
Co-Compost	1.14	1.24	1.15b	1.12
Compost + Biochar	1.13	1.25	1.16ab	1.12
No amendment	1.12	1.23	1.22a	1.13

#### **4.3.2 Total carbon and total nitrogen in whole soil and particulate organic matter fractions**

Surface-applied amendments in unfertilized Puyallup soils had no effect on total C and N concentrations in 2018 (Table 16). In 2019, total C was significantly increased in all treatments but only in comparisons with the unamended control, while total N mean values were significantly greater in the two compost treatments (compost and co-compost) and in comparisons with all other treatments (Table 16). In the POM fraction and in 2018, the significant differences detected followed a general pattern: the lowest values were observed in biochar and unamended soils and increased in treatments that included compost material (Table 16). For the POM fraction total C, significantly higher values were observed following co-compost and compost plus biochar treatments, whereas in POM fraction total N the largest values were observed in compost alone, co-compost, and compost plus biochar treatments, respectively (Table 16). Conversely, in 2019, no clear trends were observed. For example, in POM fraction total C, no differences were detected between any of the treatments, except in comparisons with the control, while in POM fraction total N, mean values in plots receiving the two compost treatments (compost and co-compost) were significantly greater in comparisons with all other treatments (Table 16).

In the first year of our study (2018), POM fraction total C and N were modestly sensitive to, and reflected, the extent of biochar conditioning revealed in the three biochar treatments: biochar alone, co-composted biochar, and compost plus biochar mixed and applied in the field. As a soil amendment, biochar is known to alter soil chemical properties owing to physicochemical properties that include a highly reactive surface (Liang et al., 2006). Recently, Hageman et al. (2017) identified a nutrient-rich organic coating that results from co-composting and attributed increases in soil chemical reactivity to this layer. The differences we observed in 2018 may reflect the varying degrees of biochar surface coating that could influence the rate at which soil chemicals are exchanged between soil surfaces and solution. In 2019, however, this mechanism

was less clear and only partially reflected in the POM fraction N observations and to a lesser extent in the total N mean values. This may have something to do with the “age” of our products as all of our amendment materials were stored for one year following use in the first-year trial. Because biochars and co-composted biochars are known to change in time (Hestrin et al., 2020; Kim et al., 2021), our second year responses may reflect these physicochemical changes.

Conversely, amendments in Mount Vernon soils significantly affected total C and N in whole soil and POM fractions for 2018 and 2019 (Table 16). In the whole soil fraction, total C concentrations were greatest in compost amended plots (compost and co-compost) and lowest in the biochar and unamended plots and this was generally true in 2018 and 2019 (Table 16). In 2018, total N concentrations for whole soil were less variable than total C, and the greatest difference between total N concentrations were observed between biochar and compost alone amended plots. In 2019 the greatest difference was observed between co-compost and biochar alone amended plots, and we observed greater separation between treatment comparisons (Table 16). In the POM fraction, total C and N concentrations were lowest in the biochar and unamended plots, moderate in plots receiving the compost plus biochar field mix, and greatest in compost alone and co-compost treated soils. This was also true for 2018 and 2019, except for POM fraction total C values in biochar amended soils for 2019, which were somewhat moderate in comparisons with all other treatments (Table 16).

The greater overall amendment effect in these soils may represent a quality difference between Puyallup and Mount Vernon soils, as organic amendments are known to produce greater effects in soils where organic matter is depleted (Medina et al., 2015). This is supported, in part, by the significant differences observed in whole soil total C and N (Table 16). Similar to Puyallup soils, the POM fraction total C and N appear, in part, to reflect the biochar conditioning (i.e., composted or not) revealing subtle differences between amendments. The more obvious difference, however, is in the quantity of material applied: compost and co-composted amendments were applied at the highest rate of material (Table 8) and therefore the observed differences at Mt. Vernon and Puyallup also reflect the additional material applied to the soil environment.

