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Mitigation of phytotoxic effect of compost by application of optimized aqueous extraction protocols

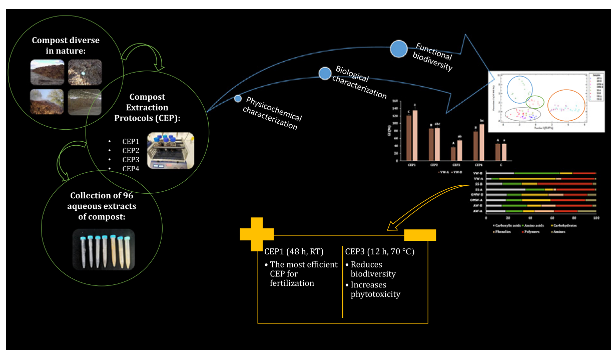
R. Lerma-Moliz, J.A. López-González *, F. Suárez-Estrella, M.R. Martínez-Gallardo, M.M. Jurado, M.J. Estrella-González, A.J. Toribio, R. Jiménez, M.J. López

Unit of Microbiology, Department of Biology and Geology, CITE II-B, Agrifood Campus of International Excellence ceiA3, CIAIMBITAL, University of Almería, 04120 Almería, Spain

HIGHLIGHTS

- Compost extracts improve the biofertilizing characteristics of the compost.
- Temperature and incubation periods mainly determine the quality of the extracts.
- The phytotoxic effect of composts is reduced by the use of compost extracts.
- Moderate temperatures guarantee the preservation of microbial functionality.

GRAPHICAL ABSTRACT



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ABSTRACT

The abuse of chemical fertilizers in recent decades has led the promotion of less harmful alternatives, such as compost or aqueous extracts obtained from it. Therefore, it is essential to develop liquid biofertilizers, which in addition of being stable and useful for fertigation and foliar application in intensive agriculture had a remarkable phytostimulant extracts. For this purpose, a collection of aqueous extracts was obtained by applying four different Compost Extraction Protocols (CEP1, CEP2, CEP3, CEP4) in terms of incubation time, temperature and agitation of compost samples from agri-food waste, olive mill waste, sewage sludge and vegetable waste. Subsequently, a physicochemical characterization of the obtained set was performed in which pH, electrical conductivity and Total Organic Carbon (TOC) were measured. In addition, a biological characterization was also carried out by calculating the Germination Index (GI) and determining the Biological Oxygen Demand (BOD_5). Furthermore, functional diversity was studied using the Biolog EcoPlates technique. The results obtained confirmed the great heterogeneity of the selected raw materials. However, it was observed that the less aggressive treatments in terms of temperature and incubation time, such as CEP1 (48 h, room temperature (RT)) or CEP4 (14 days, RT), provided aqueous compost extracts with better phytostimulant characteristics than the starting composts. It was even possible to find a compost extraction protocol that maximize the beneficial effects of compost. This was the case of CEP1, which improved the GI and reduced the phytotoxicity in most of the raw materials analyzed. Therefore, the use of this type of liquid organic amendment could mitigate the phytotoxic effect of several composts being a good alternative to the use of chemical fertilizers.

1. Introduction

The forecast that in 50 years the world population will increase to 10 billion inhabitants poses a serious supply problem (Van Dijk et al., 2021).

* Corresponding author.
E-mail address: lgj132@ual.es (J.A. López-González).

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In recent decades, the development of intensive agriculture has made it possible to increase global resource production. One of the main advances has been the use of chemical fertilizers (Rahman and Zhang, 2018). However, the abuse of this type of products is causing serious problems affecting the environment, the economy, and even human health (Asadu et al., 2020; Blanco-Vargas et al., 2020). The excess fertilization that the plant cannot assimilate and the xenobiotic nature of these compounds makes their elimination difficult, leading to contamination in soils and waters, that can even reach the human food chain (Kapeleka et al., 2020). Therefore, the competent authorities in different parts of the world are encouraging the use of other organic alternatives that cause less damage to the ecosystem (Tsai, 2020; Guo et al., 2021). Among these effective and less harmful options are the use of compost and its derivatives (Bekchanov and Mirzabaev, 2018).

