



Research article

Process and quality evaluation of different improved composts made with a smart laboratory pilot plant

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ABSTRACT

This study monitored the process and investigated the quality of compost obtained from different biomasses. Five blends of agri-food waste were composted by a laboratory pilot plant named COMPOSTER, that is designed to optimize biodegradation, and produce compost efficiently. The COMPOSTER consists of two 35-liter nearly adiabatic, aerated bioreactors that simulate an industrial process involving the typical sequence of mesophilic-thermophilic-mesophilic phases. It continuously monitors and records temperature, internal pressure, and biomass weight, while controlling and quantifying oxygen consumption and carbon dioxide emissions resulting from aerobic biodegradation. All composts were characterized for their main chemical, physical, and molecular features, as well as their suppressiveness against *Fusarium oxysporum* f.sp. *lycopersici* (FOL), tested on tomato seedlings. Optimized biodegradation yielded 50–60 % mature compost with a cumulative oxygen consumption ranging from 282 to 456 gO₂ per kg of dry matter, with peaks of 2.55 gO₂ per kg of volatile solids per hour, and carbon dioxide emissions of 22–36 % of the initial carbon content, with peaks of 5.89 g CO₂ per kg of volatile solids per hour. Blends containing more ligno-cellulosic ingredients showed higher yields and lower CO₂ emissions. Most of the nitrogen present initially was retained in the final compost; indeed, all mixtures exhibited an apparent nitrogen concentration increase due to carbon loss. Composting determined deep modifications in the molecular structure of the organic matter. ¹³C CPMAS-NMR and off-line thermochemolysis GC-MS analyses highlighted decomposition degree of polysaccharides and peptidic moieties, selective preservation of aliphatic and aromatic recalcitrant compounds, and optimal ongoing humification. All composts were non-phytotoxic, except for that including pepper crop residues, and all resulted rich in macro- and micro-elements for plant nutrition and proved to be active in controlling FOL disease. Compost comprising 81.2 % tomato crop waste exhibited the best growth performance and pathogen control on tomato. Mature, non-phytotoxic, nutrient-rich, and suppressive composts represent promising by-products that can be successfully recycled in agriculture, including high-value applications, leading to lower use of fertilizers and pesticides.

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1. Introduction

Huge amounts of biomass residues are generated every year by horticultural cropping systems. Santana-Méridas et al. [1] estimated the production of green waste for tomato and pepper crops accounting for about 72.9 and 13.8 million of metric tons of biomass, respectively; moreover, olive, and other orchards annually produce different types of by-products that can be valorised for agronomic purposes [2]. Crop residues are usually removed from the fields to ease the successive cultivation and contain the spread of pathogens, but this requires appropriate management for possible agronomic valorisation [3]. Clean agri-food biomasses are becoming increasingly attractive as a valuable source of bioactive products (i.e., molecules and mixtures) for many agricultural uses through minimal preparation, as bio-refining or composting [4,5]. Composting is an accelerated aerobic biodegradation process able to transform wet biomasses into mature, dark organic matter smelling like a soil, useful in agriculture either as soil improver/fertilizer or as ingredient of nursery substrates [6]. The cultivation of plants in containers involves a small root zone compared to what happens in the open field crops; in such a condition, the substrate plays a key role in ensuring adequate storage of water and nutrients for plants, and good oxygenation for the roots. Sphagnum peat is widely used in the formulation of substrates for soilless cultivation [7]. It usually generates by the slow decomposition of plant biomass gathered in humid ecosystems characterized by low oxygen and acid pH that lower microbial biodegradation. Peat bogs cover different areas of the globe and are mostly located in the medium-high latitudes of the northern hemisphere; they were generated in the post-glacial period, approximately 14,000–11,000 years ago, and regenerate very slowly after extraction. For this reason, since the end of the 1980s, environmentalist lobbies have been calling for a reduction in use, encouraging replacement of peat with other “renewable” products [8]. Moreover, certain types of peat are conducive to several plant diseases which represents a critical issue, especially when used alone or as the sole organic component of the growth medium [9,10]. Hence, there is an increasing effort of the scientific community to find out alternatives to replace and/or integrate peat, and to create potting soil with improved agronomic quality, possibly even better than that of peat [11–13]. Among the possible substitutes for peat, coconut peat predominates, whose use has significantly increased worldwide since the early years of this century [14]. Sometimes, other organic materials are incorporated into the growth media such as bark, wood fibres, rice husks, etc., as well as composted materials, especially when available locally; inorganic materials are often also added, such as perlite, vermiculite, expanded clay and pumice, above all to improve the physical properties of the substrate. However, research recently emphasized the prospect to obtain high quality horticultural productions using substrates including compost mixed with other components [15–19]. Composting normally takes place in three phases which follow the temperature pattern: increasing at the beginning, up to a thermophilic stage ($T \approx 60\text{ }^{\circ}\text{C}$); decreasing after the peak, in the mesophilic phase ($T < 45\text{ }^{\circ}\text{C}$) and constant, in the curing phase (\approx room temperature). During thermal composting there is a high release of carbon dioxide associated with a high consumption of oxygen, due to the rapid biodegradation of the labile carbon fraction, i.e. carbohydrates, organic acids, proteins [20,21]; subsequently, the process continues more slowly at the expense of the more recalcitrant residual ligno-cellulosic components [22]. Among biomass feedstock, tomato crop residues have been tested in some studies aimed at optimizing the thermal kinetic by means of co-composting with heterologous feedstock (i.e., chicken manure, sawdust, almond shell powder and agri-food sludge) as strategy to improve the compost quality [23, 24]. On the other hand, pepper residues are little explored as feedstock for composting: few trials showed reduced palatability by non-ligno-cellulolytic microorganisms [25], and the need for integration with other feedstock [26].

This study aims at investigating how composting performs and which kind of product it makes when different biomasses are treated. Therefore, five blends of different agri-food waste mostly available in the Mediterranean countries, including tomato and pepper crop residues, wheat straw, olive mill waste and dry olive leaves, pig slurry, chicken and rabbit manure, broadleaves and conifer pruning, hydrolyzed collagen, were designed for ideal composting. Furthermore, this study aims to achieve the following

Table 1

Composition and basic features of the initial organic mixtures used in the composting trials.

ingredients	Mix #1	Mix #2	Mix #3	Mix #4	Mix #5
tomato crop residues	–	–	–	65.0	–
pepper crop residues	–	–	–	–	59.3
wheat straw	–	–	9.6	–	–
pig slurry	5.8	6.2	2.5	–	–
dry olive leaves	–	–	7.1	–	–
OMW (twigs and leaves)	–	–	14.8	35.0	40.7
OMW (3-phase husk)	–	–	60.1	–	–
chicken manure	5.8	6.9	–	–	–
rabbit manure	4.3	4.7	–	–	–
broadleaves prunings	84.1	45.9	–	–	–
conifer prunings	–	36.3	–	–	–
hydrolyzed collagen	–	–	5.9	–	–
basic features					
C:N	34.0	31.2	20.1	19.6	18.3
moisture, %	54.0	57.4	51.0	50.6	52.6
bulk density, Kg L ⁻¹	0.44	0.37	0.45	0.38	0.43

OMW = olive mill waste.

specific goals:

- i) manage key process parameters to ensure proper biomass sanitization [27] and conduct a mass balance analysis of composting
- ii) keep oxygen levels into the biomass within the optimal range (>10 % ,vol:vol) to prevent undesired anaerobic fermentations, minimize nitrogen loss [28], and promote the formation of bioactive humic compounds
- iii) assess the potential agronomic value of the composts, in terms of chemical, physical, molecular, and biological features, with a specific focus on suppressive effects against tomato tracheofusariosis.