**Table 16: In Puyallup (Puy.) and Mount Vernon (Mt. V.) soils, mean values for total and POM (particulate organic matter) C and N (Mg ha<sup>-1</sup>) in unfertilized whole soils and POM fractions as affected by amendment for 2018 and 2019. Within a variable (e.g., C), site, and year, values followed by different letters are significantly different according to Tukey's HSD test, p<0.05.**

Treatment	C		N		POM C		POM N	
	Puy.	Mt.V.	Puy.	Mt.V.	Puy.	Mt.V.	Puy.	Mt.V.
	<u>2018</u>							
Biochar	2.8	2.6bc	0.28	0.25b	0.39b	0.53c	0.03c	0.05b
Compost	3.0	3.1a	0.30	0.31a	0.69ab	1.02a	0.04ab	0.13a
Co-compost	3.0	3.0ab	0.30	0.29ab	0.62a	0.96a	0.04ab	0.13a
Compost + biochar	3.3	2.8abc	0.31	0.28ab	0.73a	0.76b	0.05a	0.09a
No amendment	2.9	2.4c	0.29	0.27ab	0.49ab	0.38c	0.03bc	0.05b
	<u>2019</u>							
Biochar	5.8a	3.19e	0.45a	0.24e	2.09c	0.95e	0.05e	0.05de
Compost	6.0a	3.75d	0.60b	0.37d	2.06c	1.49d	0.12d	0.21c
Co-compost	6.0a	3.75d	0.58b	0.38c	2.10c	1.22de	0.12d	0.18c
Compost + biochar	5.7a	3.23e	0.49a	0.30d	1.85c	0.93e	0.06de	0.10d
No amendment	4.4b	2.37f	0.45a	0.27e	0.89d	0.38f	0.04e	0.04e

### 4.3.3 Mid-season inorganic nitrogen

In 2018 at 44 days following fertilization, fertilized Puyallup plots had an 80% increase in nitrate nitrogen (NO<sub>3</sub>-N) compared to unfertilized split plots, while no difference was detected in ammonium nitrogen (NH<sub>4</sub>-N) (Table 17). In comparisons between organically amended soils and in comparisons with the unamended control, no statistical differences were observed in either NO<sub>3</sub>-N or NH<sub>4</sub>-N levels for 2018. In 2019, however, statistical differences between amendments were the only differences detected, and this was observed in both NO<sub>3</sub>-N and NH<sub>4</sub>-N levels which, followed a general pattern: the highest values were observed in the compost treatments (compost and co-compost) and the lowest in compost+biochar, biochar and unamended plots (Table 17). Strawberry beds in the fertilized split-plots received weekly applications of calcium nitrate (15.5-0-0) and this likely accounts for the increased NO<sub>3</sub>-N for 2018. Nitrification, the biologically mediated conversion of NH<sub>4</sub>-N to NO<sub>3</sub>-N, is believed to proceed rapidly in agricultural soils and is moderated by substrate availability, oxygen, and water potential (Norton and Stark, 2011). It is unlikely that any of these environmental conditions were deficient in our irrigated research plots, and therefore, any accumulated NH<sub>4</sub>-N would have nitrified rapidly, resulting in minimal NH<sub>4</sub>-N values and differences.

The differences we observed in comparisons between amendment in 2019 are likely the result of two previously mentioned mechanisms: (1) the change in materials after one year of storage (this likely increased the amount of inorganic N), and (2) the different N content of the materials. The highest levels of soil inorganic N (NH<sub>4</sub>-N and NO<sub>3</sub>-N) were observed in the compost and co-

compost amended plots and those two materials were amended at the highest rates of application and thus, provide the highest quantity of inorganic N. Our soil observations then, reflect these differences and are in line with expectations.

**Table 17: Mean inorganic nitrogen concentrations (mg kg<sup>-1</sup>) at 30 cm in fertilized (Fert) and unfertilized (Unfert) plots following amendment in Puyallup and Mt. Vernon soils for 2018 and 2019.**

	2018							
	Mt. Vernon				Puyallup			
	NH <sub>4</sub> -N		NO <sub>3</sub> -N		NH <sub>4</sub> -N		NO <sub>3</sub> -N	
Treatment*	Fert	Unfert	Fert	Unfert	Fert	Unfert	Fert	Unfert
Bio	11.0†	-	76.0ab <sup>Á</sup>	9.0b	-	-	16.9†	-
Comp	11.8	-	79.9b	12.5ab	-	-	19.0	-
Co-C	7.1	-	65.5ab	18.3a	-	-	19.7	-
C+B	10.9	-	83.2a	14.5ab	-	-	17.8	-
NoA	7.4	-	51.4b	9.8b	-	-	16.6	-
	2019							
Bio	22.5	3.9b	39.8c <sup>3</sup>	-	3.2b <sup>3</sup>	-	10.7b <sup>3</sup>	-
Comp	23.0	8.1a	81.1a	-	4.9a	-	20.4a	-
Co-C	34.9	7.2ab	91.0a	-	5.1a	-	21.3a	-
C+B	21.6	4.7ab	58.2b	-	4.1ab	-	10.8b	-
NoA	33.5	3.8b	48.1bc	-	4.1ab	-	9.4b	-

\*Treatment labels are as follows: Bio is Biochar; Comp is Compost; Co-C is Co-Compost; C+B is Compost + Biochar; NoA is No Amendment.

