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# Valorization of Vineyard By-Products to Obtain Composted Digestate and Biochar Suitable for Nursery Grapevine (*Vitis vinifera* L.) Production

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**Abstract:** Although compost and biochar received high attention as growing media, little information is available on the potential of vineyard by-products for the production and use of composted solid digestate (CSD) and biochar (BC). In the present study, two experiments are reported on CSD and BC mixed with commercial peat (CP) for grapevine planting material production. Four doses (0, 10%, 20%, 40% vol.) of CSD and BC were assessed in the first and second experiment, respectively. CSD mixed at a dose of 10% recorded the highest values of shoot dry weight (SDW) and a fraction of total dry biomass allocated to shoot (FTS), both cropping bench-graft and bare-rooted vine. On the other hand, CSD mixed at a dose of 40% displayed the highest values of SDW and FTS, cropping two-year-old vine. BC used at a dose of 10% improved SDW, root dry weight, total dry weight, FTS, shoot diameter, and height on bare-rooted vine. The present study shows that CSD and BC, coming from the valorization of vineyard by-products, can be used in the production of innovative growing media suitable for nursery grapevine production. Further studies are needed to assess the combined applications of CSD and BC in the same growing media.

**Keywords:** vineyard by-products; composted solid digestate; biochar; grapevine planting material

## 1. Introduction

The growing demand for grapevine (*Vitis vinifera* L.) planting materials, due to the increasing worldwide viticulture, is promoting research studies to obtain useful guidelines for improving vineyard sustainability [1]. In addition, the losses caused by the failures, such as poor establishment or vigor of the vines and infection of trunk disease pathogens, are a significant but often unrecognized burden for nurseries, farmers, and wineries [2–4].

Among several agronomic practices, soil and vineyard canopy management received particular attention. In fact, soil and canopy management play a fundamental role on the vegetative and reproductive development of the vines [5,6]. Soil management practices include the fertilizer administration able to improve and/or maintain soil fertility, satisfying grapevine nutrient requests [7].

Field production and planting in the vineyard of dormant bare-rooted vines may be negatively affected by some biotic and abiotic stresses, such as root dehydration, contamination by soil borne

pathogens, and frost damage [8]. Thus, to overcome these problems, the production in greenhouses of forced bench-grafts or dormant bench-grafts grown in containers is a successful alternative to open-field propagation, allowing the transplanting of high-quality plants at any time of the year.

In greenhouse nursery, peat is the most common growing medium used by growers; however, peat is an expensive and non-renewable material [9]. Hence, the reduction of exploitation of peatlands received high attention by researchers, and currently different materials are investigated as substitute growing media [10].

Growing media originated from the valorization of agro-industrial by-products might be an alternative to the consumption of peat. Among them, compost and biochar can be an interesting ingredient for alternative and sustainable substrates [11,12]. On the other hand, one critical point when compost and biochar are used at high doses is ascribed to the alkalization of growing media, causing a reduced nutrient availability for plants [12].

Although several studies performed in a greenhouse nursery showed that compost and biochar, used at low doses, are able to improve plant growth [9,13], few studies were performed on grapevine planting material productions [14–16]. On the other hand, working in the open field, compost and biochar were used in large amounts, up to 30 and 44 t ha<sup>-1</sup>, respectively [14,17]. Moreover, to expand the range of agro-industrial by-products used as alternative substrates, the investigation of new feedstock materials should be performed. In this framework, little attention was paid to vineyard by-products.

Vineyard biomass is characterized by underutilized by-products (such as vine pruning residues and winery outstream, such as grape stalks) that can be valorized [18] by processing them in compost and other products, thus reducing the vineyard carbon footprint [19,20]. Vineyard winter pruning wood is usually destroyed by infield burning or crushing onto the soil, and the same occurs for grape stalks. On the other hand, a better valorization of vineyard pruning and winery out stream as feedstock for biogas plants [21–23] and pyrolysis [24] might increase vineyard sustainability providing green energy and promising fertilizers, such as digestate and biochar [25]. Considering that little information is reported on the use of compost and biochar coming from the valorization of vineyard by-products, the present study aimed to close the waste cycle in vineyards via the reuse of by-products.

In a vineyard, a high correlation between the development of the belowground and aboveground organs were demonstrated [26,27]. Thus, our objective was to assess the potential benefits of composted solid digestate (CSD) and biochar (BC) coming from vineyard by-products on different grapevine planting materials grown in the greenhouse. Two independent experiments were carried out. In the first experiment, CSD was assessed on greenhouse forced bench-grafts, bare-rooted vines, and two-year-old vines, while in the second experiment, BC was assessed on bare-rooted vines.

## 2. Materials and Methods

### 2.1. Composted Solid Digestate and Biochar Productions

Digestate containing grape stalks and coming from a biogas plant was composted with chips (1 cm in length) of vineyard winter pruning residues. Moreover, chips of vineyard prunings were also used in a gasifier to obtain biochar. Grape stalks and prunings derived from a vineyard of 'Lambrusco Salamino', grafted on 'Kober 5 BB' rootstock, located in the provinces of Modena and Reggio Emilia, Italy.

CSD was produced at the University of Modena and Reggio Emilia, located at Reggio Emilia, Italy, through a static pile on a farm composting for 105 days, turned weekly. Solid digestate (SD) was conferred by a local biogas plant (CAT, Correggio, Italy) and composted with vineyard prunings chips, mixed at a ratio of 15.0%–83.3% in dry weight, respectively. Feedstocks used in the anaerobic digestion were maize (*Zea mays* L.) silage (43%), triticale (*X Triticosecale* Wittmack) silage (22%), cow slurry (27%), and grape stalks (8%). The 1 m<sup>3</sup> pile was wetted through an irrigation system and manually activated on demand when the pile gravimetrically determined relative humidity (RH) was <50%. An aliquot of 1.7% dry weight of mature compost was added to the pile as a composting starter. Composting

temperatures were measured by thermoresistance sensors (PT100, Gandolfi, Parma, Italy), placed in the center of the pile at 15 cm from its base. A total of 35 days of the thermophilic phase were followed by a further 2-month curing period.

The biochar used in this study was produced in a PP20 gasifier, a commercial biomass-to-power unit manufactured by the US company, ALL Power Labs (Berkeley, CA, USA). The gasification reactor consists of a single throat, fixed bed, downdraft system. The reactor is coupled with a 4 cylinders internal combustion engine capable of producing 20 kW of electrical power at 60 Hz, or 16 kW at 50 Hz.

The biochar is extracted below the reduction zone of the reactor, where temperature decreased from the 900–950 °C of the combustion zone down to 650–700 °C of the end of the endothermic gasification zone. The temperature in the extraction point is much higher than the average dew point of polycyclic aromatic hydrocarbons, thus reducing the chance to find these compounds as toxic contaminants into the char. Biomass preparation consisted in the manual chipping of the vineyard prunings using a specifically designed rotary valve (Torex RWN05, Modena, Italy).

## 2.2. Compost and Biochar Characterisations

CSD and BC were analyzed according to the respective procedures indicated below. The content of different elements (Ca, Mg, Na, Cr, Cu, Cd, Ni, Fe, Pb, and Zn) was determined after acid digestion with a microwave oven, according to EPA 3052 (EPA 3052, 1996), with an ICP-OES (iCAP 6000 Series, Thermo Scientific, Waltham, MA, USA) on the basis of the EPA 6010D 2014 standard. The following parameters were detected according to the respective procedures: Total organic carbon (C) (UNI EN 13137:2002); total nitrogen (N) (UNI EN 13654-1:2001 and ISO 11261:1995); P<sub>2</sub>O<sub>5</sub> and K<sub>2</sub>O contents (UNI EN 13650:2002 and UNI EN ISO 11885:2009). The water content of the CSD and BC was determined after drying at 105 °C for 72 h. pH and electrical conductivity (EC) were determined on wet material (1:5 ratio) using a CRISON pH meter basic 20 (Crison Instrument, Barcelona, Spain) and CRISON GLP 31 EC meter (Crison Instrument, Barcelona, Spain), respectively. The growing media water capacity—the amount of water content held in the growing media after excess water has drained away and the downward movement has stopped—was determined by the gravimetric method, with two replicates for each control and treatment.