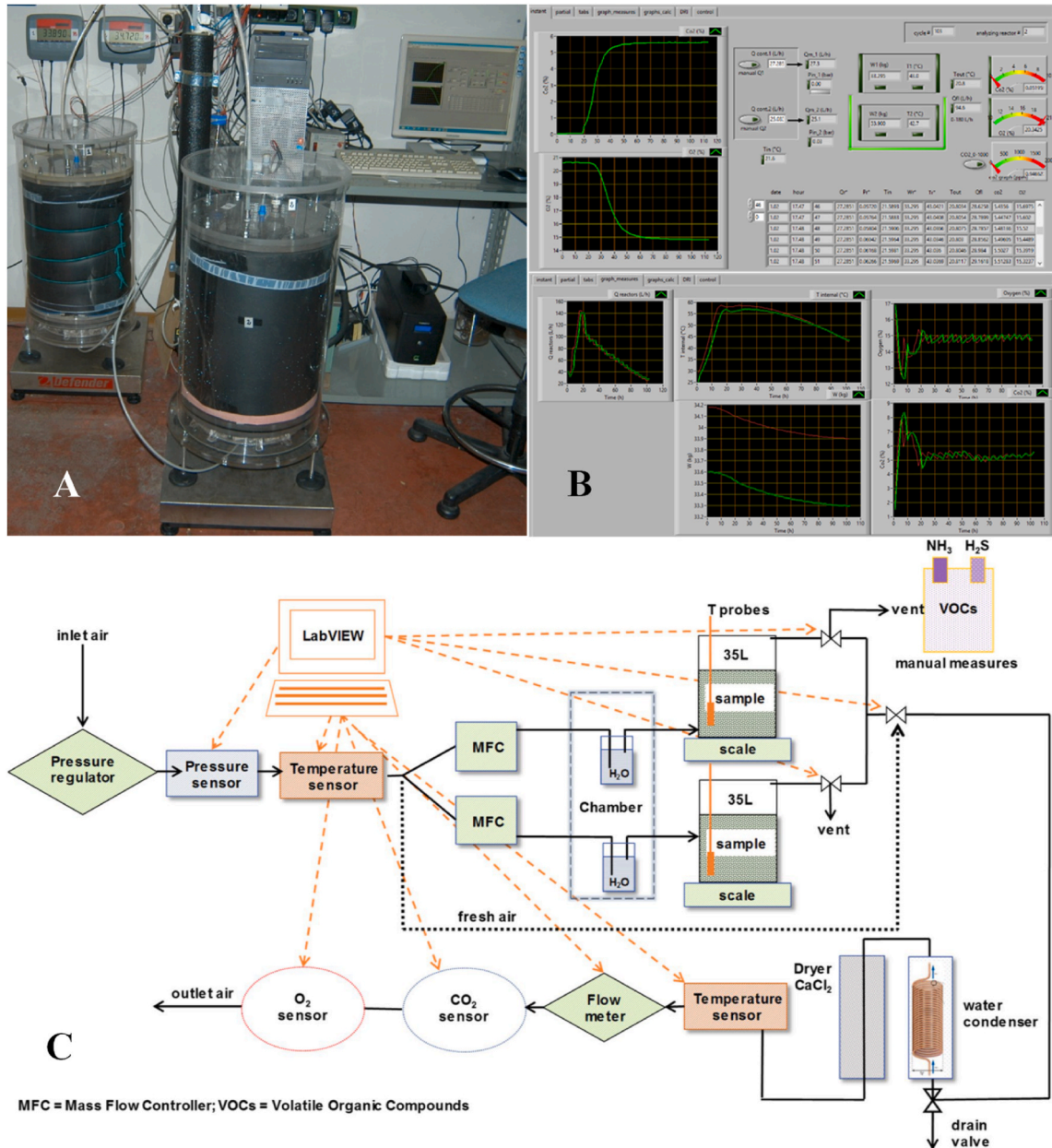


Fig. 1. Lab-scale COMPOSTER used for composting trials (A); screenshots showing the cleaning phase of the measuring gas line (B upper) and flow rates, temperature, gross weight, O₂(%) and CO₂(%) monitoring (B bottom); schematic diagram of the COMPOSTER (C), from Altieri et al. (2021).

Table 2Characterization of agri-food waste used for making compost with COMPOSTER. Data are means of triplicates with coefficient variation $\leq 10\%$.

	Moisture % (fm)	ash	C % (dm)	N % (dm)	S	Ca	Mg	K g kg ⁻¹ (dm)	Na	P	Fe	Mn	Cu	Zn	Al g Mg ⁻¹ (dm)	Pb	Cr	Ni	Mo	As
tomato crop residues	93.8	18.8	38.4	2.0	0.8	32.9	7.9	28.0	1.5	2.4	0.7	64.4	369.3	31.7	798.4	0.9	3.4	2.6	3.6	bdl
pepper crop residues	84.4	16.3	41.2	2.4	0.6	20.3	6.3	49.5	0.0	4.6	0.2	56.2	83.4	40.1	248.7	0.2	1.3	3.8	0.8	bdl
pig slurry	94.5	26.5	39.1	3.7	1.3	24.9	14.6	16.9	5.3	39.8	4.3	1224.0	2060.0	6567.0	1503.0	3.9	21.8	14.9	17.7	1.7
wheat straw	16.4	7.4	41.7	0.6	0.2	5.1	1.1	19.6	0.0	0.6	0.1	37.7	8.7	42.0	50.0	0.2	8.9	3.6	0.8	bdl
Dry olive leaves	5.0	5.4	50.0	2.2	0.1	14.5	1.3	12.3	0.1	2.0	0.1	25.3	15.4	17.9	85.2	bdl	1.3	2.1	0.3	bdl
OMW- twigs and leaves	50.0	8.5	49.4	2.4	0.3	22.2	1.9	10.6	0.1	1.4	0.3	67.6	81.4	28.3	264	1.2	2.4	6.4	bdl	bdl
OMW- 3-phases husk	42.6	1.8	50.6	1.3	0.2	1.8	0.2	8.0	0.0	3.5	0.1	6.9	6.2	8.5	24.3	0.1	2.7	1.3	0.3	0.3
chicken manure	14.6	29.0	35.3	3.4	0.5	85.7	4.9	27.5	4.0	17.2	0.6	477.8	65.0	352.0	312.0	0.2	8.2	6.2	4.8	0.6
rabbit manure	58.4	50.0	26.2	2.0	0.6	62.1	8.4	18.7	2.9	14.2	6.0	554.0	90.0	462.0	5927	4.1	27.3	20.4	4.3	1.6
broadleaves prunings	11.4	3.7	47.6	1.0	0.2	9.7	1.0	5.2	0.1	1.1	0.2	44.7	7.8	18.9	110.7	0.5	2.6	2.0	0.3	0.3
conifers prunings	29.8	7.8	48.0	1.2	0.1	20.1	0.8	5.6	0.2	2.5	1.4	94.3	22.2	76.4	538.2	7.2	4.8	3.9	1.4	0.1
hydrolyzed collagens	6.0	4.3	45.7	16.5	0.4	9.8	0.0	0.6	4.8	0.0	0.0	0.7	0.8	2.3	7.4	bdl	10.4	0.8	0.7	0.2
OMW- 2-phases husk	65.0	5.4	53.4	2.1	0.1	1.9	0.8	33.2	0.1	5.2	0.1	12.8	9.1	17.3	132.5	0.2	0.9	0.5	0.5	0.4
rice husk	8.5	17.3	40.6	0.6	0.0	1.0	0.4	2.9	0.0	0.9	0.3	214.1	2.5	16.8	46.5	0.0	40	12.3	2.3	bdl
coconut peat	11.0	3.3	49.6	0.4	0.1	1.6	0.4	5.1	1.6	0.2	0.2	10.0	4.2	10.2	93.4	bdl	22.1	7.4	1.3	bdl
sphagnum peat	51.4	3.0	49.9	1.1	0.1	3.7	0.6	0.8	0.1	0.3	0.6	19.9	1.8	12.4	651.5	4	6.1	2.3	0.5	bdl

fm = fresh matter; dm = dry matter; bdl = below detection limit; OMW = olive mill waste.

2. Materials and methods

2.1. Feedstock selected for composting

A wide array of waste from different agri-food supply chains was used for preparing five organic mixtures for composting (Table 1), carried out in a Lab pilot plant, namely COMPOSTER (Fig. 1), set up at CNR-ISAFOM, Perugia for research studies [29]. Waste rich in water were mixed with hygroscopic bulking agents including wheat straw, dry olive leaves, broadleaves and conifer pruning, able to guarantee a balanced micro and macro porosity that ensure good oxygenation and sufficient thermal inertia of the biomass during composting. The following running conditions were guaranteed at the beginning of each trial: moisture content not exceeding 50–55 % which prevents leaching during the process; bulk density around 0.4 kg dm^{-3} ; carbon to nitrogen ratio in the range 18–35. Animal residues were added in mix #1, #2 to lower the C:N of those blends rich in ligno-cellulosic ingredients; the C:N of mixture #3 was adjusted adding pig slurry and hydrolyzed collagen (HC) as main nitrogen source. HC was provided by ILSA company as fine dry powder (Etixamin DF®) and obtained through enzymatic hydrolysis of tanned leather shavings.

To ensure an easy implementation of the proposed composting approach at farm scale, it was decided to use selected organic waste without any pre-treatments, such as drying. Therefore, based on the specific chemical characteristics of each waste, as outlined in Table 2, initial blends were formulated to achieve a balance of moisture content (50–55 %) and the correct C/N ratio. However, priority was given to maintaining the appropriate moisture level, which is crucial for preventing mass leaching during composting. Consequently, the low C/N ratio in blends #4 and #5 is attributed to the inherent characteristics of the wastes used, mainly tomato and pepper cultivation residues respectively, which naturally contain high levels of nitrogen (Table 2). In the case of blend #3, the initial C/N ratio was reduced to around 20 by adding dry nitrogen-rich hydrolyzed collagen. This was done to evaluate the efficiency of the COMPOSTER in preventing potential nitrogen losses, particularly in the form of ammonia.