†Values illustrate the main effect of fertilizer.

<sup>3</sup>Values illustrate the main effect of amendment.

-Dashes indicate no significant difference.

<sup>Á</sup>Values within a year, site, variable (e.g., NH<sub>4</sub>-N), and fertilizer treatment followed by different letters are significantly different according to Tukey's HSD test, p<0.05.



In 2018 Mount Vernon soils, research plots were clearly influenced by the main fertilizer treatment, as different trends between fertilized and unfertilized split-plots were observed in mean concentrations of  $\text{NO}_3\text{-N}$  following organic amendment (Table 17). In fertilized plots for 2018, the lowest value in  $\text{NO}_3\text{-N}$  concentration was observed in unamended plots with increasing concentrations as follows: co-compost, biochar, compost alone, and compost plus biochar. However, the only statistically significant difference in  $\text{NO}_3\text{-N}$  concentration was observed between unamended plots and those receiving the compost plus biochar treatment. In unfertilized plots, biochar amended plots were lowest in  $\text{NO}_3\text{-N}$  concentrations and increased in the following order: unamended control, compost alone, compost plus biochar, and co-compost. Plots that received either type of biochar-compost combination (i.e., compost plus biochar or co-compost) were significantly higher in  $\text{NO}_3\text{-N}$  concentration than those plots receiving biochar alone.

Conversely, mean  $\text{NH}_4\text{-N}$  concentrations did not reveal an interaction between fertilized and amended plots, but did indicate significant differences among treatments (Table 17). Within organic amendment, the greatest concentrations of  $\text{NH}_4\text{-N}$  were observed in compost amended plots followed by compost plus biochar, biochar, co-compost, and unamended research plots (Table 17). In 2019 Mt. Vernon mean  $\text{NO}_3\text{-N}$  concentrations were only affected by amendment, which reflected a common trend measured in these field experiments: the highest values were observed in compost treatments (compost and co-compost), moderate values in compost+biochar treatment, and lowest values in biochar and unamended plots (Table 17).  $\text{NH}_4\text{-N}$  concentrations, on the other hand, were significantly affected by amendment but only in unfertilized plots. Not surprisingly, the  $\text{NH}_4\text{-N}$  concentrations in the unfertilized plots followed a now familiar trend: compost amended plots measured (compost > co-compost > compost+biochar) greater than the biochar and unamended plots (Table 17).

In the split-plot design, fertilized 2018 Mount Vernon soils received a total of  $108.6 \text{ kg N ha}^{-1}$  which would explain the elevated levels of  $\text{NH}_4\text{-N}$  in fertilized split-plots, as the N in this application was largely in the form of ammonium (Table 9). When mean values for  $\text{NH}_4\text{-N}$  concentrations in 2018 were separated and analyzed using a post-hoc approach, no significant differences between treatments were detected (Table 17). For 2019, elevated levels of  $\text{NH}_4\text{-N}$  were clearly the result of a similarly high ammonium fertilizer application, while the values observed in the unfertilized plots reflect the quantity of material applied, and thus the N applied.

For  $\text{NO}_3\text{-N}$  concentrations in 2018, two points should be considered: 1) 89% of pre-plant fertilizer applications are not recovered in end of season potato biomass (Rens et al., 2016), and 2) nitrification in potato hills proceeds at a relatively rapid rate (Zebarth and Milburn, 2003). Thus, our fertilized split-plots were likely flooded with excess  $\text{NO}_3\text{-N}$ , an environmental condition that may explain the varied trends in  $\text{NO}_3\text{-N}$  concentration following amendment in fertilized and unfertilized split-plots. In addition, when biochar is amended to soils, what can result is “nitrate-capture” (Hagemann et al., 2017). This function, if it were operating here, along with the additional N provided by compost, would explain why the greatest levels in  $\text{NO}_3\text{-N}$  were observed in the compost plus biochar treatment. The elevated  $\text{NO}_3\text{-N}$  levels following co-compost amendment in unfertilized soils could then be attributed to a function like the one just

mentioned or, it may be that the co-compost is acting as source of slowly available nitrogen (Kamman et al., 2015).