In particular, compost, a humified product resulting from the biodegradation of organic waste in the composting process (Libutti et al., 2020), has been studied by numerous authors for its beneficial properties for soil and plants (Eldridge et al., 2018; Kranz et al., 2020; De Corato, 2020; Moreno et al., 2021). Furthermore, thanks to the sanitization that occurs during the process, this organic amendment is free of pathogens and rich in nutrients (López-González et al., 2021). Thus, in terms of circular economy, the use of compost is an ideal alternative to the use of agrochemicals, allowing the practice of a more sustainable and environmentally friendly agriculture (Pecorini et al., 2020). All of the above is reflected in the commitment of major world economies to the implementation of composting as the main sustainable alternative for organic solid waste management (Razza et al., 2018).

Even with the advantages derived from the use of compost, it is relatively common to find a certain reluctance on the part of farmers to use it (Ayilara et al., 2020). One of the disadvantages of compost is that it cannot be used by fertigation. This drawback can be solved by obtaining aqueous extracts from composted materials (Marín et al., 2013). These extracts are the result of mixing compost and water in different proportions during an extraction conditions (González-Hernández et al., 2021). The resulting liquid organic amendment is composed of bioactive molecules of different nature and microorganisms beneficial to plants and soil. However, the microbial richness of the extracts is determined by the characteristics of the original compost and the incubation conditions (Eudoxie and Martin, 2019). They also provide the plant with better resistance to pathogens and diseases (Zouari et al., 2020). This is because the extracts increase the amount of nutrients in the plant, activate the plant defense responses and even enhance the growth of soil microorganisms that act as biocontrol agents (Li et al., 2020). Therefore, the use of aqueous extracts is a simple, cheap and effective alternative to chemical pesticides.

The application of aqueous compost extracts capable of promoting plant growth means less dependence on inorganic fertilizers (Hargreaves et al., 2009), which is particularly advantageous in intensive agriculture. In addition, the negative effects of the abusive use of agrochemicals are avoided. There are previous studies on the biofertilizing effect of the aqueous extract but, always, obtained under standardized and fixed conditions (Naidu et al., 2013; Otero et al., 2019). However, in the present study, the compost was subjected to different extraction protocols ranging from mild to more severe incubation conditions. On the other hand, the use of different raw materials makes it possible to establish a diverse spectrum of applicability depending on the fertilizing properties of the extract and the starting raw material. Furthermore, for a good characterization, it is important to determine if there are differences with respect to the solid compost from which they were obtained. Therefore, based on the above, the aim of this study was to analyze and determine the phytostimulant capacity of aqueous extracts obtained by different protocols in comparison with the starting compost. For this purpose, the following challenges were determined: i) obtaining a collection of aqueous compost extracts from different types of industrial compost; ii) characterizing the aqueous extracts obtained from a physical, chemical and biological point of view; iii) analyzing the phytotoxic character of the extracts; iv) determining the microbial functional diversity of the extracts; v) performing a statistical correlation of all the parameters studied, in order to establish a pattern of the agronomic behavior of the aqueous compost extracts.

2. Materials and methods

2.1. Raw materials and obtaining aqueous extracts

This work was carried out on compost samples from industrial composting process of Agri-food Waste (AW-A, AW-B), Olive Mill Waste (OMW-A, OMW—B), Sewage Sludge (SS-A, SS-B) and Vegetable Waste (VW-A, VW-B). All samples came from eight industrial facilities that process organic waste generated in southwestern Europe, specifically in Spain. Two facilities were selected for each type of waste analyzed. The final material generated in the composting processes of each facility was used to obtain the aqueous extracts. The characterization of the starting composts is shown in Table 1. Aqueous extracts were obtained by applying four different Compost Extraction Protocols (CEP) in terms of time and temperature. A 1:5 dry weight suspension in sterile distilled water was made. In extraction protocol 1 (CEP1), the mixture was kept at room temperature for 48 h. In the case of extraction protocol 2 (CEP2), the incubation time was 24 h at 40 °C. The incubation conditions for extraction protocol 3 (CEP3), were 70 °C and 12 h. Finally, the samples extracted with protocol 4 (CEP4) were incubated for 14 days in darkness at room temperature. All samples were incubated under agitation at 200 rpm, except those extracted with protocol CEP4. Once all incubation periods were completed, the samples were centrifuged for 20 min at 4000 rpm. After this, each sample was filtered first through a filter paper and then through a fiberglass filter. Finally, the extracts obtained were aliquoted and frozen at −20 °C until analysis. A total collection of 96 aqueous compost extracts was obtained.