2.2. Composting in a lab pilot plant

As closed-vessel system, COMPOSTER can monitor, control, and optimize the thermal phase of composting: it includes two almost adiabatic and aerated 35 L bioreactors (Fig. 1) that perform as an industrial process, resulting in typical sequence of mesophilic-thermophilic-mesophilic phases. Every 10 s, it regularly records temperature, internal pressure, and weight of the biomass and controls and quantifies oxygen consumption and carbon dioxide emission resultant from aerobic microbial biodegradation. Each bioreactor was loaded with about 12–14 kg of organic matter (fresh weight). Separate mass flow controllers (Model F-201CV-10K-AGD-22-V, 500 Ln h^{-1} , Bronkhorst® High Tech, NL) modulate the oxygen in the exhausted air at chosen values (14–16 vol% O_2), thus ensuring proven aerobic biodegradation. Fresh air flows from the bottom (inlet) to the top (outlet) of the vessels. Carbon dioxide and oxygen are detected by specific online gas detectors: zirconium dioxide oxygen sensor (model XYA5M, equipped with oxygen sensor circuit board ZBXYAF, FirstSensor, DE) and Gascheck CO_2 Sensor (0–10 vol% CO_2) by Edinburgh Sensors Ltd (UK). An alternate measuring/cleaning sequence ensures proper monitoring of O_2 and CO_2 concentrations, collecting one data set per hour. O_2 and CO_2 concentration (vol: vol), assessed in each measurement cycle, were automatically converted to mass values, and then cumulated, with the following general equation (1):

$$CO_{2\text{sample}} = \frac{Q \cdot CO_2(\text{or } O_2) \cdot \Delta t \cdot M(CO_2 \text{ or } O_2)}{V_m}, \quad (1)$$

where Q is the flow rate measured with the mass flow controllers, CO_2 or O_2 is the concentration measured with the online gas detectors and corrected for the background CO_2 or O_2 concentrations of the inlet fresh air, Δt is the period (1 h) of the measurement cycle, $M(CO_2$ or $O_2)$ is the molar mass of CO_2 or O_2 , and V_m is the volume occupied by 1 mol of CO_2 or O_2 at the exhaust-air temperature, as determined by the local in-line sensor. The molar volume of CO_2 or O_2 is calculated using equation (2):

$$V_m = R \cdot \frac{T_{out}}{P_{atm}}, \quad (2)$$

where R is the ideal gas constant, T_{out} is the temperature of the outlet flow and P_{atm} is the atmospheric air pressure constantly checked by the online pressure transmitter 628CR (Dwyer®). Acquisition of signals, control of actuators, remote software supervision, data elaboration and monitoring perform by means of fit-for-purpose integration of hardware and software created in NI LabVIEW® environment. Parameters are displayed and plotted in spreadsheets showing biodegradation patterns in real time (SM). When temperature drops below 40°C and without stopping data logging, bioreactors are temporarily emptied and mixtures turned, thus assuring faster and more homogeneous biodegradation. At the end of the thermophilic phase, that usually lasts 4–5 weeks, bioreactors are emptied, and the biomass placed in a tray, allowing the mesophilic curing phase to take place at room temperature. An adequate moisture content (50–60 wt%) is ensured up to the end of the curing.

2.3. Chemical-physical analysis

All organic ingredients and mixtures, sampled at the beginning and end of composting, were analyzed for their chemical-physical and biological properties. Total carbon, nitrogen and sulfur were determined on aliquots (50–100 mg) of dry samples by CHNS analyzer Vario Macro Cube (Elementar, Germany). Dry samples were previously ground ($<1 \text{ mm}$) using a milling system (Retsch ZM

200). The ash content was determined as weight loss on dry samples at 650 °C for 24 h in a muffle furnace and used for estimating the organic matter (OM) loss, as described by Viel et al. [30]. Total phosphorus and metals were determined on dry, digested samples in a closed-vessel microwave system (Ethos D, Milestone, Germany), using the US-EPA method 3051 A [31]. Digested samples were then analyzed by Inductively Coupled Plasma Optical Emission Spectrometry (ICP-OES ICAP 7200, Thermo, USA). Electrical conductivity (EC), pH, ammonia-N, soluble inorganic anions and water soluble C and N were determined on filtered deionized water extracts obtained from fresh samples shaken for 15 min (1:10 w:v). Ammonia-N was determined according to the method 4500-NH₃ Nitrogen [32]. Inorganic anions, phytotoxicity, soluble carbon and nitrogen were determined with procedures detailed in Seggiani et al. [33]. The water extracts, previously acidified with nitric acid, were also analyzed by ICP-OES ICAP 7200 (Thermo, USA). Cation Exchange Capacity (CEC) was determined on 0.2 g air-dried samples, as described by Harada and Inoko [34]. Dynamic respiration index (DRI) was calculated according to the method UNI/TS 11184 [35]; DRI values were hourly corrected for volatile solids loss, estimated on the base of carbon dioxide emission recorded. The physical properties of samples were checked in triplicates using standard procedures [36]. Particle density (PD) was calculated basing on volatile solids (VS%) and ash% contents, using the following equation:

$$PD \left(\frac{\text{kg}}{\text{L}} \right) = \frac{1}{\frac{VS\%}{(100 \cdot 1.55)} + \frac{ash\%}{(100 \cdot 2.65)}}$$

As well, based on the relationships existing between physical properties, the following equation were also considered:

$$TP (\%) = 100 \cdot \left(1 - \frac{BD}{PD} \right)$$

$$TP = \text{WHC} + \text{AS}$$

where TP = total porosity; BD = bulk density; PD = particle density; WHC = water holding capacity; AS = air space.

2.4. ¹³C CPMAS NMR and thermochemolysis GC-MS

Fine-powdered samples were analyzed by solid-state NMR spectroscopy (¹³C CPMAS NMR) on a Bruker AV300 Spectrometer (Bruker Instrumental Inc, Billerica, MA, USA) with operational parameters usually applied for complex matrices [37,38]. The different signals highlighted in solid state ¹³C NMR spectra, are conventionally grouped into 6 main chemical shift regions, representative of the major types of carbon functional groups: alkyl-C (0–45 ppm); methoxyl-C and N-alkyl-C (45–60 ppm); O-alkyl-C (60–110 ppm); un-substituted and alkyl-substituted aromatic-C (110–145 ppm); O-substituted aromatic-C (145–160 ppm); carboxyl- and carbonyl- C (160–190 ppm) [37]. The ith area of each functional group (Riabs) was divided by the sum of all spectral zones, to obtain a semi-quantitative evaluation of carbon distribution (MestreNova 6.2.0 software, Mestre-lab Research, 2010). To summarize the structural differences among diverse organic materials, specific dimensionless structural indexes are calculated from the relative amount of specific C functionalities in the NMR spectra [38]: Hydrophobic index (HB), Alkyl ratio (A/OA), and Lignin ratio (LR); the corresponding formulas are included in supplementary materials (SM). The HB and A/OA have been extensively applied to estimate the biochemical stability and bioactivity organic materials, while the LR is a useful indicator to discriminate between signals owing to lignin and other phenolic derivatives (lower LR) with respect to the prevalent inclusion of peptidic moieties in the 45–60 ppm interval (larger LR). For the off-line thermally assisted hydrolysis and methylation (TAHM) GC-MS about 100 mg of compost samples were moistened with 0.5 ml of tetramethylammonium hydroxide (25 % in methanol) solution and subjected to pyrolysis at 400 °C for 30 min). The released molecules were finally redissolved in 1 ml of chloroform and transferred to a glass vial for GC-MS analysis [37,39]. Compound identification was based on comparison of mass spectra with the NIST library database, and published spectra.

2.5. In planta compost suppressiveness assay

Solanum lycopersicum L. landrace so called ‘Cuore di bue’ seeds (Enza Zaden, Tarquinia, Italy) were sown in sterile peat-filled 20 mL pots and let them to germinate in the dark at 25 °C. Seedlings were then maintained in growth chamber at 25 °C with a 12 h photoperiod, and the irrigation was manually performed twice a week. After 15 day-growth, seedlings were gently removed from the pots and thoroughly rinsed in sterile water and submitted to artificial infection with *Fusarium oxysporum* f. sp. *lycopersici* (Sacc.) Snyder & Hans. Race 1 (FOL). Root tips were then cut, and the infection was carried out dipping roots in a FOL aqueous conidial suspension (10⁷ conidia mL⁻¹) for 10 min. FOL conidial suspension was obtained from ten-day-old cultures grown on potato dextrose agar medium (PDA, Condalab, Madrid, Spain) maintained at 25 °C. Infected seedlings were then transferred to 50 mL pots containing sterilized peat amended with 30 % (w/w) of compost. Healthy controls were obtained dipping roots into sterile water. Pots containing 3 plants each, were incubated at 26 °C in growth chamber for 20 days. Experimental design included 12 pots per compost, infected (FOL) and healthy control (CNT). Disease incidence (DI, %), and disease severity assessments (DS, %) were carried out at 20-days post infection (dpi). DI% was calculated as the percentage of symptomatic plants on the total ones while DS was assessed using a 0–4 scale adapted from Larkin and Honeycutt [40]: 0 = no symptoms, 1 = wilting, 2 = stunting, 3 = yellowing/desiccation and 4 = dead plants. DS was then elaborated according with the formula reported by Chiang et al. [41]. At the end of the assay, biometric parameters (root and stem length, fresh and dry weight, number of leaves) as well as plant mortality (%) were also assessed.

2.6. Statistical analyses

Ordinary one-way ANOVA was applied to evaluate compost effects on the measured parameters. Analysis of variance was corrected for multiple comparisons by the Bonferroni hypothesis test, considering a p-value ≤ 0.05 .