In 2019, similar to other trends reported here, mean values in soil NO<sub>3</sub>-N reflected the quantity of material applied, and thus the N provided by the amendment. This, of course, is unsurprising.

In addition, the low soil NO<sub>3</sub>-N concentrations we measured in biochar amended plots is an important observation for a producer to consider. Biochar, uncomposted and applied alone, is a rather chemically inert soil amendment which does not provide soil nutrient benefits in short term soil fertility responses.

#### **4.3.4 Post-season soil chemical properties and soil macro- and micronutrients**

In unfertilized soils and after one growing season, CEC values in Puyallup research plots were significantly affected by amendment but only in 2018. In Mount Vernon soils, no significant difference in CEC was detected in any of the four amended soils and in comparisons with the unamended control in either year, 2018 or 2019 (Table 18). In Puyallup soils, the greatest CEC values were observed in biochar, compost alone, and co-compost amended plots which all increased CEC in comparisons with the native soil (i.e., unamended). CEC values in the compost plus biochar amendment, however, did not differ significantly with those of the unamended control. CEC is a measure of soil adsorption properties and arises from the reactive surface properties of clay minerals and organic matter (Parfitt et al., 1995; Oorts et al., 2003). Biochar, with its high surface charge density (Liang et al., 2006), has been shown to increase soil CEC values in agricultural soils (Laird et al., 2010; El-Naggar et al., 2018) and our observations are consistent with those reports.

Additionally, and similar to the effects we observed, compost products like our co-compost and compost alone treatments are also known to increase soil CEC (Duong et al., 2012; Bhatta and Weatherley, 2018). This effect is generally attributed to carboxyl groups on the organic matter surface (Kleber et al., 2015), and since there was very little difference in chemical properties and rates of application between the co-compost and compost alone treatments, we would not expect to see detectable differences between the two. Interestingly, the compost plus biochar amendment did not increase CEC values as compared to the control. In our study, the rates at which biochar and compost were applied in the compost plus biochar treatment were exactly half that of biochar alone and other compost treatments (Table 8); this suggests that in these Puyallup soils, a minimal biochar and compost application rate must be met before CEC values will increase.

**Table 18: Mean CEC values (meq 100g<sup>-1</sup>) at 15 cm in unfertilized split plots following amendment in Mt. Vernon and Puyallup soils for 2018 and 2019. Within a location (e.g., Mt. Vernon) and year, values followed by different letters are significantly different according to Tukey's HSD test, p<0.05.**

Treatment	Mt. Vernon		Puyallup	
	2018	2019	2018	2019
Biochar	13.32	11.98	14.07a	12.03
Compost	13.20	13.00	13.80a	13.65
Co-Compost	13.44	13.05	13.76ab	13.35
Compost+Biochar	13.01	13.00	12.88bc	12.35
No amendment	12.86	11.75	12.75c	12.70

Following one growing season in Mount Vernon, the greatest mean concentration of NO<sub>3</sub>-N in 2018 was detected in plots that received the compost plus biochar treatment, which was significantly greater than the unamended control and biochar alone treatments (Table 19). Mean NO<sub>3</sub>-N concentrations in compost alone and co-compost amended plots were indistinguishable from the highest and two lowest values. This is unlike observations in 2019, where NO<sub>3</sub>-N concentrations followed an expected pattern: high levels of NO<sub>3</sub>-N in the two compost treatments (compost and co-compost) and lower levels in all other treatments (biochar+compost, biochar, and unamended control) (Table 19).