## 2.3. Phytotoxicity Characterisations of CSD and BC

CSD was evaluated for phytotoxicity, as suggested by Zucconi et al. [28]. A total of 20 seeds of garden cress (*Lepidium satibum* L.) were incubated at 20 °C in three-replicated Petri dishes, in which 4 mL of CSD water extract (50 g L<sup>-1</sup>) was poured over sterile filter paper. At 36 h after germination, the germination index percentage (GI%) was calculated on both roots and shoots. For roots, the following formula was used:

$$GI\% = 100 \times (G1/G2) \times (R1/R2) \quad (1)$$

where G1 and G2 are germinated seeds in the sample and control and R1 and R2 are the mean root length for the sample and for the control, respectively. For shoots, a similar formula adopted for roots, was used:

$$GI\% = 100 \times (G1/G2) \times (S1/S2) \quad (2)$$

where G1 and G2 are germinated seeds in the sample and control and S1 and S2 are the mean shoot length of the sample and control, respectively.

## 2.4. Microbiological and Suppressiveness Characterisations of CSD

The abundance of culturable filamentous fungi, total bacteria, and spore-forming bacteria in CSD were evaluated by a serial 10-fold (10<sup>-1</sup> to 10<sup>-7</sup>) dilution method in triplicate. Fungi were counted on the potato dextrose agar (PDA, Oxoid) pH 6, supplemented with 150 mg L<sup>-1</sup> of nalidixic acid and 150 mg L<sup>-1</sup> of streptomycin. Total bacteria were counted on the selective medium (glucose 1 g L<sup>-1</sup>, protease peptone 3 g L<sup>-1</sup>, yeast extract 1 g L<sup>-1</sup>, potassium phosphate buffer 1 g L<sup>-1</sup>, agar 15 g L<sup>-1</sup>)

supplemented with 100 mg L<sup>-1</sup> of actidione. Spore-forming bacteria were counted by plating 10-fold dilutions of CSD on nutrient agar previously heated at 90 °C for 10 min. Population densities expressed as a colony forming unit (CFU) g<sup>-1</sup> dry weight of CSD. Coliform, *Escherichia coli*, and *Salmonella* spp. detection were performed following the methods reported by Cekmecelioglu et al. [29].

The suppressiveness ability of CSD on two important soil-borne pathogens, such as *Rhizoctonia solani* and *Sclerotinia minor*, was assessed testing garden cress as a host plant. Potting mixes were done by amending commercial peat (CP) with CSD at a rate of 20% by vol. [30]. The bioassays were performed on 10 pots per treatment, and the pots filled up only with non-amended CP were used as the control. Pathogen inoculations and pot assessment were performed as reported by Pane et al. [31]. The bioassay was done in duplicate.

### 2.5. Growing Media Preparation and Characterisation

In the first experiment, four different growing media (GM) were composed by mixing ingredients as follows (% vol.): Commercial peat (CP) (Fondolinfa® Universale, Linfa Spa, RE, Italy) 100% (GM1); CP 90% + CSD 10% (GM2); CP 80% + CSD 20% (GM3); CP 60% + CSD 40% (GM4). In the second experiment, four different GM were tested mixing ingredients as follows (% vol.): CP 100%; CP 90% + biochar (BC) 10% (GM2); CP 80% + BC 20% (GM3); CP 60% + BC 40% (GM4). CP exhibited 70% of organic matter, 35% of organic carbon, and 0.6% N. Fertilisation was performed once a week throughout the growth cycle, administering, in total, 6 g of N per pot. Growing media pH and EC were determined on the wet material (1:5 ratio) using a CRISON pH meter basic 20 and a CRISON GLP 31 EC meter, respectively.

### 2.6. Nursery Greenhouse Experiments

Potting experiments were performed in a nursery greenhouse, located at Reggio Emilia, Italy, with programmed temperature ranging from 19 to 25 °C (day/night), relative humidity ranging from 50% to 70%, and with a natural photoperiod and solar radiation. At present, different grapevine planting materials are available in commerce. For the planting of a new vineyard, the most used material is the 1-year-old dormant bare-rooted vines (hereafter called 'bare-rooted vines'). This material is obtained by bench grafting dormant one-bud cuttings of the cultivar onto dormant hardwood cuttings of the rootstock, followed by callusing in a greenhouse and subsequently transferring in the open field for the development of roots and shoots. For marketability, the materials are excavated, the roots and shoot pruned, and plants sold with the bare root. For late planting in soil with lower water content, or to replace the dead vines in a new vineyard, nursery growers suggest the use of material similar to bare-rooted vines, but obtained using a scion bench grafted on rooted rootstocks and potted for callusing and hardening in a controlled greenhouse (hereafter called 'bench-grafts'). In fact, this material, having a preformed root-ball, could be more suitable for overcoming abiotic stresses than bare-rooted vines. Finally, to replace adult plants (having two or more years, but affected by diseases), the use of 2-year-old vines (obtained cropping bare-rooted vines in pots for two years, and hereafter called '2-year-old vines') is suggested by nursery growers in the vineyard. Hence, considering the availability of different planting material, in the first experiment, the following kinds of vines were used: (1) bench-grafts; (2) bare-rooted vines; (3) 2-year-old vines. Meanwhile, in the second experiment, only the bare-rooted vine was assessed. The combination scion/rootstock was cultivar 'Lambrusco Salamino' grafted on 'Kober 5BB' in all the three types of grapevine planting materials and in both experiments.

Grapevine planting materials were transplanted manually (one plant per pot) in plastic pots (4.5 L per bench-graft and bare-rooted vine and 9.0 L per 2-year-old vine) filled with the GM mixtures. Pots were arranged in a completely randomized design with five replicates. Pots were irrigated every night by spinner-type sprinklers in order to maintain the substrate at water capacity. Pests were controlled according to the integrated production rules of the Emilia Romagna Region, Italy. Grapevine planting materials were pruned, leaving one shoot that was trimmed at 120 cm at 259 day of year (DOY) and at 289 DOY in the first and second experiments, respectively. During the growing season,

the main phenological growth stages were recorded following the BBCH-scale (BBCH = Biologische Bundesanstalt, Bundessortenamt and Chemische Industrie, Germany) [32].

In the first and second experiments, at 177 DOY and 170 DOY, respectively, the following parameters were measured: the leaf chlorophyll content (CHL), leaf flavonoid content (FLAV), nitrogen balance index (NBI) (the ratio between CHL and FLAV, NBI), the leaf anthocyanin content (ANT), the basal shoot diameter, shoot height, number of leaves and nodes, and the shoot height/shoot diameter (H/D) ratio. CHL, FLAV, NBI, and ANT were estimated on the youngest fully expanded leaf by Dualex 4 Scientific (Dx4, FORCE-A, Orsay, France), an optical leaf-clip meter for non-destructive assessment of the physiological status of the plants [33]. At 273 DOY in the first experiment and 275 DOY in the second experiment, CHL, FLAV, NBI, ANT, shoot diameter, shoot height, and H/D ratio were recorded. At 314 DOY in the first experiment and 362 DOY in the second experiment, the shoot fresh weight (SFW), the aboveground part of the rootstock fresh weight (RSFW), the root fresh weight (RTFW), total fresh weight (TFW), shoot dry weight (SDW), aboveground part of rootstock dry weight (RSDW), root dry weight (RTDW), total dry weight (TDW), fraction of total dry weight allocated to shoot (FTS), fraction of total dry weight allocated to the aboveground part of rootstock (FTRS), and fraction of total dry weight allocated to root (FTRT), were recorded.