3. Results

3.1. Characterization of feedstocks used for composting trials

Table 2 shows the main chemical features of the organic ingredients used for making compost for this study. The high ash content, found mainly in tomato (TC) and pepper (PC) crop residues, in chicken and rabbit manure, can be justified by their contamination with soil during harvesting. Among plant biomasses, TC, PC, and olive mill waste (OMW) showed high N, K and Ca content, as well as micronutrients, that can be all usefully stored in the mature compost if they do not leach during composting. TC and PC showed a C to N ratio slightly below 20 (Table 2) thus providing adequate N to support reproduction and metabolism of microbes driving degradation [42]. Micronutrients and heavy metals were low in content in all plant biomasses, except Cu in TC; the large use of cupric fungicides used against common tomato diseases may explain the abundant Cu in the tomato residues, especially found in the leaves (data not showed); therefore, the use of TC must be careful managed to ensure Cu below the allowed limit (230 ppm) in the mature compost [43, 44]. The high N content (16.5%), most delivered as amino acids (data provided by ILSA spa), and dryness of HC helped to optimize the C:N of the mixture #3, while keeping moisture around the target value. Among animal wastes, pig slurry was the richest in metals, especially Cu and Zn; therefore, it must be added in low amount to prevent excesses in the mature compost. Dry wheat straw, olive leaves, conifer and broadleaves pruning were used as bulking agents to better aerate biomass and inhibit leaching during composting. Conifer pruning showed higher concentration of minerals than broadleaves, but still very low, if compared to the other ingredients used in this study. Both OMW residues, used in the mixture #3, #4 and #5, showed high amount of N and K that make them interesting for agronomical purposes; indeed, previous investigation confirmed OMWs as excellent by-products to compost, resulting in end products rich in humus-like substances that showed good agronomic performances, both in field and potted cultivation trials [45–47]. Table 2 also includes analyses gathered on rice husk, coconut, and sphagnum peat, used here just for comparison and database: they all showed a very low potential for plant nutrition, with heavy metals largely under the limits set by the rules; therefore, they offer few nutrients and just the physical support to grow the plants, requiring frequent fertigation if used as cultivation substrate.

3.2. Composting trials

The optimized biodegradation carried out in COMPOSTER returned approximately 50–60% of mature compost, in terms of volatile solids, typical values of composting [48]. As soon as blended, biomasses started to self-heat soon, showing intense microbial activity since the beginning; indeed, all mixtures entered the thermophilic phase ($T > 45^\circ\text{C}$) in less than 24 h. The thermal phase proceeded for several days, mix #1 mainly including broadleaves pruning, the shortest period, 8 days, and mix #3 mainly including olive mill waste, the longest one, 26 days (Fig. 2), all trials ensuring the minimal target for proper biomass sanitization [43]. During composting all mixtures were turned twice, except mix #3, only once on day 21st, because of the optimal ongoing thermal phase. Mixing always resulted in peaks of oxygen consumption and carbon dioxide emission, as showed by DRI and CO_2 emission growing trends (Fig. 2). In all cases, the second mixing had a lower effect on temperature, oxygen consumption and carbon dioxide emission, suggesting a progressive disappearance of the labile C fraction (carbohydrates, proteins, fatty acids, etc.) that supported microbial metabolisms. Despite differences in the initial C:N, all mixtures required hourly average aeration rates comparable to each other's (Table 3) and

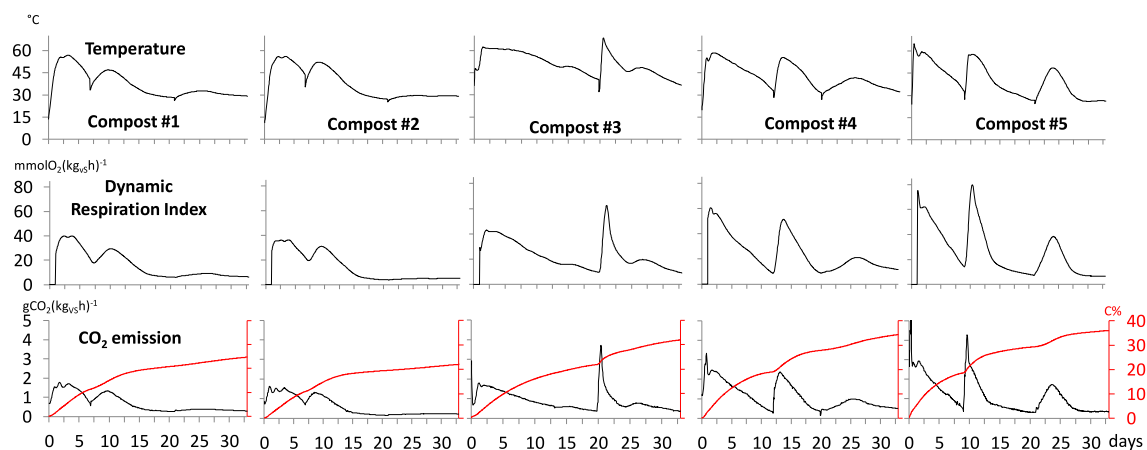


Fig. 2. Temperature, dynamic respiration index and CO_2 emission patterns recorded during the thermal phase of composting carried out in COMPOSTER.

Table 3Composting trials: mass balance assessed at the end of the thermal phase, aeration rate, cumulated O₂ consumption and CO₂ emission.

COMPOSTING TRIALS		Mix #1	Mix #2	Mix #3	Mix #4	Mix #5
weight loss	%	11.2	10.3	15.6	16.3	12.1
dry matter loss		28.0	25.0	36.5	36.3	40.1
volatile solids loss		31.3	27.3	38.1	45.3	46.2
carbon loss		24.7	22.0	31.8	34.3	36.0
water gain		3.1	0.6	6.7	3.1	13.0
aeration rate, hourly average	L (h kg _{DM}) ⁻¹	8.4	7.8	10.2	7.8	9.6
aeration rate, max		20.4	19.2	32.4	27.6	36.6
oxygen consumption	gO ₂ kg _{DM} ⁻¹	309	282	456	380	444
carbon dioxide emission	gCO ₂ kg _{DM} ⁻¹	411	367	539	533	590

DM = dry matter.

largely lower than those reported in literature [49]: the lack of constant airflow control, as provided in this study, probably led authors to give a surplus of airflow rate which wastes energy and can even trigger undesired dehydration and lower the speed of biodegradation. Mix #3 recorded the highest cumulated O₂ consumption, 456 gO₂ kg_{DM}⁻¹, and the highest hourly aeration rate, 10.2 L (kg_{DM}h)⁻¹; mix #3 consumed more oxygen even than mix #5 mainly including pepper crop residues, that showed the highest CO₂ emission (590 gCO₂ kg_{DM}⁻¹), corresponding to 36.0 % of carbon loss (Table 3). The explanation of this apparent anomaly could lie in the fact that mix #3, being rich in organic N, could have triggered more intense oxygen-driven nitrification [50], proven by the presence of nitrate in the water extract (Table 5). DRI is a well-recognized index of the compost biological stability [51]: basing on that, the thermal phase of composting was considered over on day 33rd for all trials. Indeed, DRI at that time reached values lower than 25 mmol (kg_{vsh})⁻¹, the limit considered safe even for a direct agronomic use [44]. Table 3 shows also mass balances recorded at the end of the thermal phases: in comparison to mix #1 and #2, most including ligno-cellulosic ingredients, mix #3, #4 and #5, richer in labile carbon, showed the highest losses; moreover, all mixtures showed water gain resulting from aerobic degradation of organic compounds, the highest, 13 %, in the mixture #5; water gain occurred differently, in part offsetting the weight loss imputable to CO₂ emission.

3.3. Chemical-physical and biological features of the compost

Tables 4 and 5 show physical and biological properties of the five experimental organic mixtures. Biodegradation, quantified by the CO₂ emission recorded (Fig. 2), determined a strong reduction in the C:N, more evident for mix #1 and #2 both mainly including pruning and animal residues; however, C:N values tend to align with each other's at the end, regardless of the initial composition; the optimal operating conditions [49] ensured by the COMPOSTER, such as good aeration and humidity, and thermal inertia, allowed to lessen the emission of ammonia and to store most of the initial nitrogen in the biomass, even in the case of mix #3 where nitrogen was added in a labile form (amino acids): as a result, the nitrogen content increased in all different composts, doubling in the majority of cases (Table 4). The composting trials were not carried out at the same time but in a temporal sequence, starting from mixture #1 up to #5: it follows that the first compost had a longer *curing* time and gradually decreased for the subsequent ones. This could explain the higher content of mineral N (mainly nitrates) found in the first three composts resulting from a longer lasting organic-N mineralization over time [52]. However, most of the nitrogen was still in organic form (>80 %), as required by the current legislation [43], even in the case of compost #1 which underwent a longer *curing*. Among the other macro-elements of plant nutrition, compost #4 and #5, mainly including tomato and pepper crop residues respectively, showed higher K, 29.9 and 41.5 g kg⁻¹, respectively (Table 4), explained by the richness in K of the main ingredients used (Table 2). Compost #1 and #2 resulted richer in Ca, 52.8 and 51.2 g kg⁻¹, respectively, while compost #4 and #5 were higher in Mg, 6.3 and 7.0 g kg⁻¹, respectively. All cured composts show little amount of phosphorus, with the maximum, 9.4 g kg⁻¹, found in compost #2 mainly including conifer pruning. Regarding Mn, Cu, Zn, Mo, Fe, microelements of plant nutrition, there is means of them in all compost. In compost #1, #2, and #3 Zn exceeded the safe limit (500 ppm). Zn clearly derives from pig slurry (Table 2) which must be carefully managed in composting when it aims at high-quality end-products. Also, Cu exceeded the allowed limit (230 ppm) in some cases: the origin of Cu relies on pig slurry and chicken manure for compost #1 and #2, respectively, and reasonably to cupric pesticides, accumulated in tomato crop residues, for compost #4. Regarding heavy metals, all compost were largely within the limits allowed by the current legislation. Al, high in compost #1, #2 and #4, derived from contamination with soil particles; however, Al solubilized little in water (Table 4), making it unavailable for plants thus reducing possible phytotoxic effects. Looking at water extracts, most of the K (from 69 % up to 86 %) and ammonia (>54.8 %) solubilized; therefore, they are both easily available for crops and responsible of the related high EC recorded. K contributed to over 70 % of the whole cation content in the water extract, with peaks of 90 % for compost #4 and #5 mainly including tomato and pepper crop residues, respectively. On the other hand, given their lower overall solubility, Ca and Mg resulted low in the water extracts, despite their high content found in all bulk compost, especially Ca. Similarly, the low solubility of Cu and Zn kept them at low concentration in the water extracts, even in the richer compost #1 and #2. As regards anions, compost #1, #2 and #3 showed a valuable nitrate content, in line with expectations due to different times left for nitrification which was the only natural source of nitrates. All compost showed small content of phosphates, while compost #4 and #5 large amount of sulphates and chlorides, perhaps deriving from foliar fertilizers and/or pesticides commonly used in tomato and pepper cultivation and accumulated in the crop residues (i.e. copper sulphate, broadly used as fungicide, and/or calcium chloride, widely applied against apical fruit-rot). The longer curing of compost #1,