In 2018, the compost plus biochar amended plots contained roughly half the total N (wt. %) supplied by the compost or co-compost treatments (Table 7 and Table 8) and half the total amended biochar (by weight), and thus, it was unexpected for the largest mean NO<sub>3</sub>-N concentrations to be observed in these plots. However, research into biochar amended soils has indicated that significant chemical and physical changes occur as biochar ages (Cheng and Lehmann, 2009; Fang et al., 2015), and these changes may significantly alter soil nutrient cycling (Liu et al., 2013). Considering the compost plus biochar treatment included additional C, and labile C is known to have considerable effects on N transformations, particularly in combination with biochar (Fiorentino et al., 2019), the elevated NO<sub>3</sub>-N concentrations we observed may reflect a synergistic effect between aging biochars and the additional C input. This

helps, but does not fully explain why the lowest NO<sub>3</sub>-N concentrations were observed in the biochar alone amended plots, where the biochar rate is twice that of the compost plus biochar treatment, and why these unexpected values were only observed in 2018. Biochars produced at high temperatures (871°C), like the one used in this study, generally bear a negative charge (Cheng et al., 2008) and as a result, were previously thought unlikely to sorb negatively charged anions, like NO<sub>3</sub>-N (Iqbal et al., 2015), which would explain the low NO<sub>3</sub>-N values observed in our 2019 biochar alone plots. But, as others have shown, alternate chemisorption pathways are possible (Amonette and Joseph, 2009; Mukherjee et al., 2011) and are often attributed as the cause of the previously described “nitrate capture”. Moreover, as Haider et al. (2016) noted, our current extraction methods may not fully quantify the amount of inorganic N that is retained in biochar amended soils and composts, and short term N dynamics in biochar amended systems should be carefully interpreted.

In contrast, in Puyallup, mean NO<sub>3</sub>-N concentrations for 2018 were greatest in the co-compost treatment followed by compost alone, the unamended control, compost plus biochar, and biochar alone (Table 19). Comparisons between the greatest and two lowest mean NO<sub>3</sub>-N values, the co-compost amended plots and the biochar alone and compost plus biochar treatments, respectively, revealed the only significant differences in Puyallup soil. In 2019, only comparisons between the compost treatment and those with the biochar and unamended control plots were significantly different and these followed previously identified trends (i.e., more material and N applied results in higher NO<sub>3</sub>-N). It is not clear why such stark differences in NO<sub>3</sub>-N concentrations following amendment were observed between the sites, Mount Vernon and Puyallup, but the differences in cropping system may partially explain the variation in observed responses. It is well understood that root exudates are a large contributor to soil organic C (Haichar et al., 2008), and that differences in quantity and quality of exudate exist between plant species (Badri and Vivanco, 2009). Additionally, soil microbial communities, largely responsible for soil inorganic N availability (Mooshamer et al., 2014), are themselves shaped by variation in root exudate and thus shape soil nutrient transformations and availability (Guyonnet et al., 2017). Although we did not measure root exudates or soil microbial properties, differences in cropping system (i.e., potato versus strawberry) are likely contributing, in part, to observed differences in soil chemical properties following amendment and between the two sites.

**Table 19: Mean values for select soil macro- and micronutrients in unfertilized plots following amendment and one growing season in Mt. Vernon and Puyallup soils from 2018 and 2019. Within a year and location (e.g., Mt. Vernon), values in a variable row (e.g., K) followed by different letters are significantly different according to Tukey's HSD test,  $p < 0.05$ .**

	2018									
	Mt. Vernon					Puyallup				
V*	Bio**	Comp	Co-C	C+B	NoA	Bio	Com P	Co-C	C+B	NoA
NO <sub>3</sub> -N (mg kg <sup>-1</sup> )	23.3ab	26.8ab	34ab	41.1a	19.4 b	8.8a	19.4b c	26.2c	10.9bc	12.4b c
K (mg kg <sup>-1</sup> )	341bc	381.8ab	411.8a	370.5a bc	307.5c	254.5 d	319.2 5ef	341.5g	266.5d e	263.2 5d
Ca (meq 100g <sup>-1</sup> )	8ab	8.6a	8.8a	8.35ab	7.32b	-	-	-	-	-
Mg (meq 100g <sup>-1</sup> )	1.1ab	1.25a	1.28a	1.18ab	1.0b	0.88d	1.1c	1.03cd	0.9cd	0.9cd
Na (meq 100g <sup>-1</sup> )	0.18b	0.33a	0.37a	0.27ab	0.14b	0.16	0.24	0.26	0.17	0.17
S (mg kg <sup>-1</sup> )	11.3b	13.0ab	14.5a	12.6ab	11.3b	-	-	-	-	-
Zn (mg kg <sup>-1</sup> )	1.92c	2.7ab	3.2a	2.5abc	2.0bc	0.73e	1.6d	1.7d	1e	0.98e
	2019									
NO <sub>3</sub> -N (mg kg <sup>-1</sup> )	43.0b	105.4a	134.4a	67.1b	60.0b	16.8d	34.9c	21.7cd	24.9cd	13.0d
NH <sub>4</sub> -N (mg kg <sup>-1</sup> )	8.13b	17.4a	17.7a	11.3b	11.0b	-	-	-	-	-
P (mg kg <sup>-1</sup> )	80	89	93.3	93	77	-	-	-	-	-
OM %	2.6b	3.4a	3.4a	3.1a	2.5b	3.0de	3.7c	3.8c	3.2d	2.8e
Ca (meq 100g <sup>-1</sup> )	6.6b	8.4a	8.7a	7b	6.5b	-	-	-	-	-