2.2. Physicochemical and biological characterization of the starting compost and the collection of extracts obtained

From the aqueous extracts obtained, pH, Electrical Conductivity (EC), Total Organic Carbon (TOC) and Biological Oxygen Demand (BOD_5) were analyzed. In addition, the Bulk Density (BD) of the starting compost was determined. The pH and EC of the extracts were measured using the Crison BASIC 30 (Crison Instruments, S.A., Barcelona, Spain). In the case of compost, a 10^{-1} dilution in distilled water was used to determine these two parameters. To calculate the bulk density of the compost, the raw materials was dried in an oven at 60 °C for 24 h. The difference of the weights before and after drying was used to calculate the BD with the expression (1). To obtain TOC and BOD_5 data, a 10^{-2} dilution of the extracts was done. In order to establish the TOC values, a TOC-VCSN analyzer (Shimadzu Co., Kyoto, Japan) was used. In the case of BOD_5, the different measurements were performed with a BOD analysis sensor (BOD Sensor, Velp Scientifica, Lombardia, Italy) following commercial indicators. Sterile distilled water was used as a control. BOD_5 was calculated using the expression (2).

$$BD = \text{dry sample weight}/\text{cylinder volume}. \tag{1}$$

Table 1
Physicochemical characteristics of the starting compost.

	AW-A	AW-B	OMW-A	OMW-B	SS-A	SS-B	VR-A	VR-B
pH	6.84	8.47	7.11	9.35	7.52	8.36	9.26	9.82
EC (mS cm^{-1})	7.43	2.21	4.68	3.66	4.65	2.28	8.84	9.71
SOC (%)	0.04	0.16	0.39	0.80	0.33	0.21	0.71	0.42
BD (g cm^{-3})	0.15	0.29	0.53	0.26	0.40	0.44	0.22	0.29

Data are mean values (n = 3), data have SD < 5 %. All these composts comply with current European legislation on microbiological contamination (EU, 2019). All data are on a dry weight basis. Abbreviations: EC: Electrical Conductivity; SOC: Soluble Organic Carbon; BD: Bulk Density; TOC: Total Organic Carbon; AW-A. B: Agri-food Waste; OMW-A. B: Olive Oil Mill Waste; SS-A. B: Sewage Sludge; VW-A. B: Vegetable Waste.

$$BOD_5 = (BOD_5 sample \times dilution\ factor) - BOD_5 control. \quad (2)$$

2.3. Determination of germination index (GI)

The study of the biostimulant capacity of each extract was carried out by following the method described by Zucconi et al. (1985) with some modifications. For this purpose, 100 *Lepidium sativum* seeds were used. After 48 h of incubation at 25 °C in darkness, the germinated seeds and the radicle length were measured. In addition, a germination test was performed on the compost samples to compare the germination results with those obtained from the extracts. The GI was obtained according to the equation below (3):

$$GI\ (\%) = (G_S \times L_S)/(G_W \times L_W) \times 100 \quad (3)$$

G_S: percentage of germinated seeds in the presence of the compost aqueous extracts.

G_W: percentage of germinated seeds in the presence of distilled water.

L_S: is the mean of radicle elongation (mm) in the presence of the compost aqueous extracts.

L_W: is the mean of radicle elongation (mm) in the presence of distilled water.

2.4. Analysis of functional biodiversity

To analyze the functional biodiversity of the samples, the method described by Martínez-Gallardo et al. (2021) was followed, with slight variations. Briefly, a 10^{-3} dilution in distilled water of each extract was done. Thereafter, 150 μL of the diluted extract were added to each well of the Biolog EcoPlates™ microplates (Biolog, USA) and incubated at 25 °C for 72 h. Then, an EON microplate spectrophotometer (Biotek, USA) was used to measure the Optical Density (OD) at 590 nm. A correction of the OD of each well was done with the control sample (water). Finally, based on the 31 substrates included in the Biolog EcoPlates™ system, the following biodiversity indicators were calculated: functional richness (R), sum of the number of cells where OD was greater than or equal to 0.15; functional activity intensity as Average Well Color Development (AWCD); and Substrate Average Well Color Development (SAWCD) for each of substrate categories (carboxylic acids, amino acids, carbohydrates, phenolics, polymers and amines).