Table 4Characterization of the starting mixtures and cured compost; water extracts relate to cured composts. Data are means of triplicates with coefficient variation $\leq 10\%$.

	dm % (fm)	ash % (dm)	C	N	S	Ca g kg ⁻¹ (dm)	Mg	K	Na	P	Fe	N-NH4+ g Mg ⁻¹ (dm)	Mn	Cu	Zn	Al	Pb	Cr	Ni	Mo	As	C/N	OMloss %
COMPOST #1																							
starting mix	46.0	19.4	45.5	1.3	0.3	17.2	2.3	7.7	0.8	4.8	0.7	–	159.9	133.5	436.4	450.5	0.9	5.0	3.7	1.7	0.5	34.0	–
cured	27.5	25.9	40.9	2.8	0.4	52.8	4.5	17.4	1.5	8.0	2.6	120.3	341.7	250.3	1007.8	2049.4	5.7	14.6	12.7	4.1	1.7	14.4	31.3
we (1:10 w:v)	–	–	0.7	0.2	–	0.7	0.3	12.8	0.7	0.9	0.0	65.9	0.6	5.5	10.3	5.2	1.0	bdl	2.8	0.1	0.9	–	–
COMPOST #2																							
starting mix	42.6	22.5	45.4	1.5	0.3	22.1	2.4	8.2	0.9	5.7	1.2	–	189.1	147.1	486.4	639.4	3.3	6.1	4.6	2.3	0.4	31.2	–
cured	28.9	28.6	38.3	2.9	0.5	51.2	4.7	16.0	1.8	9.4	3.3	145.4	369.8	311.7	1292.8	2364.7	9.5	16.6	15.3	5.0	1.9	13.1	27.6
we (1:10 w:v)	–	–	0.6	0.3	–	1.2	0.6	11.1	0.8	0.9	0.0	87.6	0.4	5.0	9.8	3.2	0.7	bdl	2.6	0.1	0.8	–	–
COMPOST #3																							
starting mix	51.7	5.3	49.0	2.4	0.2	7.1	1.0	9.6	0.5	3.5	0.2	–	49.8	68.6	176.7	102.0	0.4	4.1	2.6	0.8	0.2	20.1	–
cured	32.9	8.4	48.3	4.1	0.4	11.2	1.8	15.2	2.2	3.9	0.5	192.7	117.0	170.5	530.4	309.2	1.9	5.6	5.6	1.9	1.3	11.9	38.8
we (1:10 w:v)	–	–	1.2	0.2	–	0.9	0.3	11.8	1.1	2.5	0.0	117.0	0.4	5.5	12.5	1.6	1.0	0.1	2.4	0.0	0.6	–	–
COMPOST #4																							
starting mix	49.2	21.0	42.2	2.2	0.6	29.1	5.8	21.9	1.0	2.1	0.5	–	65.5	268.5	30.5	611.3	1.0	3.1	3.9	2.3	bdl	19.6	–
cured	43.5	32.8	35.1	2.7	0.8	45.6	6.3	29.9	1.3	6.3	4.2	95.7	120.2	283.8	82.1	3791.8	5.1	23.8	16.6	3.2	1.9	13.1	45.5
we (1:10 w:v)	–	–	1.0	0.0	–	1.2	0.6	25.8	1.0	0.2	0.0	90.7	0.5	6.0	5.2	3.4	1.0	0.0	1.8	0.5	0.7	–	–
COMPOST #5																							
starting mix	47.4	14.5	44.5	2.4	0.5	21.1	4.5	33.7	0.1	3.3	0.2	–	60.8	82.6	35.3	254.9	0.6	1.7	4.9	0.5	0.0	18.5	–
cured	39.7	23.9	42.5	3.1	0.7	35.3	7.0	41.5	0.8	5.8	1.0	70.8	57.8	64.2	54.7	550.2	1.0	3.7	4.3	0.7	0.4	13.8	46.1
we (1:10 w:v)	–	–	1.7	0.1	–	0.5	0.4	32.6	0.6	2.3	0.0	96.5	1.3	3.9	1.8	0.8	0.6	0.0	0.1	0.0	0.3	–	–

fm = fresh matter; dm = dry matter; below detection limit; we = water extract.

Table 5Other chemical-physical and biological features of cured compost. Data are means of triplicates with coefficient variation $\leq 10\%$.

COMPOST		#1	#2	#3	#4	#5
bulk density	$Mg\ m^{-3}(dm)$	0.14	0.16	0.17	0.20	0.19
particle density		1.74	1.76	1.61	1.79	1.72
total porosity	$m^3\ m^{-3}$	0.92	0.91	0.90	0.89	0.89
water holding capacity		0.56	0.56	0.57	0.62	0.61
air porosity		0.36	0.35	0.32	0.26	0.28
pH		6.86	6.77	6.35	7.50	9.25
EC	$dS\ m^{-1}$	1.41	1.67	1.84	4.35	4.06
phosphates	$g\ kg^{-1}\ (dm)$	1.3	1.3	3.9	0.1	0.4
sulphates		2.0	2.8	bdl	13.6	9.7
chlorides		2.9	2.8	5.3	18.1	13.1
nitrates		12.7	15.1	11.4	bdl	bdl
CEC	$me\ 100g^{-1}(dm)$	92	94	75	56	56
GI	%	90.2	100.5	99.7	81.2	24.8

bdl = below detection limit, CEC = cation exchange capacity; GI = germination index.

#2 and #3 reflected in sub-acid pH, while compost #4 and #5 showed a sub-alkaline reaction typical of fresh, younger compost (Table 5). A possible explanation of the pH reduction over time is the following: after the thermal phase, compost is colonized by fungi which slowly biodegrade the residual recalcitrant components (ligno-cellulosic moieties) producing organic acids; at the same time, oxidative polymerization reactions take place and slowly incorporate ammonia in the molecular structures of the organic matter, buffering the alkalinity mostly ascribed to ammonia. The biological nitrification gradually removes ammonia as well, lowering the compost pH [53,54]. The high EC especially found in compost #4 and #5 (Table 5) may limit their value-added uses, such as ingredient of growing substrates; however, preliminary water leaching or mixing with materials lower in soluble salts (i.e. peat moss, coconut peat, etc.) could guarantee the final substrate safeness. All compost, except #5 mainly including pepper crop residues, resulted no phytotoxic with GI values $> 60\%$ [43]. The shortest and probably not completed maturation of compost #5, associated to an alkaline pH (9.25), may explain why it hindered the germination of the *L. sativum* seeds. All compost showed CEC values in line with those reported in the literature for compost differently aged [55]; moreover, a clear rising trend of CEC emerged with the progress of curing time, confirming this parameter as suitable index of compost maturity [56]. All composts showed total porosity in the optimal range ($>80\text{--}85\%$ vol.) for plant nursery substrates. Moreover, compost #4 and #5 showed water holding capacity 62.4 and 61.3 % vol., respectively, slightly higher than those found in the first three compost. Also, the air porosity almost matched the target value (30 % vol.) recommended for substrates while the particle and bulk density values were in line with those reported in the literature for other organic matrices often used in the formulation of nursery substrates [14].

3.4. ^{13}C CPMAS NMR analyses

The ^{13}C CPMAS NMR spectra of starting biomasses (SM) revealed the predominance of O-alkyl-C components, in carbohydrates and polysaccharides which relative content ranged the 57.5 % up to the 60.0 % of total area, respectively (Table 6). This finding must be associated with the inclusion of large amount of plant residues and by-products in the initial mixtures (Table 1). The various peaks shown in the 60–110 ppm chemical shift, are the typical resonances of O-alkyl-C in monomeric units of polysaccharide chains such as cellulose and hemicellulose of plant tissues [37]. The carbon nucleus in position 6 at 64 ppm is followed by the intense coalescence around 73 ppm composed by the spectral overlay of carbon 2, 3, and 5, in pyranoside structure. The less evident shoulders at 83/88 ppm derive from the carbon 4, in either crystalline or amorphous structures, involved in the glycosidic bond with most deshielded

Table 6Relative distribution (%) of signal area over chemical shift regions (ppm) in ^{13}C -CPMAS-NMR spectra and derived structural index of starting organic mixtures (t_0) and final composts (t_{fin}).