Mg (meq 100g <sup>-1</sup> )	1.3b	1.9a	1.9a	1.4b	1.3b	0.8d	1.1c	1.1c	0.9d	0.8d
Na (meq 100g <sup>-1</sup> )	0.2c	0.6a	0.6a	0.3b	0.1c	0.1e	0.2d	0.2d	0.1e	0.1e
S (mg kg <sup>-1</sup> )	43.7ab	45.6a	47.4a	40.6ab	37.4b	-	-	-	-	-
Zn (mg kg <sup>-1</sup> )	1.7c	3.8a	4.0a	2.5b	1.7c	0.7f	1.9d	1.9d	1.1e	0.7f
<p>*V lists the chemical variable.</p> <p>**Treatment labels are as follows: Bio is Biochar; Comp is Compost; Co-C is Co-Compost; C+B is Compost + Biochar; NoA is No Amendment.</p> <p>- Dashes indicate no significant difference.</p>										

Soil mean macro- and micronutrients were significantly affected by amendment but observations were inconsistent across site and year (Table 19). In 2018, K, Mg, Na, and Zn were all significantly affected in Mt. Vernon and Puyallup, while significant effects to Ca and S were observed only in Mt. Vernon soils. In 2019, OM (%), Mg, Na, and Zn were significantly affected in Mt. Vernon and Puyallup soils while NH<sub>4</sub>-N, P, Ca and S were affected in Puyallup soils only (Table 19).

The variation in nutrient effects between sites is due, in part, to differences in cropping system (i.e., potato vs. strawberry) and soil type (i.e., different starting concentrations for each nutrient), while the variation in macro- and micronutrients between years most likely reflects legacy cropping systems as different treatment amendments and management strategies are evaluated at these research centers. Still, in general, mean values for macro- and micro-nutrients were significantly increased in compost and co-compost amended plots as compared to the biochar and unamended plots (Table 19), and this makes sense given the additional nutrients that are available in composted materials (Alvarenga et al., 2015; Zheljazkov and Warman, 2004; Zhang et al., 2006; Zheljazkov et al., 2006). The compost+biochar treatment had moderate effects on soil macro – and micronutrients, which is due to the moderate rate at which the compost portion was applied. The biochar, however, had very little effect on soil macro- and micro-nutrients and this is not surprising given the variation in soil response to biochar amendment. For example, Lopez-Cano et al. (2018) observed no effect on soil macro- and micronutrients following biochar amendment, while Laird et al. (2015), in a soil column study, reported significant differences in soil P, K, Ca, and Mn, but only at the highest rates of biochar incorporation. Additionally, the biochar used for this study was produced from softwood, a product typically low in elemental concentrations (Suliman et al., 2016). This suggests that: 1) this biochar may never provide additional nutrients or that, 2) our application rate was too low for differences in soil macro- and micronutrients to be detected.

## 5. Conclusions

### 5.1 Impact of biochar on gas emissions during composting

Volatile organic compound data collected during field sampling at both Lenz and WSU compost facilities displayed large variability in flux densities for a particular compost age and pile type. Because of this, it was not possible to conclude if biochar reduced VOCs emissions from composting processes through field sampling. At Lenz, positive or negative air flow through the aerated static piles likely resulted in measurements of high and low flux values depending on the direction of the air flow. For aerated static piles, the flux isolation chamber approach for measuring emission rates may not work well. Measured flux densities under positive flow may be overestimated because of the artificially high helium tracer dilution correction factor resulting from air flow from the pile itself. This aeration flow would dilute the helium tracer but be a strong source of VOCs. Another factor that may have caused variations in flux densities at Lenz and WSU compost facilities could be heterogeneity of surface emissions. At WSU, the three piles of similar type could display very different flux densities, implying that it is difficult to replicate the composting process. Large differences in pile temperature are evidence of this. The potential for compound loss in the air sampling apparatus under high humidity conditions was also observed. Water vapor from the hot piles often condensed in the flux chamber and sample lines and in the canisters themselves. Condensation of water would impact sampling efficiency of water soluble compounds such as light alcohols and aldehydes. Sample loss upon storage of S-containing and N-containing compounds in the canisters may have also contributed to flux variability for these compounds. For future field sampling, in-situ VOCs measurements using PTR-MS instrumentation would avoid some of the problems of sampling and storage losses of light alcohols, S-compounds, and N-compounds inherent in grab sampling methodologies.