2.5. Data processing

All tests were fulfilled in triplicate. Statistical analyses performed included a Fisher's least difference test (LSD) at $P < 0.05$ and an analysis of variance (ANOVA). In addition, Spearman's correlations were carried out to determine possible relationships between all the parameters studied based on each extraction protocol. Discriminant function analysis was performed to confirm the variability of the samples used. These analyses were performed with Statgraphics Centurion XIX version 19.2.01 (Stat-Point, Inc., Virginia, USA).

3. Results and discussion

3.1. Physicochemical characterization

In the present work, different compost extraction protocols were tested. All of them were selected based on previous experiences described by other authors (Oka and Yermiyahu, 2002; Bernal-Vicente et al., 2008; Koné et al., 2010). This was intended to cover a wide range of extraction conditions, in order to obtain conclusions on the agronomic suitability of the different extracts. The results of the physicochemical characterization of the aqueous compost extracts are shown in Table 2. Regarding pH, this parameter revealed the more or less suitable degree of maturity of the starting composts (Lin et al., 2020). This is a fundamental parameter, since values far from neutrality can compromise plant growth. In general terms, values were slightly alkaline in all extracts. These data agreed with those of Azim et al. (2018), who reported that pH values close to 8 were suitable for the growth of microorganisms and crops. No compromising pH levels were detected from any of the applied extraction protocols. With respect to EC, the results varied between the different extracts (3.8–16.13 mS cm^{-1}). Higher EC values were obtained in aqueous extracts compared to solid composts (Table 1). This was also observed in the study of Kim et al. (2015), in which the EC values of the extracts increased after one day of incubation. These data were to be expected since stirring times to obtain extracts increase the presence of salts in solution. The differences in the conductivity of the extracts were mainly ascribed to the nature of the composts.

The highest results were found in the VW extracts, particularly in the extract from CEP1 of VW-B with an EC of 16.13 mS cm^{-1}. These residues came from intensive agriculture. Therefore, the high conductivity shown by VW-A and VW-B could have been caused by the accumulation of inorganic fertilizers when using drip irrigation (Siles-Castellano et al., 2020) or due to the irrigation of plant residues with leaching water during the composting process. This was also observed in the starting composts of vegetables waste (Table 1). In the case of TOC, there was a clear trend in all extracts when CEP3 protocol was applied. The aggressive conditions of this extraction method may have resulted in the extraction of a more concentrated fraction of soluble carbon than the other treatments (Fernández-Delgado et al., 2020).

3.2. Biological analysis

3.2.1. Germination bioassay

The calculated GI values are shown in Fig. 1 (A-D). The values of the extracts were compared with the reference compost from which they were

Table 2
Results of physicochemical characterization of aqueous compost extracts.

		AW-A		AW-B		OMW-A		OMW-B		SS-A		SS-B		VW-A		VW-B	
pH	CEP1	7.61	a	8.12	a	8.19	ab	8.40	a	7.69	a	7.95	a	8.12	a	8.2	a
	CEP2	7.93	b	8.19	ab	8.31	b	8.47	a	7.80	ab	8.05	b	8.22	a	8.3	b
	CEP3	8.35	c	8.44	b	7.96	a	8.57	a	8.01	b	8.34	d	8.43	b	8.66	c
	CEP4	8.36	c	8.18	ab	8.19	ab	8.44	a	7.93	ab	8.26	c	8.61	c	8.63	c
EC (mS cm^{-1})	CEP1	9.1	b	7.97	b	4.73	a	7.73	b	8.33	ab	5.5	b	12.63	a	16.13	b
	CEP2	8.4	b	7.33	a	6.20	b	4.83	a	8.7	b	3.8	a	12.63	a	15.63	b
	CEP3	5.6	a	9.27	d	6.40	b	7.80	b	8.7	b	5.4	b	12.53	a	15.33	b
	CEP4	6.3	a	8.53	c	5.30	ab	8.23	b	7.87	a	5.1	b	13.07	a	12.07	a
TOC (mg L^{-1})	CEP1	5.15	ab	35.68	a	4.63	a	52.84	a	21.93	a	13.06	a	38.80	ab	27.48	ab
	CEP2	5.45	b	38.10	a	4.56	a	79.72	ab	24.02	ab	12.38	a	37.69	a	28.91	b
	CEP3	9.29	c	54.17	b	12.38	b	145.00	d	26.12	b	20.11	b	49.24	b	48.72	c
	CEP4	4.42	a	34.96	a	3.62	a	98.53	c	20.75	a	14.93	ab	31.26	a	17.69	a