### 2.7. Data Analysis

Considering that the agronomic and physiological performance of the fruit-trees could be affected by the plant age and by the nutrients accumulated in the previous cropping seasons, the experimental data were analyzed separately for each grapevine planting material and experiment, using GenStat 17th software (VSN International, Hemel Hempstead, UK) for analysis of variance (ANOVA). Significant means were separated by Duncan's test at  $p < 0.05$ . Experimental data were also processed for a principal component analysis (PCA) in order to evaluate the existing relationships with original variables and growing media for each grapevine planting material and experiment.

## 3. Results and Discussion

Grapevines are relatively easy to propagate in the nursery greenhouse. However, high skill and organization are required to produce planting materials with the high standard quality requested every year by growers for new planting or replanting uneconomic vineyards or to replace plants affected by trunk disease pathogens [8].

Chemical and physical growing media characteristics can affect cutting establishment [34]. Growing media should be friable, free of weeds and pathogens, with good water capacity and drainage [35,36]. Among them, peat is the most commonly used, due to its positive hydrological, physiochemical, and agronomic characteristics [9,37]. However, to improve agricultural sustainability, researchers are called to study alternatives to peat utilization as innovative growing media. In the present study, vineyard winter prunings and grape stalks were valorized to obtain CSD and BC. CSD and BC were tested in two experiments as innovative ingredients for growing media preparation, used for the production in nursery greenhouses of grapevine planting material.

### 3.1. Compost, Biochar, and Growing Media Characteristics

During the composting process, CSD exceeded 60 °C for 16 days, achieving sanitation [38]. The main chemical characteristics of CSD and BC are reported in Table 1.

CSD and BC showed pH values of 7.9 and 10.0 with an EC of 1.5 and 2.5 dS m<sup>-1</sup>, respectively. Total N in CSD was more than six times compared to CP (3.2% vs. 0.6%), while BC showed a similar value to CP. The C/N ratio was 13.2 and 80.9 for CSD and BC, respectively, suggesting that the CSD was a suitable ingredient for growing media, since a C/N ratio less than 20.0 is required [39]. On the other hand, a direct use of BC is not indicated, suggesting dilution with other growing media. For organic C and total N, CSD and BC were within the framework previously reported by other studies [7,40]. CSD and BC showed levels of heavy-metal content below the limit established for the

commercialization of amendment in the European Community, according to the European Regulation CE 2003/2003. According to chemical parameters, CSD and BC appeared to be suitable for growing media preparation, although the high pH of CSD and BC and the high C/N ratio of BC encourages their dilution in a peat-based mixture, as previously suggested by other authors [7,41]. For this reason, in the present study, different growing media were composed by replacing CP from 10% to 40% vol. with CSD and BC.

**Table 1.** Main chemical characteristics of composted solid digestate (CSD) and biochar (BC).

Parameter	CSD Value	BC Value
pH	7.93	10.00
EC (dS m <sup>-1</sup> )	1.51	2.45
TOC (%)	41.79	56.60
Total N (%)	3.17	0.70
P <sub>2</sub> O <sub>5</sub> (%)	18.88	0.11
K <sub>2</sub> O (%)	27.47	19.09
Ca (%)	21.26	0.12
Mg (%)	9.81	0.31
Na (%)	0.88	0.22
C/N (–)	13.18	80.85
Cd (mg kg <sup>-1</sup> )	<0.50	<0.50
Ni (mg kg <sup>-1</sup> )	7.42	10.51
Cr (mg kg <sup>-1</sup> )	14.00	4.58
Zn (mg kg <sup>-1</sup> )	152.00	82.26
Cu (mg kg <sup>-1</sup> )	35.40	59.50
Hg (mg kg <sup>-1</sup> )	<0.20	<0.20
Pb (mg kg <sup>-1</sup> )	13.80	1.81
Fe (mg kg <sup>-1</sup> )	3.75	64.20
H <sub>2</sub> O (%)	77.30	71.37

Total organic carbon (TOC). All values as dry weight.

The germination assays indicated no phytotoxicity problems, both for the CSD and BC tested in the present study. In fact, the germination index displayed values higher than 50% for both CSD and BC, which is considered the threshold value for phytotoxicity [28]. In particular, the germination index showed higher values for the root and shoot of the sensitive reference species ‘garden cress’ when treated with a water extract of CSD (Table 2).

**Table 2.** Phytotoxicity assessment of CSD and BC and microbiological and suppressiveness characterization of CSD.

Index	Value
CSD-GI root (%)	138.00 *
CSD-GI shoot (%)	113.00 *
BC-GI root (%)	68.00 *
BC-GI shoot (%)	59.00 *
Fungi (CFU g <sup>-1</sup> )	9.63E + 06
Bacteria (CFU g <sup>-1</sup> )	3.61E + 02
Coliform bacteria (CFU g <sup>-1</sup> )	0.40E + 01
<i>Escherichia coli</i> (CFU g <sup>-1</sup> )	Absent
<i>Salmonella</i> spp. (CFU g <sup>-1</sup> )	Absent
<i>Clostridia</i> spp. (CFU g <sup>-1</sup> )	Absent
<i>Rhizoctonia solani</i> damping-off (%)	98.47
<i>Sclerotinia minor</i> damping-off (%)	46.56 *

Composted solid digestate (CSD), biochar (BC), germination index (GI). \* = statistically different compared to the control.

The microbial community within the compost was shown to be one of the major factors involved in the biological control of soilborne disease through different antagonistic mechanisms linked to the relationship between microbes [42]. Moreover, fungal populations were suggested as the main contributors of biological control in organic matrices [43]. Levels of microbial populations in CSD are shown in Table 2. Populations of total fungi, total bacteria, and *Coliform* bacteria were  $9.6E + 06$ ,  $3.6E + 02$ , and  $0.4E + 01$  (CFU  $g^{-1}$ ), respectively (Table 2). In addition, a total absence was recorded for *Escherichia coli*, *Clostridia* spp., and *Salmonella* spp., in line with the ecolabel criteria established by Decision 2001/688/CE.

The ability of compost to suppress soil-borne disease is also an important added value when compost is used as a component of growing media [44]. In the present study, suppressive bioassays displayed that garden cress damping-off caused by *S. minor* was significantly reduced by CSD, whereas no effects were reported on the control of *R. solani* (Table 2).

Growing media displayed pH and EC ranging between 7.6 and 8.5 and between 0.2 and 1.5  $dS m^{-1}$ , respectively (Table 3); values which are suitable for the production of several horticultural crops [12,45].

**Table 3.** Growing media assayed on grapevine planting materials.

Substrates	Formulation	pH		EC ( $dS m^{-1}$ )		GMWC (%)	
		CSD	BC	CSD	BC	CSD	BC
GM1	CP	7.6	7.6	0.21	0.21	85.0	85.0
GM2	CP 90% + CSD or BC 10%	7.6	7.8	0.22	0.46	83.2	81.3
GM3	CP 80% + CSD or BC 20%	7.7	8.1	0.39	0.87	84.6	79.1
GM4	CP 70% + CSD or BC 40%	7.8	8.5	0.44	1.48	84.8	77.0

Growing media (GM), commercial peat (CP), composted solid digestate (CSD), biochar (BC), electrical conductivity (EC), growing media water capacity (GMWC).

### 3.2. Grapevine Planting Material Productions

In the first experiment, morpho-physiological and agronomic parameters of bench-grafts, bare-rooted vines, and two-year-old vines were affected by CSD applications that, in general, induced a plant growth comparable to plant crops using only CP (Tables 4–6). In particular, bench-grafts and bare-rooted vines, when grown in amended pots with CSD at 10% (GM2), showed similar or higher values of FLAV, NBI, shoot diameter, shoot height, number of leaves and nodes, and H/D ratio, compared to CP at 177 DOY, corresponding to the BBCH-scale 19 “9 or more unfolded leaves”. In addition, the same growing media (GM2) recorded the highest values of RSFW, RTFW, TFW, SDW, and FTS at 314 DOY, corresponding to the BBCH-scale 97 “end of leaf-fall”, both on the bench-grafts and bare-rooted vines (Tables 4 and 5).