For the WSU compost piles, monoterpenes and sulfur compounds, such as DMDS, were the most abundant VOCs. At Lenz, monoterpene emissions dominated the VOC flux density. For both the Lenz and WSU piles, monoterpenes emissions decreased with pile age, and as the pile aged, S-compound emissions for the WSU piles became a larger fraction of the emissions. For the WSU piles, we observed that N-compound emissions from the 10% biochar piles were often much lower than the control piles, hinting at the potential effect of biochar absorbing these types of compounds.

Lab-based composting experiments contrasting emissions from a control pile and a pile with 10% biochar were conducted twice. Material was supplied from the WSU composting facility. In the first trial the material contained no food waste and a lower fraction of manure than a typical WSU material mix. The second trial was a more typical WSU mix with food waste and green waste material mixed with manure. Volatile organic compounds were continuously measured by PTR-MS. The second trial had much higher emission fluxes of monoterpenes, alcohols, and ketones than first trial. Both first and second trials provided evidence that addition of 10% biochar can reduce emissions of monoterpenes, DMDS, other compounds not yet identified. The reduction of monoterpene and DMDS emissions would help reduce compost

facility odor. Since monoterpenes were the most abundant VOCs emitted at the Lenz and WSU compost facilities, its emission reduction also has the potential to be useful in reducing total VOC emissions for regulatory compliance. Emissions of alcohols, ketones and some S-containing compounds (H<sub>2</sub>S and DMS) were fairly similar between the control and biochar tanks. Analysis of greenhouse gas emissions for the first trial reveals that biochar reduced methane and nitrous oxide emissions.

The lab based experiments for quantifying the impact of biochar on compost emissions had better success than field sampling. While from the two lab trials there was consistency on the impact of biochar at reducing emissions of some VOCs, further trials are necessary to convincingly quantify its impact at reducing emissions from green waste materials, including reducing emissions of greenhouse gases.

As far as we are aware, using the PTR-MS in a laboratory composting test was the first time this type of VOC measurement technology was applied to compost emissions. The PTR-MS provides the ability to continuously measure the light alcohols and monoterpenes that appear to dominate VOC mass emission rates at WSU compost facilities. In addition, several compounds that contribute to odor including ammonia, H<sub>2</sub>S, DMS, and DMDS can also be measured by PTR-MS. The PTR-MS measurement method is ideally suited to measure the most important VOCs emitted in composting. Continuous measurements reveal that for some compounds, a large fraction of emissions occurred in the first few days of composting. For the first 3 days of the food / green waste / manure mixture, emissions of ethanol, methanol, and acetone were very high. Discrete sampling, for example starting on day 3 as was done for Lenz and WSU facility sampling, would appear to miss this large initial flux of light alcohols and ketones. It can be concluded that for future sampling either in the field or laboratory, continuous measurements starting at day 1 should be conducted to properly determine VOC emission rates and impact of biochar amendments.

The laboratory composting experiment using large volume tanks demonstrated that it may be possible to substitute lab based experiments for field sampling to derive VOC emission factors from compost (lbs VOC / ton wet material). The advantage of the lab based method is that the emission sampling can be done continuously to better determine how VOC emission rates change with time. In addition, there is much better control of the sampling conditions which improves the quality of the VOC flux density measurement. This includes better control of the dilution flow and humidity and the avoidance of compound losses from condensed water in sampling lines, and the apparent impact of wind on dilution of the flux isolation chamber. What has to be demonstrated is that lab based composting yields similar flux densities to composting piles at facility scale.