Data are mean values ($n = 3$), those with the same letter in the same column are not significantly different from each other (LSD $p < 0.05$). Abbreviations: EC: Electrical Conductivity; TOC: Total Organic Carbon; CEP1: Compost Extraction Protocol 1; CEP2: Compost Extraction Protocol 2; CEP3: Compost Extraction Protocol 3; CEP4: Compost Extraction Protocol 4; AW-A. B: Agri-food Waste; OMW-A. B: Olive Oil Mill Waste; SS-A. B: Sewage Sludge; VW-A. B: Vegetable Waste.

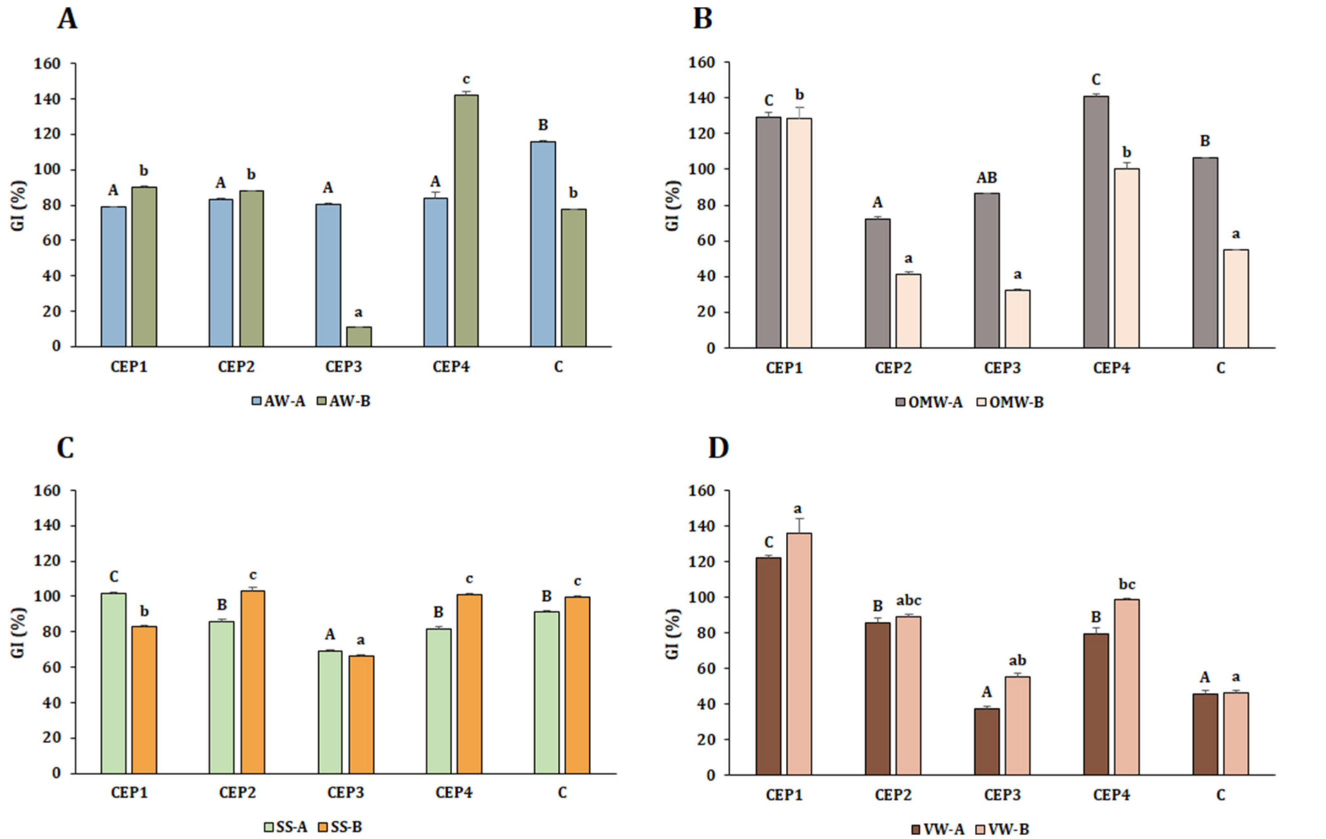

Fig. 1. Germination Index (GI) of the aqueous compost extracts and their solid compost control. Data are mean values ($n = 3$), those with the same letter in the same column are not significantly different from each other (LSD $p < 0.05$). A: Germination Index in percentage (GI%) of Agri-food waste (AW-A. B); B: Germination Index in percentage (GI %) of Olive Oil Mill Waste (OMW-A. B); C: Germination Index in percentage of Sewage Sludge (SS-A. B); D: Germination Index in percentage of Vegetable Waste (VW-A. B).

obtained. Through this bioassay, the quality of the aqueous extracts from each compost was determined as a function of the extraction protocol applied. In general, the application of the different protocols modified the GI of the extracts compared to the composts. In fact, after the application of CEP1 and CEP4, better results were obtained. On the opposite side, the worst results were detected in CEP3. This protocol did not increase the GI of any extract, indeed, it decreased it in some cases. In particular, the AW-B extract gave the worst result, 10.72 %, and turned out to be the one with the lowest GI (Fig. 1A). The explanation for this fact may be supported by the TOC results. The TOC values from the application of the CEP3 protocol were the highest, which may justify the adverse effect on the promotion of seed growth. Based on Bohacz (2019), the extraction of a higher proportion of soluble organic compounds negatively affects the GI of the extracts. However, much more outstanding were the results obtained in other extraction protocols. While compost samples of OMW—B, VW-A and VW-B were phytotoxic, with GI values of 55.01 %, 45.78 % and 46.43 %, respectively (Fig. 1B and Fig. 1D), the application of the CEP1 protocol on these same samples resulted in phytostimulant extracts. In this sense, the OMW-B and VW-A extracts obtained after applying the CEP1 protocol showed a high germination index in watercress seeds (128.52 % and 135.75 %, respectively). Particularly, the extracts with the highest biostimulant capacity were those