For two-year-old vines, plants grown in amended pots with CSD at 40% (GM4) highlighted similar or higher values of the CHL, FLAV, ANT, NBI, shoot diameter, shoot height, number of leaves and nodes, and H/D ratio, compared to CP at 177 DOY. Moreover, GM4 recorded the highest values of SDW, RTDW, TDW, FTS, and FTTR at 314 DOY (Table 6).

**Table 4.** Parameters assessed on greenhouse bench-grafts grown on growing media containing composted solid digestate.

<b>(A) Morphological and physiological parameters recorded at 177 day of year (DOY)</b>															
Treatment	CHL (-)		FLAV (-)		NBI (-)		ANT (-)		DIAMETER (mm)		H (cm)	LEAVES (no.)	NODE (no.)	H/D (-)	
GM1	<b>22.34</b>	a	1.25		18.15		0.15	b	4.10		140.00	30.20	26.20	<b>357.00</b>	a
GM2	17.74	b	1.06		17.65		<b>0.21</b>	a	3.74		135.00	30.20	24.80	<b>370.40</b>	a
GM3	17.12	b	1.06		17.19		0.20	ab	4.32		107.00	29.60	23.80	231.90	b
GM4	16.60	b	1.10		15.70		<b>0.24</b>	a	4.12		115.00	30.80	24.20	280.10	ab

<b>(B) Morphological and physiological parameters recorded at 273 DOY</b>													
Treatment	CHL (-)		FLAV (-)		NBI (-)		ANT (-)		DIAMETER (mm)		H/D (-)		
GM1	<b>12.98</b>	a	<b>3.38</b>	a	3.70	b	<b>0.85</b>	a	<b>5.15</b>	a	234.50	b	
GM2	10.98	b	3.06	b	3.59	b	0.68	b	4.22	b	<b>283.10</b>	a	
GM3	9.45	c	2.58	d	3.67	b	0.46	c	4.15	b	<b>292.90</b>	a	
GM4	11.26	b	2.82	c	<b>4.22</b>	a	0.34	d	4.61	ab	261.70	ab	

<b>(C) Morphological and physiological parameters recorded at 314 DOY</b>																						
Treatment	SFW (g plant <sup>-1</sup> )		RSFW (g plant <sup>-1</sup> )		RTFW (g plant <sup>-1</sup> )		TFW (g plant <sup>-1</sup> )		SDW (g plant <sup>-1</sup> )		RSDW (g plant <sup>-1</sup> )		RTDW (g plant <sup>-1</sup> )		TDW (g plant <sup>-1</sup> )		FTS (-)		FTRS (-)		FTRT (-)	
GM1	<b>20.88</b>	a	24.38	d	113.30	c	158.60	c	7.08	b	<b>23.80</b>	a	21.58	d	47.80	0.16	b	<b>0.52</b>	a	0.48	c	
GM2	17.62	b	<b>35.17</b>	a	117.90	b	<b>169.90</b>	a	<b>8.61</b>	a	19.32	d	23.94	c	45.00	<b>0.20</b>	a	0.45	b	0.55	b	
GM3	13.59	d	30.15	b	105.40	d	149.20	d	5.11	d	22.80	b	27.40	b	46.33	0.10	d	0.45	b	0.55	b	
GM4	15.64	c	26.77	c	<b>123.50</b>	a	165.90	b	6.50	c	20.26	c	<b>32.21</b>	a	55.00	0.12	c	0.39	c	<b>0.61</b>	a	

GM1 = commercial peat (CP) 100%; GM2 = CP 90% + composted solid digestate (CSD) 10%; GM3 = CP 80% + CSD 20%; GM4 = CP 60% + CSD 40%. Leaf chlorophyll content (CHL); leaf flavonoid content (FLAV); nitrogen balance index (NBI); leaf anthocyanins (ANT); shoot diameter (DIAMETER); plant height (H); number of leaves (LEAVES); number of nodes (NODE); height diameter ratio (H/D); shoot fresh weight (SFW); rootstock fresh weight (RSFW); root fresh weight (RTFW); total fresh weight (TFW); shoot dry weight (SDW); rootstock dry weight (RSDW); root dry weight (RTDW); total dry weight (TDW); fraction to shoot (FTS); fraction to rootstock (FTRS); fraction to root (FTRT); parameter without unit of measure (-). Means followed by the same letter do not significantly differ at  $p < 0.05$ .



**Table 5.** Parameters recorded on bare-rooted vines grown on growing media containing composted solid digestate.

<b>(A) Morphological and physiological parameters recorded at 177 day of year (DOY)</b>																					
Treatment	CHL (-)		FLAV (-)		NBI (-)		ANT (-)		DIAMETER (mm)		H (cm)		LEAVES (no.)		NODE (no.)		H/D (-)				
GM1	28.30	b	0.76		39.28		<b>0.10</b>	a	3.94		136.40	ab	27.20		24.20		350.30				
GM2	<b>35.92</b>	a	0.96		38.11		0.06	b	3.78		124.80	ab	30.80		25.80		353.10				
GM3	29.30	b	0.77		42.54		<b>0.10</b>	a	3.66		114.00	b	32.40		26.00		311.20				
GM4	30.50	b	0.73		42.34		<b>0.10</b>	a	4.24		<b>140.80</b>	a	32.00		25.00		340.00				

<b>(B) Morphological and physiological parameters recorded at 273 DOY</b>											
Treatment	CHL (-)		FLAV (-)		NBI (-)		ANT (-)		DIAMETER (mm)		H/D (-)
GM1	<b>32.09</b>	a	0.78	b	<b>41.80</b>	a	<b>0.06</b>	a	5.01	ab	240.90
GM2	<b>35.28</b>	a	<b>1.05</b>	a	33.69	b	0.03	b	5.12	ab	235.50
GM3	28.37	b	0.69	bc	<b>41.49</b>	a	0.05	ab	<b>5.82</b>	a	210.50
GM4	25.48	b	0.60	c	<b>42.72</b>	a	<b>0.06</b>	a	4.90	b	247.90

<b>(C) Agronomic parameters recorded at 314 DOY</b>																						
Treatment	SFW (g plant <sup>-1</sup> )		RSFW (g plant <sup>-1</sup> )		RTFW (g plant <sup>-1</sup> )		TFW (g plant <sup>-1</sup> )		SDW (g plant <sup>-1</sup> )		RSDW (g plant <sup>-1</sup> )		RTDW (g plant <sup>-1</sup> )		TDW (g plant <sup>-1</sup> )		FTS (-)		FTRS (-)		FTRT (-)	
GM1	24.05	ab	<b>18.99</b>	a	102.40	b	145.40	b	9.55	b	<b>12.34</b>	a	<b>32.60</b>	a	<b>53.91</b>	a	0.18	b	<b>0.23</b>	a	0.61	b
GM2	<b>24.61</b>	a	<b>19.42</b>	a	<b>109.77</b>	a	<b>154.20</b>	a	<b>11.53</b>	a	10.97	c	31.32	b	<b>53.70</b>	a	<b>0.21</b>	a	0.20	b	0.58	b
GM3	23.43	b	16.27	b	71.02	d	120.90	d	6.27	d	9.68	d	30.40	c	46.76	c	0.13	c	0.21	b	<b>0.65</b>	a
GM4	22.23	c	15.56	b	83.92	c	133.30	c	8.48	c	11.43	b	29.36	d	49.01	b	0.17	b	<b>0.23</b>	a	0.60	b

GM1 = commercial peat (CP) 100%; GM2 = CP 90% + composted solid digestate (CSD) 10%; GM3 = CP 80% + CSD 20%; GM4 = CP 60% + CSD 40%; Leaf chlorophyll content (CHL); leaf flavonoid content (FLAV); nitrogen balance index (NBI); leaf anthocyanins (ANT); shoot diameter (DIAMETER); plant height (H); number of leaves (LEAVES); number of nodes (NODE); height diameter ratio (H/D); shoot fresh weight (SFW); rootstock fresh weight (RSFW); root fresh weight (RTFW); total fresh weight (TFW); shoot dry weight (SDW); rootstock dry weight (RSDW); root dry weight (RTDW); total dry weight (TDW); fraction to shoot (FTS); fraction to rootstock (FTRS); fraction to root (FTRT); parameter without unit of measure (-). Means followed by the same letter do not significantly differ at  $p < 0.05$ .