## **5.2 Impact of biochar soil amendment on crop productivity and soil physicochemical properties**

We tested biochar, compost, co-compost and a field-mixed biochar plus compost as soil amendments, and compared those responses to an unamended control in a variety of different



cropping systems and sites: basil (field, Colbert), basil (greenhouse), strawberry (greenhouse), strawberry (field, Puyallup), and potato (field, Mount Vernon). To provide additional insight into the effects of these amendments, we examined phytochemical composition of basil produced with these amendments in field and greenhouse conditions. We also compared a number of soil physiochemical properties in response to amendments in the field trials in Puyallup and Mount Vernon.

Amendment with compost, co-composted biochar, and compost plus biochar resulted in some productivity increase in sweet basil and strawberries. However, the effects were not uniform and varied by compost, biochar, and crop. The same amendments to the soil did not significantly affect the phytochemical composition of field- or greenhouse-grown sweet basil.

Crop yield and soil health attributes were generally affected, and in many cases, significantly so, by two amendments, the co-compost and compost plus biochar treatments. For the potato field trial, co-compost amendments were the only amendment whose application resulted in crop yield increases, but this was demonstrated only in fertilized split-plots. Co-compost and the compost plus biochar tended to affect soil physicochemical properties beneficially. Co-compost and compost influenced nutrient availability. These two treatments tended to increase K, Ca, and Mg in soils. Our results suggest that blending compost with biochar optimizes the physical and chemical properties of each, but that this effect is somewhat moderate and dependent upon the native soil and crop. Co-compost and biochar both significantly improved soil physical properties during one site-year. Continual application at the same rate or a larger one-time rate would likely lead to more significant and consistent changes.

The use of biochar as a soil amendment in cropping systems may be beneficial, but its use, and the intent of its use, need to be carefully considered and clearly defined. The moderate differences we observed between biochar products (i.e., co-composted biochar or biochar alone) warrant this consideration, as growers using one or the other will likely see drastic differences in performance, and so, expectations for yield and soil responses should be appropriate to the product. Biochar applied alone did not tend to increase soil nutrient status or crop yield. There may be some positive effects to soil properties when biochar is applied alone (e.g. increased cation exchange capacity as we measured in one site-year) but soil fertility was not positively impacted. It should also be noted that because the characteristics of the biochar and compost impact chemical and biological processes, the use of different types of biochar or compost in these studies would be expected to yield different results.

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## Appendix

**Table A.1: Flux densities ( $\mu\text{g m}^{-2} \text{min}^{-1}$ ) of the thirty most abundant compounds measured in the Day 3 samples collected from an aerated static pile at Lenz Enterprises.**

Name	Cntrl 1	Cntrl 1D	Cntrl 2	Biochar 1	Biochar 2	Equip Blank
d-limonene	29847	30813	15732	22126	6687	2.1
$\alpha$ -pinene	6635	5477	3742	9784	1770	0.2
$\alpha$ - thujone	5949	9870	3539	26270	2498	0.7
$\beta$ -pinene	2569	1880	1410	6085	921	0.1
3-Carene	2551	1881	1188	5437	713	0.1
camphor	2173	3006	2186	6187	1963	1.7
$\beta$ -thujone	2099	3079	1220	8442	823	0.3
$\alpha$ -phellandrene	2001	1484	928	4253	535	0.1
acetone	1866	1353	528	3339	232	0.1
3-methylbutanal	1772	1305	687	6495	9	0.0
$\gamma$ -terpinene	1363	1028	607	2629	408	0.1
terpinolene	1312	1007	631	3265	424	0.0
ethanol	744	412	53	1701	11	0.1
2-butanone	677	428	186	1206	130	0.0
$\beta$ -myrcene	636	446	274	1007	150	0.1
2-methylbutanal	622	468	255	2649	2	0.0
$\beta$ -phellandrene	497	363	200	1003	144	0.0
2-methylpropanal	475	321	178	1586	1	0.0
3-methyl-1-butanol	466	290	102	1929	9	0.0
camphene	324	225	135	534	68	0.0
2,3-butadione	223	175	87	159	3	0.0
furfural	113	66	20	1	0	0.0
hexanal	93	60	40	150	1	0.0
DMS	49	32	7	13	13	0.0
Styrene	37	21	11	60	6	0.0
Isoprene	35	24	10	14	11	0.0
DMDS	27	17	10	125	31	0.0
toluene	19	13	5	31	5	0.0
trans-2-Butene	4	3	2	7	1	0.0
1-Pentene	2	3	1	3	1	0.0