COMPOST	#1		#2		#3		#4		#5	
	t_0	t_{fin}	t_0	t_{fin}	t_0	t_{fin}	t_0	t_{fin}	t_0	t_{fin}
Carboxyl-C (190–160)	5.4	5.4	4.7	8.6	3.7	7.7	5.7	6.3	6.4	6.3
O-Aryl-C (160–145)	5.1	4.6	4.4	4.7	2.8	5.8	4.9	3.4	4.7	4.0
Aryl-C (145–110)	10.1	10.6	10.0	11.7	7.4	13.5	9.6	10.7	9.3	11.0
O-Alkyl-C (110–60)	59.0	42.5	57.5	42.7	60.0	43.8	57.9	42.0	59.3	46.5
CH3O/CN (60–45)	7.1	12.8	7.3	11.5	10.6	12.4	7.9	12.1	7.4	12.7
Alkyl-C (45-0)	13.2	23.9	16.0	20.9	15.5	16.9	14.1	25.5	13.0	19.5
HB	0.5	0.8	0.5	0.8	0.4	0.7	0.5	0.8	0.4	0.7
A/OA	0.2	0.6	0.3	0.5	0.3	0.4	0.2	0.6	0.2	0.4
LR	1.4	2.8	1.6	2.5	3.7	2.2	1.6	3.5	1.6	3.2

$$HB = \frac{\Sigma[(0-45\text{ ppm})+(45-60)/2+(110-160)]}{\Sigma[(45-60)/2+(60-110)+(160-190)]; A/OA = (0-45)/(60-110); LR = (45-60)/(110-140).$$

di-O-alkyl anomeric carbon 1 placed at 104/105 ppm (SM). The second more abundant organic components were represented by the aliphatic compounds pertaining to the 0–45 ppm region (Table 6). This spectral interval includes the apolar C functions assigned to the alkyl chains of various lipid compounds, such as fatty acids, waxes, and components of plant bio-polyesters (e.g. cutin and suberin). The bands around 30 and 33 ppm, are related to bulk methylene segments of straight chains, while the shoulder around 21 ppm and at 38–40 ppm, indicate, respectively, the presence of methyl substituents in alkyl chains, hemicelluloses and glucosidic group of flavonoids, and the inclusion of tertiary (C–H) and quaternary (C–R) carbons in assembled rings of sterol and terpene derivatives (SM). The resonance centered around 56/57 ppm may combine either the methoxyl substituent on the aromatic rings of guaiacyl and syringyl units in lignin components, as well as the C–N bonds in peptidic moieties [38]. The resonances shown along the aryl-C region (110–140 ppm) originate from both un-substituted and C-substituted phenyl carbon of lignin fragments as well as to condensed aromatic compounds. The subsequent interval (140–160 ppm) highlighted the presence of O-substituted ring carbons in phenolic compounds, such as the nuclei in position 3, 4, and 5 of lignin monomers in coumaryl, guaiacyl and syringyl structures. Finally, the peaks ranging between 172 and 175 ppm of NMR spectra indicated the inclusion of carbonyl groups pertaining to various compounds such as aliphatic acids, amino acid moieties, side groups of pectin derivatives etc. The shared lower values found for the HB, A/OA structural indexes in starting mixtures (Table 6), are typical features of fresh biomasses thereby confirming the prevalence of degradable polar components in the initial organic materials. The lower value of Lignin ratio found in initial biomasses (Table 6) indicated the significant contribution of polyphenolic derivatives in the signal area in of O-aryl function in the 145–160 chemical shift combined with the presence of lignin units. The larger value of LR found for sample #3 mainly including olive mill waste, may be associated with a larger N content of the corresponding initial residues, which included the hydrolyzed collagen, and the relative increase of C–N functional groups in the 45–60 ppm regions. The main variation found at final composting process was related to the expected decrease of most bioavailable components represented by carbohydrates and polysaccharides (SM); the various biomasses showed a marked decrease O-alkyl-C functions which relative amounts ranged from the 42–46.5 % of total area (Table 6). The decrease or disappearance of the shoulders associated to the O-alkyl-C at 82–88 ppm in mature composts (SM) confirm the progressive dissolution of β 1→4 glycosidic linkage in the decomposition of plant polysaccharides. The increase found for HB and A/OA indexes summarized the selective preservation in final mature composts of stable apolar aliphatic and aromatic organic compounds (Table 6). The final composts #1, #2, #4 and #5 were mainly characterized by a relative increase of alkyl-C molecules, while compost #3 showed the larger improvement of aromatic groups and phenolic signals, also highlighted by the lower value of LR parameter (Table 6). Conversely, the marked increase of Lignin ratio revealed by the samples #1, #2, #4 and #5 suggest the depolymerization of polyphenolic structures and lignin biopolymer and the incorporation of low weight molecules. The relative increase of the signal associated to methoxyl substituents in lignin units at 56/57 ppm (SM) may be also related to the preservation in final compost of nitrogen derivatives thereby confirming data of elemental analysis of the final composts (Table 4). The larger final values of LR in compost #4 and #5 mainly including tomato and pepper crop residues, respectively, may also be related with the incorporation of nitrogen derivatives (e.g. peptides) in final matrices.

3.5. Thermochemolysis GC–MS analyses

The thermochemolysis applied to initial mixtures and final bulk composts released about hundred recognizable different molecules (SM), which were identified as methyl esters and ethers of natural compounds (SM). Most of the identified monomers was represented by lignin components, fatty acids, aliphatic biopolymers, alcohols, and cyclic components (Table 7, SM). As compared with the results of NMR analyses, significant lower quantity of carbohydrates was found among the pyrolysis products of organic materials. This finding has been related to the reduced efficiency of off-line pyrolysis techniques to detect carbohydrate units of polysaccharides in complex matrices [37]. The specific monomers of lignin units have been determined by the main fragmentation pattern and were associated to the current symbols used to distinguish the different structural units: P, p-hydroxyphenyl; G, guaiacyl (3-methoxy, 4-hydroxyphenyl); S, syringyl (3,5-dimethoxy, 4-hydroxyphenyl). The increasing relative abundance of lignin derivatives found in thermochemolysis products of final composts in respect to initial fresh mixtures, indicate the selective preservation of aromatic moieties in the humification step of composting process (Table 7). Among the identified lignin fragments, the oxidized products of both di- and tri-methoxy phenylpropane molecules represented by the aldehydic (G4, S4), ketonic (G5, S5) and benzoic-acid (G6, S6) forms are related to microbial processed materials. Conversely, the concomitant release from the thermochemolysis of 1-(3,

Table 7

Yield (mg g^{-1}) of alkyl (lipid + biopolyesters) and lignin derivatives and lignin oxydation indexa found in thermochemolysis of initial mixtures (t_0) and final compost samples (t_{fin}). Data are means of triplicates with coefficient variation ≤ 16 % (mean value).

COMPOST	#1		#2		#3		#4		#5	
	t_0	t_{fin}	t_0	t_{fin}	t_0	t_{fin}	t_0	t_{fin}	t_0	t_{fin}
Alkyl	70.8	88.6	90.6	87.8	84.4	99.6	89.3	94.7	73.4	95.6
Lignin	27.2	33.8	27.2	41.3	18.5	68.3	30.6	31.4	26.3	44.1
GAd/Al	2.2	3.5	1.5	3.4	1.1	4.1	2.0	4.1	1.8	5.2
S Ad/Al	3.7	4.1	1.5	3.6	1.4	3.9	1.8	4.4	2.1	4.2
Γ	1.0	2.9	0.9	3.2	1.5	3.1	1.3	2.8	1.1	3.6
$\Gamma\Sigma$	1.4	3.0	1.2	5.2	1.6	3.4	1.1	3.4	1.4	4.0

a Ad/Al = G6/G4, S6/S4; GG = G6/(G14 + G15); GS=S6/(S14 + S15).