**Table 6.** Parameters recorded on two-year-old vines grown on growing media containing composted solid digestate.

<b>(A) Morphological and physiological parameters recorded at 177 day of year (DOY)</b>																		
Treatment	CHL (-)		FLAV (-)		NBI (-)		ANT (-)		DIAMETER (mm)		H (cm)		LEAVES (no.)		NODE (no.)		H/D (-)	
GM1	19.34		2.50		7.95		0.18		5.40		83.40	c	21.80	ab	13.60	b	154.70	b
GM2	18.64		2.21		8.56		0.14		5.50		119.60	b	21.80	ab	<b>16.00</b>	<b>a</b>	220.50	ab
GM3	19.16		2.66		7.36		0.21		5.20		131.40	ab	<b>24.40</b>	<b>a</b>	<b>16.20</b>	<b>a</b>	<b>264.70</b>	<b>a</b>
GM4	21.98		2.31		9.52		0.16		5.50		<b>147.40</b>	<b>a</b>	17.00	b	13.40	b	<b>282.60</b>	<b>a</b>

<b>(B) Morphological and physiological parameters recorded at 273 DOY</b>														
Treatment	CHL (-)		FLAV (-)		NBI (-)		ANT (-)		DIAMETER (mm)		H/D (-)			
GM1	<b>15.12</b>	<b>a</b>	<b>3.45</b>	<b>a</b>	<b>4.47</b>	<b>a</b>	<b>0.72</b>	<b>a</b>	<b>6.23</b>	<b>a</b>	193.80	b		
GM2	12.08	c	3.23	b	3.74	b	0.56	b	5.63	b	<b>213.20</b>	<b>a</b>		
GM3	13.62	b	3.14	b	4.30	a	0.33	c	<b>6.31</b>	<b>a</b>	190.30	b		
GM4	9.38	d	2.75	c	3.43	c	0.16	d	<b>6.52</b>	<b>a</b>	184.50	b		

<b>(C) Agronomic parameters recorded at 314 DOY</b>																						
Treatment	SFW (g plant <sup>-1</sup> )		RSFW (g plant <sup>-1</sup> )		RTFW (g plant <sup>-1</sup> )		TFW (g plant <sup>-1</sup> )		SDW (g plant <sup>-1</sup> )		RSDW (g plant <sup>-1</sup> )		RTDW (g plant <sup>-1</sup> )		TDW (g plant <sup>-1</sup> )		FTS (-)		FTRS (-)		FTRT (-)	
GM1	145.40	b	42.29	d	<b>182.40</b>	<b>a</b>	243.40	b	8.44	c	27.40	c	47.58	d	83.42	d	0.10	ab	<b>0.32</b>	<b>a</b>	0.58	b
GM2	<b>154.20</b>	<b>a</b>	54.13	b	156.60	d	224.20	d	7.48	d	29.38	b	51.90	c	88.76	c	0.08	c	<b>0.33</b>	<b>a</b>	0.59	b
GM3	120.90	d	<b>62.71</b>	<b>a</b>	174.70	b	<b>258.60</b>	<b>a</b>	9.74	b	<b>33.46</b>	<b>a</b>	57.94	b	101.14	b	0.10	b	<b>0.34</b>	<b>a</b>	0.56	b
GM4	133.30	c	45.65	c	167.10	c	238.00	c	<b>11.36</b>	<b>a</b>	25.58	d	<b>71.04</b>	<b>a</b>	<b>107.98</b>	<b>a</b>	<b>0.11</b>	<b>a</b>	0.25	b	<b>0.64</b>	<b>a</b>

GM1 = commercial peat (CP) 100%; GM2 = CP 90% + composted solid digestate (CSD) 10%; GM3 = CP 80% + CSD 20%; GM4 = CP 60% + CSD 40%; Leaf chlorophyll content (CHL); leaf flavonoid content (FLAV); nitrogen balance index (NBI); leaf anthocyanins (ANT); shoot diameter (DIAMETER); plant height (H); number of leaves (LEAVES); number of nodes (NODE); height diameter ratio (H/D); shoot fresh weight (SFW); rootstock fresh weight (RSFW); root fresh weight (RTFW); total fresh weight (TFW); shoot dry weight (SDW); rootstock dry weight (RSDW); root dry weight (RTDW); total dry weight (TDW); fraction to shoot (FTS); fraction to rootstock (FTRS); fraction to root (FTRT); parameter without unit of measure (-). Means followed by the same letter do not significantly differ at  $p < 0.05$ .

The most likely explanation for the different behavior shown by bench-grafts and bare-rooted vines vs. two-year-old vines could be ascribed to the different plant age and is probably linked to the different root expansion (higher in the two-year-old vine) as already suggested by Bozzolo et al. [7]. In addition, as reported by Ericsson et al. [46] the growth dynamics of the fruit-tree depend on the changes in the source-sink balance. The same authors reported that a competition for intensive shoot growth can affect the root development, and a similar trend was displayed in the present study. In fact, a higher percentage of biomass was allocated to the shoot (and lower to the root) in bench-grafts and bare-rooted vines, grown on GM2. On the other hand, for the two-year-old vines, the highest values of biomass allocation, both to the shoot and root, were recorded using GM4. These results support one of the nursery grower requests, which is a planting material with a high dry matter content in the shoot.

Results about improved biomass production (using CSD) are in agreement with those of Raviv et al. [47] and Ronga et al. [12], who reported that composts obtained from separated cow manure with wheat straw, grape marc, orange peels, and spent coffee grounds positively affected potted plants grown in greenhouses. Similarly, vine root development in the vineyard was stimulated by the addition of compost from vine pruning waste supplied in the under-row of cultivar 'Cabernet Sauvignon' [48]. The improved biomass was ascribed to the presence of some substances working as growth promoters in the CSD as suggested by Bernal-Vicente et al. [49]. However, further research is needed to validate this hypothesis. In addition, the similar or highest dry weight displayed by growing media containing CSD is fundamental for successful transplanting as it is linked with an increase resistance to environmental stresses [9].

In the second experiment, BC was assessed on bare-rooted vines. BC applied at 10% (GM2) positively affected the ANT, shoot diameter, shoot height, number of leaves, and nodes at 170 DOY, corresponding to the BBCH-scale 19 (Table 7). Moreover, GM2 also increased the FLAV, ANT, and shoot diameter at 275 DOY, corresponding to the BBCH-scale 95 (50% of leaves fallen) and SFW, RTFW, TFW, SDW, RTDW, TDW, FTS, and FTRT at 362 DOY, corresponding to the BBCH-scale 97 (end of leaf-fall). Our results are in accordance with those of other authors, who reported increments in plants aboveground and belowground biomass, applying biochar at low doses to different species [15,50,51]. In grapevine, a significant increase of fine root biomass has been observed following the application of biochar as an amendment to the vineyard soil [52].

**Table 7.** Parameters recorded on bare-rooted vines grown on growing media containing biochar.

<b>(A) Morphological and physiological parameters recorded at 170 day of year (DOY)</b>																				
Treatment	CHL (-)		FLAV (-)		NBI (-)		ANT (-)		DIAMETER (mm)		H (cm)		LEAVES (no.)		NODE (no.)		H/D (-)			
GM1	18.80	b	3.84	a	4.93	b	0.27	b	4.90	c	85.00	c	19.20	b	14.80	b	113.90	a		
GM2	15.40	c	3.80	a	4.05	c	0.34	a	5.90	a	121.40	a	24.20	a	19.40	a	94.20	b		
GM3	17.18	b	3.50	a	4.89	b	0.30	b	5.41	b	107.80	b	18.20	b	12.20	b	74.70	c		
GM4	21.78	a	2.99	b	6.55	a	0.26	b	3.54	d	67.20	d	8.80	c	6.00	c	39.80	d		

<b>(B) Morphological and physiological parameters recorded at 275 DOY</b>														
Treatment	CHL (-)		FLAV (-)		NBI (-)		ANT (-)		DIAMETER (mm)		H (cm)		H/D (-)	
GM1	19.92	a	3.14	b	6.43	a	1.18	b	5.01	b	115.60	c	231.00	bc
GM2	12.76	b	3.66	a	3.49	b	2.09	a	5.89	a	126.70	b	215.30	c
GM3	13.78	b	3.25	b	4.24	b	1.87	a	5.48	ab	134.50	a	246.00	ab
GM4	12.94	b	3.06	b	4.29	b	1.26	b	3.86	c	102.20	d	261.60	a

<b>(C) Agronomic parameters recorded at 362 DOY</b>																						
Treatment	SFW (g plant <sup>-1</sup> )		RSFW (g plant <sup>-1</sup> )		RTFW (g plant <sup>-1</sup> )		TFW (g plant <sup>-1</sup> )		SDW (g plant <sup>-1</sup> )		RSDW (g plant <sup>-1</sup> )		RTDW (g plant <sup>-1</sup> )		TDW (g plant <sup>-1</sup> )		FTS (-)		FTRS (-)		FTRT (-)	
GM1	7.02	b	34.80	c	58.40	b	100.20	b	4.19	b	20.90	b	18.88	b	43.97	b	0.09	b	0.48	b	0.43	b
GM2	13.48	a	35.90	bc	81.80	a	131.20	a	6.84	a	19.83	b	22.55	a	49.23	a	0.14	a	0.40	c	0.49	a
GM3	14.80	a	48.00	a	49.80	b	112.60	b	7.36	a	28.44	a	15.85	c	51.66	a	0.14	a	0.55	a	0.36	c
GM4	4.30	c	40.40	b	59.60	b	104.30	b	2.62	c	22.82	b	17.75	b	43.19	b	0.06	c	0.53	a	0.41	b

GM1 = commercial peat (CP) 100%; GM2 = CP 90% + biochar (BC) 10%; GM3 = CP 80% + BC 20%; GM4 = CP 60% + BC 40%; Leaf chlorophyll content (CHL); leaf flavonoid content (FLAV); nitrogen balance index (NBI); leaf anthocyanins (ANT); shoot diameter (DIAMETER); plant height (H); number of leaves (LEAVES); number of nodes (NODE); height diameter ratio (H/D); shoot fresh weight (SFW); rootstock fresh weight (RSFW); root fresh weight (RTFW); total fresh weight (TFW); shoot dry weight (SDW); lklrootstock dry weight (RSDW); root dry weight (RTDW); total dry weight (TDW); fraction to shoot (FTS); fraction to rootstock (FTRS); fraction to root (FTRT); parameter without unit of measure (-). Means followed by the same letter do not significantly differ at  $p < 0$ .

Summarizing, the results of the two experiments are broadly in agreement with those of previous works, which reported that an increase of plant biomass is one of the macroscopic effects induced by the partial substitution of peat in the growing media with agro-industrial by-products, such as compost, digestate, and biochar [12,22,23]. The ability of agro-industrial by-products to increase plant biomass is probably due to an improved nutrient availability and uptake by the plant and/or to the presence of some microorganisms and compounds able to increase the plant growth [49]. In fact, when these substances are added in the growing media, they are able to improve the biochemical activity of the plants, similar to plant hormone-like promoters [53].

From a physiological point of view, the addition of CSD and BC reduced the CHL values, apart from bare-rooted vine grown on GM2 and GM4 containing CSD and BC, respectively, which recorded higher values compared to GM1. However, the lower values of CHL did not affect the biomass production and its distribution to shoot in each grapevine planting material. These results are in accordance with those reported by Bozzolo et al. [7].

Chlorophyll, flavonoid, and anthocyanin leaf contents are indices of leaf photosynthetic capacity and plant vigor status, which are linked to the N uptake [33]. In particular, high levels of anthocyanins in the leaf might allow the plant to increase resistance to abiotic and biotic stresses [54]. Thus, our results suggest that CSD and BC applied at 10% may increase plant resistance to stress on bare-rooted vines cropped in a pot in a greenhouse.

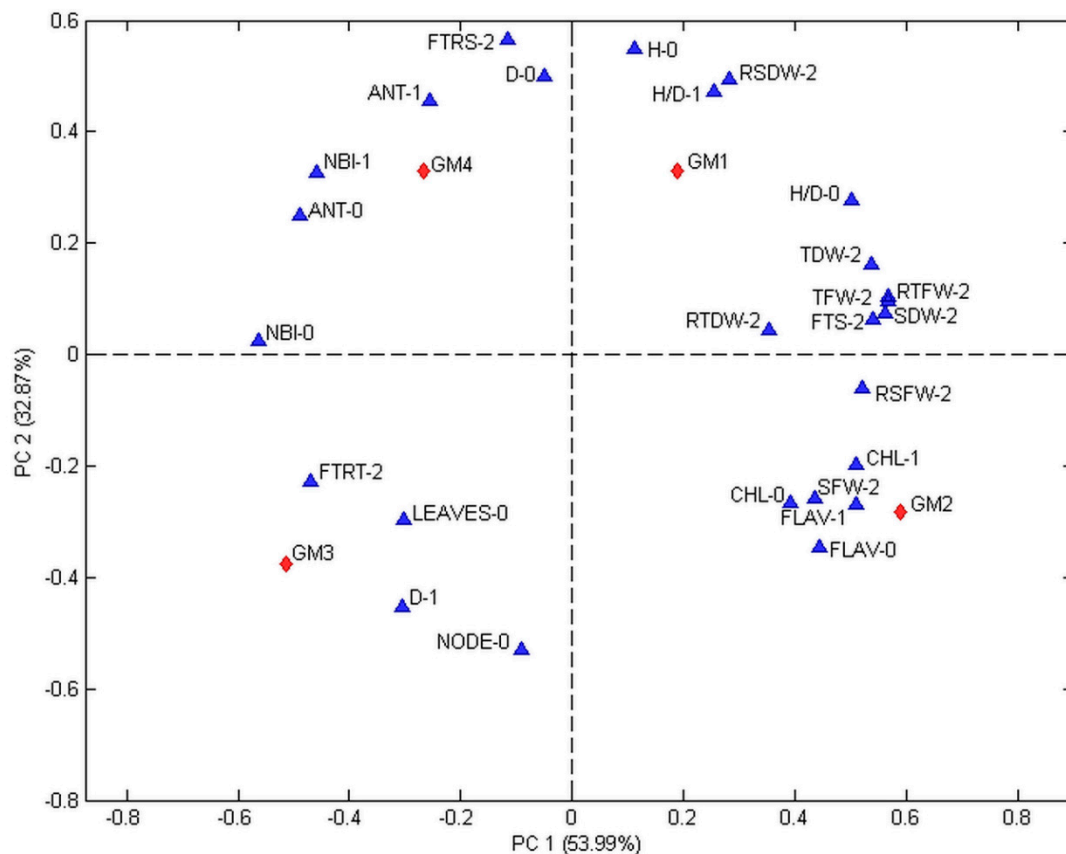
CSD and BC used at doses higher than 10% negatively affected the biomass production of bench-grafts and bare-rooted vines. These results were due to the high dosage of CSD and BC that induced alkaline pH to the relative growing media. Bozzolo et al. [7] also reported the same trend on grapevine and on other horticultural crops, such as tomato, basil, and lettuce [9,12].