4-dimethoxyphenyl)-1(3)-methoxy-propene (G10/11, G13) and 1-(3,4,5-trimethoxyphenyl)-1(3)-methoxy-propene (S10/11, S13), as either cis or trans isomers (SM), may be related to the incorporation of less decomposed lignin debris. Moreover, the identification of the enantiomers of 1-(3,4-di-methoxyphenyl)-1,2,3-trimethoxypropane (G14 and G15) and 1-(3,4,5-trimethoxyphenyl)-1,2,3-trimethoxypropane (S14 and S15), confirmed the presence of integral lignified plant components [39]. These lignin molecules allow the evaluation of structural parameters associated to either microbial processed organic materials or to undecomposed plant debris, used to estimate the extent of decomposition of lignin biopolymers during the composting process. In fact, as previously noted, while the aldehydic (G4, S4) and acidic forms (G6, S6) of lignin structures result from the progressive oxidation processes, the corresponding homologues with the integral hydroxylated side chain (G14/15, S14/15) are indicative of unaltered lignin components, which retain the propyl ether intermolecular linkages (Fig. 3). Therefore, the ratio of acidic structures over that of, both, the corresponding aldehydes ($Ad/AlG = G6/G4$, $Ad/AlS = S6/S4$) and over the sum of peak areas for the threo/erythro isomers ($\Gamma G = G6/[G14 + G15]$; $\Gamma S = S6/[S14 + S15]$), are considered to be reliable indicators of the oxidative transformation of lignin polymers into lower molecular weight components [37,39]. The data of decaying index (Table 7) suggested a progressive lignin oxidation in mature composts as compared to slightly decomposed lignin fragments in initial mixtures thereby also supporting the larger release of lignin monomers found in the thermochemolysis products of final composts (Table 7). Large amounts of aliphatic components were found in the pyrograms of both initial mixtures and final composts, revealing the contribution of long linear and cyclic alkyl molecules (Table 7). The methyl ester of alkanolic acids, mostly represented by the ubiquitous hexadecanoic and octadecanoic saturated and unsaturated homologues, also included a marked relative presence of heavier molecules largely dominated by even chain length components (C16–C28) thus suggesting the plant waxes as prevalent source of the straight chain aliphatic acids (SM). The off-line pyrolysis also produced a notable yield of the methylated form of ω -hydroxy alkanolic acids and alkane-dioic acids as minor fraction (SM), which are the main constituents of the external protective barriers of fresh and lignified plant tissues, namely cutin and suberin [39]. A direct input from microbial activity was revealed in the final compost samples by the detection of branched chain of fatty acid methyl esters (FAME). Among these, the most abundant compounds were the 12- and 13-methyl tetradecanoic and hexadecanoic acids (iso and anteiso C15 and C17 FAME (SM), which are common microbial constituents of natural organic matter. Within the lipid components were also found noticeable amounts of C24–C28 aliphatic alcohols, as well cyclic compounds including sterols of both tetra and pentacyclic triterpene origins and diterpene derivatives mainly represented by the so-called resin acids which were shown mainly in the pyrogram of compost #2 mostly including conifer pruning. The abietic, pimaric, and isopimaric acids were the main original precursors found among the diterpenoid molecules, followed by diagenetic products in final compost, such as dehydroabietic acid and labdane acid derivatives. The distribution of aliphatic compounds in initial and final samples revealed the typical decrease of bio-available free lipid fractions, mainly composed by alcohol, fatty acids and the selective preservation of more recalcitrant bio-polyesters components made up by long chain hydroxy and dicarboxylic acids, with a significant preservation of tetra and pentacyclic triterpene derivatives.

3.6. In planta compost suppressiveness assay against tomato tracheofusariosis

In planta compost suppressiveness assay showed the comparative effect of the composted matrices on the progression of the symptoms on tomato due to FOL infections. A significant reduction of disease severity (–30 %) was observed on plants put in touch with compost #1, #3 and #4, compared to infected control (FOL); all the other treatments showed the disease severity around 70 % (Table 8). No significant changes of disease incidence were observed among compost treatments as respect to control (FOL) (Table 8). Plant mortality (Table 8) was also assessed to investigate the effect of compost in helping plants to overcome the biotic stress or

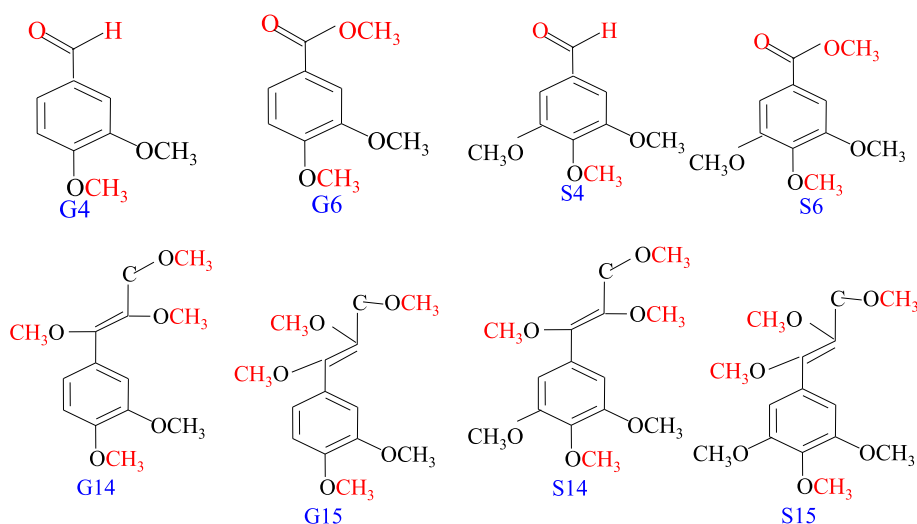


Fig. 3. Lignin units used to determine the structural index.

Table 8

Biometric evaluation, mortality, disease incidence and disease severity assessments on tomato plants at the end of the in planta compost suppressiveness assay, carried out against *Fusarium oxysporum* f.sp. *lycopersici* (FOL). Values \pm standard deviation with lowercase lettering indicates significant differences ($p < 0.05$), according to ANOVA and Bonferroni correction test for multiple comparisons.

	fresh weight, mg		dry weight, mg		length, cm		number of leaves	mortality, %	disease incidence, %	disease severity, %	
	root	stem	root	stem	root	stem					
Healthy control	100 \pm 40	a 643 \pm 183	a 19 \pm 10	a 15 \pm 6	ab 5.1 \pm 1.8	a 7.1 \pm 1.8	a 5.1 \pm 0.5	a 0.0 \pm 0.0	c 0.0 \pm 0.0	c 0.0 \pm 0.0	c
FOL-infected	109 \pm 94	a 209 \pm 156	d 5 \pm 5	b 6 \pm 5	c 3.0 \pm 3.1	b 1.9 \pm 1.6	c 2.5 \pm 1.5	c 33.3 \pm 37.6	b 100.0 \pm 0.0	a 78.3 \pm 13.7	a
Compost #1	105 \pm 48	a 291 \pm 148	cd 6 \pm 4	b 9 \pm 3	bc 5.0 \pm 2.7	a 3.3 \pm 1.4	b 3.5 \pm 0.87	b 8.3 \pm 15.1	c 100.0 \pm 0.0	a 50.5 \pm 13.2	b
Compost #2	105 \pm 63	a 482 \pm 144	b 7 \pm 4	b 12 \pm 8	ab 4.9 \pm 3.6	a 3.1 \pm 2.1	b 2.9 \pm 1.4	bc 16.7 \pm 30.2	c 83.3 \pm 30.2	a 49.4 \pm 26.3	b
Compost #3	125 \pm 53	a 374 \pm 278	bc 9 \pm 9	b 11 \pm 11	bc 4.2 \pm 4.8	ab 2.6 \pm 2.8	bc 2.4 \pm 1.3	cd 11.1 \pm 29.6	c 91.7 \pm 15.1	a 72.2 \pm 19.0	a
Compost #4	131 \pm 68	a 431 \pm 163	bc 9 \pm 5	b 18 \pm 9	ab 4.7 \pm 2.5	a 3.0 \pm 1.4	b 3.2 \pm 1.3	bc 13.9 \pm 30.0	c 97.2 \pm 9.6	a 47.2 \pm 23.2	b
Compost #5	32 \pm 38	b 119 \pm 77	d 7 \pm 9	b 6 \pm 5	c 1.2 \pm 2.0	c 1.5 \pm 2.1	c 1.6 \pm 0.7	d 58.3 \pm 20.7	a 100.0 \pm 0.0	a 83.9 \pm 10.4	a

phytotoxicity: it resulted that only 10 % of plants, primed with compost #1, #2, #3 and #4, and around 35 % of FOL-infected plants, were heavily damaged; on the other hand, the highest mortality was observed on infected plants put in contact with compost #5 (around 60 %), confirming a phytotoxic impact of this compost primarily including pepper crop residues; indeed, compost #5 showed severe phytotoxicity symptoms since the beginning and just after transplantation (data and images not showed), even before FOL infection could manifest any symptoms. Confirming the phyto-pathological assessment, biometric parameters (Table 8) highlighted a negative impact of compost #5 on plant growth which cannot be attributed to the FOL infection or inability in potential biocontrol but to other possible causes. Even the GI test confirmed such a phytotoxicity only found on compost #5. On the other hand, a positive effect on biometric parameters was highlighted on stem fresh weight for compost #2, #3, #4, and stem dry weight for compost #2 and #4, all resulting significantly higher than FOL and comparable to healthy control. Root length of plants treated with compost #1, #2 and #4 resulted significantly improved compared to FOL-infected plants.