One interesting aspect of our results is that the FTS was stimulated by CSD and BC applications used up to 10%, compared to CP. Moreover, BC applied at 10% (GM2) also showed the highest FTRT. On the other hand, working with the two-year-old vines, the highest values of FTS and FTRT were recorded applying CSD at a dose of 40% (GM4). The highest allocation of dry matter to the aboveground part of the vine might suggest the highest N uptake [55], confirming the role of CSD and BC in improving the nutrient plant uptake [7,50]. In addition, these results suggested that shoot growth response to CSD and BC was not only due to a higher nutrient uptake by plants, but that also other factors might be involved, such as the presence of humic substances and an improved porosity in the growing media, respectively [56,57].

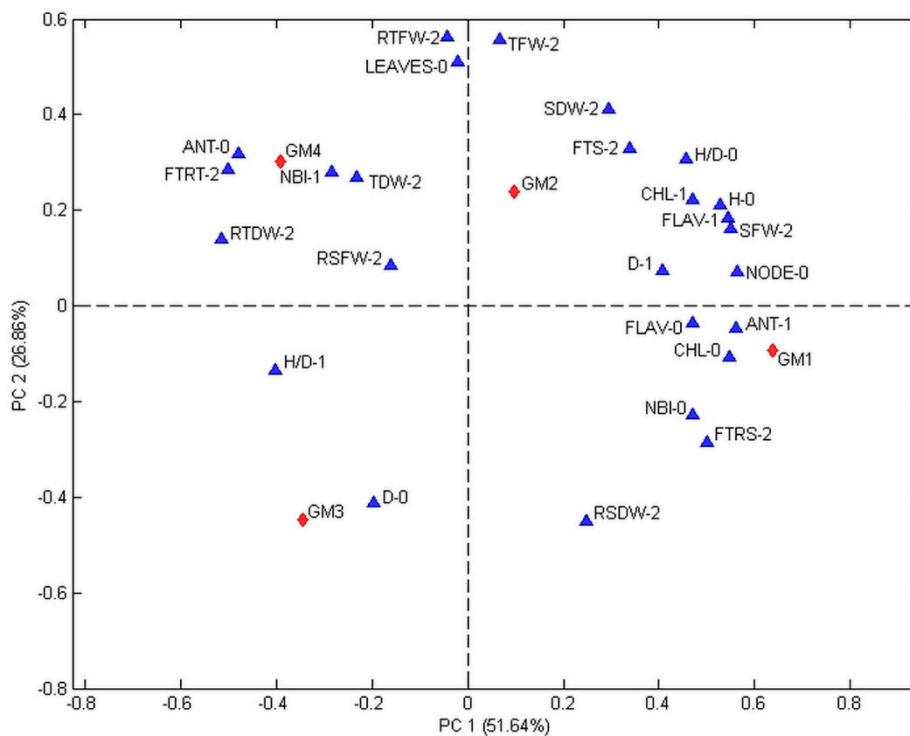
### 3.3. Relationships between Recorded Parameters and Growing Media

The correlations between data of growing media variables measured on grapevine planting materials were studied by PCA analysis. Figures 1–3 report ordination biplots of the PCA output of the first experiment on bench-grafts, bare-rooted vines, and two-year-old vines, respectively, assessing CSD. Figure 4 reports ordination biplot of the PCA output of the second experiment on bare-rooted vine, assessing BC. In the first experiment, for the bench-grafts, the PC1 accounted for 53.99% of the variance, while PC2 accounted for 32.87%. For the bare-rooted vines dataset, the two principal components, 1 and 2, accounted for 51.64% and 26.86% of the variation, respectively. Meanwhile, for two-year-old vines, the PC1 accounted for 49.14% of the variance, while PC2 accounted for 31.52%.

Growing media containing CSD at 10% (GM2) clustered along the positive side of PC1 both on the bench-grafts (Figure 1) and bare-rooted vines (Figure 2). GM2 performed as well as CP (GM1) and was associated with the most important parameters, such as CHL, FLAV, SDW, and FTS (Figures 1 and 2).

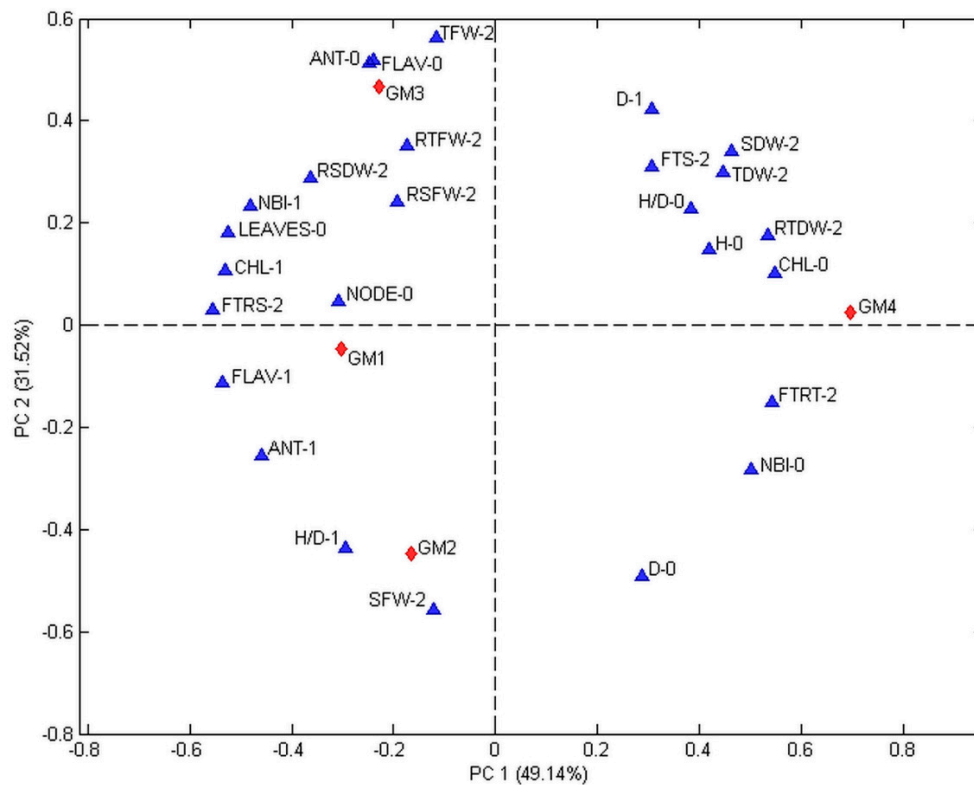


**Figure 1.** Ordination biplots of principal component analysis outputs of the data from bench-grafts grown on growing media (GM), containing composted solid digestate (CSD) at different rates. GM1 = commercial peat (CP) 100%; GM2 = CP 90% + CSD 10%; GM3 = CP 80% + CSD 20%; GM4 = CP 60% + CSD 40%. Labels in the triangles indicate the investigated parameters: Leaf chlorophyll content (CHL); leaf flavonoid content (FLAV); nitrogen balance index (NBI); leaf anthocyanins (ANT); plant height (H); height diameter ratio (H/D); shoot fresh weight (SFW); rootstock fresh weight (RSFW); root fresh weight (RTFW); total fresh weight (TFW); shoot dry weight (SDW); rootstock dry weight (RSDW); root dry weight (RTDW); total dry weight (TDW); fraction to shoot (FTS); fraction to rootstock (FTRS); fraction to root (FTRT). 0 = parameter recorded at 170 day of year (DOY); 1 = parameter recorded at 275 DOY; 2 = parameter recorded at 362 DOY.



**Figure 2.** Ordination biplots of principal component analysis outputs of the data from bare-rooted vines grown on growing media (GM) containing composted solid digestate (CSD) at different rates. GM1 = commercial peat (CP) 100%; GM2 = CP 90% + CSD 10%; GM3 = CP 80% + CSD 20%; GM4 = CP 60% + CSD 40%. Labels in the triangles indicate the investigated parameters: Leaf chlorophyll content (CHL); leaf flavonoid content (FLAV); nitrogen balance index (NBI); leaf anthocyanins (ANT); plant height (H); height diameter ratio (H/D); shoot fresh weight (SFW); rootstock fresh weight (RSFW); root fresh weight (RTFW); total fresh weight (TFW); shoot dry weight (SDW); rootstock dry weight (RSDW); root dry weight (RTDW); total dry weight (TDW); fraction to shoot (FTS); fraction to rootstock (FTRS); fraction to root (FTRT). 0 = parameter recorded at 170 day of year (DOY); 1 = parameter recorded at 275 DOY; 2 = parameter recorded at 362 DOY.

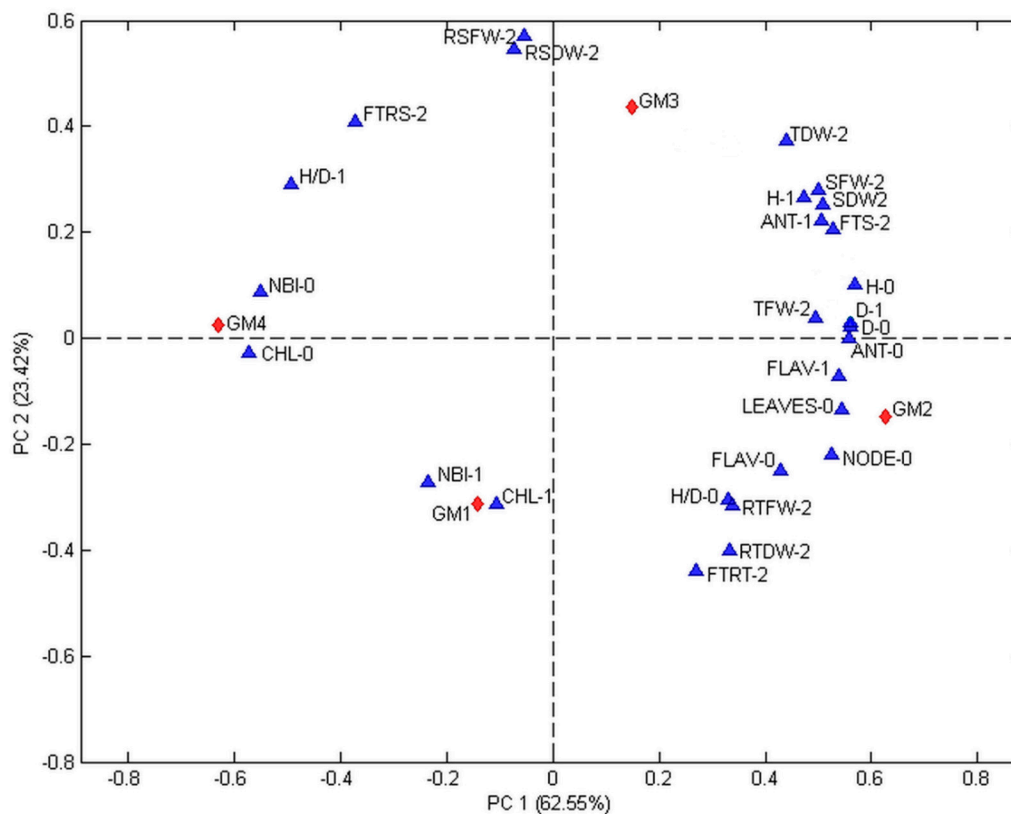
For two-year-old vines, CSD applied at 40% (GM4) recorded the best agronomic performance and was positively linked with several parameters, such as SDW, FTS, TDW, RTDW, and the shoot diameter (Figure 3).



**Figure 3.** Ordination biplots of principal component analysis outputs of the data from two-year-old vines grown on growing media (GM) containing composted solid digestate (CSD) at different rates. GM1 = commercial peat (CP) 100%; GM2 = CP 90% + CSD 10%; GM3 = CP 80% + CSD 20%; GM4 = CP 60% + CSD 40%. Labels in the triangles indicate the investigated parameters: Leaf chlorophyll content (CHL); leaf flavonoid content (FLAV); nitrogen balance index (NBI); leaf anthocyanins (ANT); plant height (H); height diameter ratio (H/D); shoot fresh weight (SFW); rootstock fresh weight (RSFW); root fresh weight (RTFW); total fresh weight (TFW); shoot dry weight (SDW); rootstock dry weight (RSDW); root dry weight (RTDW); total dry weight (TDW); fraction to shoot (FTS); fraction to rootstock (FTRS); fraction to root (FTRT). 0 = parameter recorded at 170 day of year (DOY); 1 = parameter recorded at 275 DOY; 2 = parameter recorded at 362 DOY.

In the second experiment, using BC for the bare-rooted vines, the PC1 accounted for 62.55% of the variance, while PC2 accounted for 23.42%. Growing media containing BC at 10% (GM2) clustered along the positive side of PC1, performing as well as growing media containing BC at 20% (GM3), and were associated with the most important parameters, such as CHL, FLAV, the number of leaves and nodes, the shoot height, SDW, TDW, and FTS (Figure 4).





**Figure 4.** Ordination biplots of principal component analysis outputs of the data from bare-rooted vines grown on growing media (GM) containing biochar (BC) at different rates. GM1 = commercial peat (CP) 100%; GM2 = CP 90% + BC 10%; GM3 = CP 80% + BC 20%; GM4 = CP 60% + BC 40%. Labels in the triangles indicate the investigated parameters: Leaf chlorophyll content (CHL); leaf flavonoid content (FLAV); nitrogen balance index (NBI); leaf anthocyanins (ANT); plant height (H); height diameter ratio (H/D); shoot fresh weight (SFW); rootstock fresh weight (RSFW); root fresh weight (RTFW); total fresh weight (TFW); shoot dry weight (SDW); rootstock dry weight (RSDW); root dry weight (RTDW); total dry weight (TDW); fraction to shoot (FTS); fraction to rootstock (FTRS); fraction to root (FTRT). 0 = parameter recorded at 170 day of year (DOY); 1 = parameter recorded at 275 DOY; 2 = parameter recorded at 362 DOY.

These results indicated that CSD and BC applied at 10% are able to increase the growth of potted bench-grafts and bare-rooted vines, reducing the consumption of CP. Moreover, working with plants with a preformed and well-developed root apparatus, such as in a two-year-old vine, CSD might be applied up to 40%. Finally, CSD and BC applied in low doses did not show any signs of abiotic and biotic stresses, in accordance with several previous reports [7,58,59].

#### 4. Conclusions

Results obtained in the present study suggest that CSD and BC could be a suitable ingredient for alternative growing media, recycling, and valorizing by-products coming from vineyards, such as winter prunings and grape stalks. CSD and BC showed good agronomic performances compatible with the development of grapevine planting materials. To our knowledge, this is the first study that investigated the suitability of CSD and BC valorizing vineyard prunings and grape stalks as an innovative ingredient for alternative growing media able to replace peat aliquots, for the production of different grapevine planting materials. Bench-grafts and bare-rooted vines grown in mixtures containing CSD at a dose of 10% displayed agronomic parameters higher than those recorded using only CP, and the same was displayed using BC on bare-rooted vines. On the other hand, CSD at a dose of 40% showed the highest agronomic performance on two-year-old vines. Agronomic measurements

suggest that the CSD or BC induce an enhanced plant-growth response that is particularly expressed at shoot level. The replacing of CP aliquots with CSD or BC could be an excellent sustainable practice to reduce the consumption of CP, recycling, and valorizing the underutilized vineyard prunings and grape stalks, thus supporting the current tendency to enhance the soilless propagation of grapevine. However, further research is needed to assess the combined use of CSD and BC as an ingredient of growing media for grapevine planting material productions.

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