4. Discussion

The use of renewable organic substrates such as compost as effective substitute of peat is a current challenge in the protected cultivation systems. A possible limitation is related to the variable or unsteady characteristics of recycled biomasses in contrast with the almost uniform properties of geochemical materials. However, the careful check of composting process coupled with the detailed molecular and biological appraisal of compost obtained from suitable organic biomasses, may be a straightforward tool to delineate a technological optimal solution for specific needs and to conceive a possible tailoring of composting processes suited for dedicated cropping systems. As compared to peat materials, besides the basic physical and chemical requirements of growing substrate, the humified compost may provide additional beneficial bioactive components able to either counteract biotic and abiotic stresses and/or act as bio-stimulant for root development and crop growth [37,38]. An important target in the evaluation of bioactivities of natural organic compounds is the identification of structural-activity relationship. In the present paper all tested composts, except compost #5 including mainly pepper plant residues, showed shared positive response on both diseases control and plant development parameters. The molecular characterization by both NMR and thermochemolysis GC-MS revealed a noticeable preservation of aliphatic and aromatic hydrophobic components in final biomasses. Moreover, the structural index related to lignin derivatives outlined the incorporation of microbial processed molecules thus suggesting the prevalence of low molecular weight bioavailable phenolic compounds. Although no univocal or unambiguous mechanism has been so far clearly elucidated, previous studies have stressed the occurrence of combined effects of compost and compost-derived component in the triggering of bioactive properties. A common feature of tested materials is represented by the progressive increase, during composting and humification stage, of apolar or hydrophobic fraction and the decomposition and depolymerization of polar bio-decomposable compounds like rigid ligno-cellulose fibers [37,57]. The final simultaneous and almost fair distribution of contiguous depolymerized hydrophilic and hydrophobic domains is in fact, considered a viable combination to ensure the formation of pliable micelle like structure able to both act as carrier of bioactive compounds and provide a suitable interaction with the cell membranes of plant roots systems [39,58]. The proposed structural-activity interactions rely on either indirect/mediated bioactive activities or direct release of biostimulants [58,59]; with respect to mediated actions, the adhesion of organic micelle-like conformation, dissolved or suspended in rhizosphere microenvironments, on root membranes may promote a mild-stress effect on root systems. The plant is hence induced to activate a physiological tropism-like response based on integrated enzymatic and biochemical pathways marked by specific intermediates (e.g. reactive oxygen species, ROS, nitrous oxide, NO, abscisic acid, ABA). This signaling train and condition cells and tissues into an alert or warning stage thus favoring a potential prompt defense against subsequent actual biotic or abiotic stresses. In the hypothesized direct structural-activity relationship, the flexible hydrophobic-hydrophilic arrangements upon interaction with the roots micro-environments (e.g. roots exudates, siderophore, ionic strength, etc.) may alter their conformational structure providing the release of small bioactive compounds retained in the hydrophobic core of micelle-like structure. Different molecules, including phenolic units from lignin and peptidic moieties, originated from the decomposition humification step of composting process, have been shown an effective hormone-like activity able to either promote the crop development or to modulate the plant reactivity against biotic harmful components [6,37,59].

The bioactive properties of compost may allow additional technological diversification of beneficial use as plant growth promoters. In fact, the bulk humified compost may undergo to easy green extraction procedures, based on water solutions, to isolate the most bioactive fractions represented by humic substances and compost tea. These organic water-suspensions have shown to act as either effective plant biostimulant or ecofriendly agrochemical to counteract the incidence of biotic or abiotic stresses [60,61]. The preliminary isolation of bioactive fractions did not affect the properties of compost to act as growing substrate and soil amendment. Furthermore, both bulk compost biomasses and bioactive fractions may be effectively combined with selected beneficial microorganisms, depending on molecular features of compost materials and the required agronomic application. The combination of organic substrates and defined microorganisms has been proved to widen either the availability and crop uptake of nutrients, increase the root and shoot development and to effectively improve the crop resilience against pathogens and environmental constraints [39].

Plant disease suppressiveness is a remarkable biological property of composts. In this study, compost #1, #2 and #4 managed to significantly reduce severity of tracheofusariosis symptoms on susceptible tomato, leading to composting pipelines as functional compost production models. Compost #1 and #2 obtained from pig slurry mixed with other animal manures, and #4, originated from tomato crop residues and woody materials, showed the potential to reduce the effects of pathogen infection and preserve growth performances of tomato. The use of compost able to suppress plant diseases is considered a new promising agro-ecological tool for a more environmentally sustainable control of plant pathogens in agriculture, both in nursery and field crops. The search for the mechanistic basis of this property, representing a true ecosystem service, aims at standardizing production of functional composts with high value-added. Biotic and abiotic components of the composts dynamically contribute to suppression of pathogenesis, as well

described feedback model [62] where the antagonistic microbial complex and the organic matter components play a pivotal role interfering in pathogenesis directly against the pathogen population [63], or indirectly by interacting with the plants. Evidence from the current literature on the three-way compost-*F. oxysporum*-plant complex interaction shows just that the ability of the organic amendment to reduce tracheofusariosis can follow a dual pattern. While direct action of a suppressive compost can undoubtedly lead to a reduction in the population levels of the fungus in the soil, in our case, with the infection in progress, started via root openings and continued in the vessel, a plant-mediated action would be more effective. Due to the endophytic course of FOL infection compost should exert a kind of priming effects on tomato plants to suppress developing symptoms. As matter of the fact, Yogev et al. [64], by using a split-root system, clearly demonstrated additional induced resistance mechanisms involved in the suppression of melon Fusarium wilt by composted tomato-plant residues mixed to cattle manure fraction. On the other hand, Serra-Wittling et al. [65], by using the system *F. oxysporum* f. sp. *lini-flax*, discussed for the first time the preponderance of compost-induced nutrient and space competition mechanisms over the biotic component eliminated by heating. Outcomes of the interaction of suppressive composts with plant/fusarium pathosystems have been observed in some previous investigations which converge on the stimulative role declined by the quality of organic matter. Saadi et al. [66] found significant relation of dissolved organic matter as maturity indicator with the suppressive capacity of composted tomato plants and separated cow manure against *F. oxysporum* f. sp. *melonis* on melon. While shorter availability of the labile carbon sources is characteristic of the composition of grape marc compost and cork compost found to be suppressive towards FOL [67], and FOL and *F. oxysporum* f. sp. *dianthi* [68], respectively. Overall evaluation of compost parameters indicates no clear correlation with suppressive functions. However, in agreement with findings of Borrero et al. and Castaño et al. [67, 68], the suppressive composts were also higher in the hydrophobicity index arising from imbalance in the C distribution between recalcitrant and polysaccharidic C. The chemical-physical characterization of the five experimental compost here studied clearly show how any type of biodegradable organic waste can be effectively treated through optimized composting, giving rise to products with satisfactory final agronomic features; however, it emerges that some ingredients, in particular animal manures, while giving the compost a good supply of macro elements of plant nutrition (especially N and K), can nevertheless bring excessive levels of some microelements (particularly Zn and Cu). These ingredients must therefore be used wisely and possibly in small dosages. Furthermore, the highly odorous nature of animal waste must be considered, with strong emissions that are released especially in the pre-composting storage phases, which can further hinder its use in farm composting. On the other hand, compost based just on vegetable wastes, as the cases of compost #4 and #5 most including residues from tomato and pepper crops, have the advantage of their easier availability associated with a lower production of smelling emissions. Furthermore, both tomato and pepper crop waste transfer a rich supply of N and K to the final compost. This study also highlighted that compost made from pepper waste resulted highly phytotoxic, as assessed both in germination and suppressiveness tests; this is reasonably attributable to the alkaline pH (9.25) which, associated with a high N content (3.1 %), may have determined the release of ammonia in gaseous form, notoriously phytotoxic, for the shift in the chemical equilibrium affecting the NH_4^+ present in the water solution. It should be also underlined that compost #5 underwent shorter maturation times than the others investigated.

5. Conclusions

The study demonstrated how composting of any organic waste can be optimized through the correct design of the initial mixture and the careful control of oxygen content during the process, resulting in a 50–60 % yield of end-product. This approach effectively minimized undesirable emissions from fermentations and reduced ammonia loss. During composting cumulative oxygen consumption ranged from 282 to 456 $\text{gO}_2\text{kg}_{\text{DM}}^{-1}$, with peaks of 2.55 $\text{gO}_2(\text{kg}_{\text{VSh}})^{-1}$, and carbon dioxide emissions resulted up to 22–36 % $\text{C}_{\text{INITIAL}}$, with peaks of 5.89 $\text{gCO}_2(\text{kg}_{\text{VSh}})^{-1}$. The process yielded stable and mature compost faster, while preserving the essential plant nutrients from the original organic materials by preventing leaching. The study also identified compost with promising suppressive effects against tomato tracheofusariosis, particularly that derived from tomato cultivation residues which exhibited high microbial biodiversity, based on preliminary evaluations not reported here. The findings suggest best practices for an appropriate farm composting of such agricultural waste, enabling effective recycling back into tomato cultivation, and potentially reducing the need for fertilizers and pesticides.

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Data availability

Data associated with this study have not been deposited into a publicly available repository. Data from this study is included in article/supplementary material/referenced in article.

Publishing ethics

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

Authors have not used generative AI and AI-assisted tools in scientific writing nor in making figures, images, and artwork.

CRedit authorship contribution statement

Roberto Altieri: Writing – review & editing, Writing – original draft, Supervision, Resources, Methodology, Investigation, Funding acquisition, Formal analysis, Data curation, Conceptualization. **Riccardo Spaccini:** Writing – original draft, Investigation, Formal analysis, Data curation. **Catello Pane:** Writing – original draft, Methodology, Investigation, Formal analysis, Data curation. **Gelsomina Manganiello:** Writing – original draft, Methodology, Investigation, Formal analysis, Data curation. **Silvana Cangemi:** Investigation, Data curation. **Mariavittoria Verrillo:** Investigation. **Vitale Stanzione:** Investigation. **Alessandro Esposito:** Writing – original draft, Investigation, Formal analysis.

Declaration of competing interest

The authors declare the following financial interests/personal relationships which may be considered as potential competing interests: Roberto Altieri reports financial support was provided by European Union's PRIMA programme. Gelsomina Manganiello reports financial support was provided by Ministero dell'Università e della Ricerca, Italy. If there are other authors, they declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Appendix A. Supplementary data

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