obtained with CEP4. Specifically, the extracts of AW-B and OMW-A obtained from CEP protocol showed germination rates around 140 % (Fig. 1A and B). This enhancement in plant growth after the application of aqueous compost extracts was also detected by Pant et al. (2012). These extracts provide microorganisms and different compounds beneficial to plants and soil. Based on these results, it is possible that the application of some of the extraction protocols favored the extraction of the phytostimulant compounds present in the solid matrix of the compost, also eliminating the phytotoxic fraction in the extracts. This work has confirmed that in some of the extracts obtained, the phytotoxic effect of compost solid matrices is mitigated, thus increasing the phytostimulant capacity of the raw material.

3.2.2. Biological oxygen demand analysis

The data related to the BOD_5 analyses are shown in Fig. 2 (A-D). The Biological Oxygen Demand (BOD) is a measure of the oxygen consumed by microorganisms to metabolize organic compounds in a sample over 5 days (BOD_5) at 20 °C (Jouanneau et al., 2014). Therefore, a higher BOD indicates a high organic matter content, although this fact does not necessarily imply high plant toxicity. In general, this was corroborated in the CEP4 extracts of AW, OMW and VW, as they showed the highest BOD_5 values, ranging from 60 to 80 g L^{-1} (Fig. 1A-B-D). In particular, the highest value was obtained in the AW-B sample extracted with the CEP4 protocol. On the other hand, the BOD_5 values for most of the extracts obtained with CEP1, CEP2 and CEP3 protocols were lower. These three treatments corresponded to incubation periods of 48, 24 and 12 h, respectively, while it was 14 days for the protocol CEP4. Thus, in view of these results, it was determined that the incubation time is an influential factor on the BOD_5 levels of the extracted samples and thus on the biofertilizing capacity of the extracts. That is, at shorter incubation times, the temperature causes more toxic components to be extracted. In fact, in the study by Kwok et al. (2005) they also detected an increment in oxygen consumption with increasing temperature. This means that with the increase in temperature, more organic compounds were extracted and, therefore, BOD_5 increased. Our results are in agreement with those observed by other authors in compost tea (Carballo et al. (2009).

3.3. Functional biodiversity evaluation

The analysis of the functional biodiversity of the microbiota present in the aqueous compost extracts is shown in Fig. 3 (A-B). This assay was

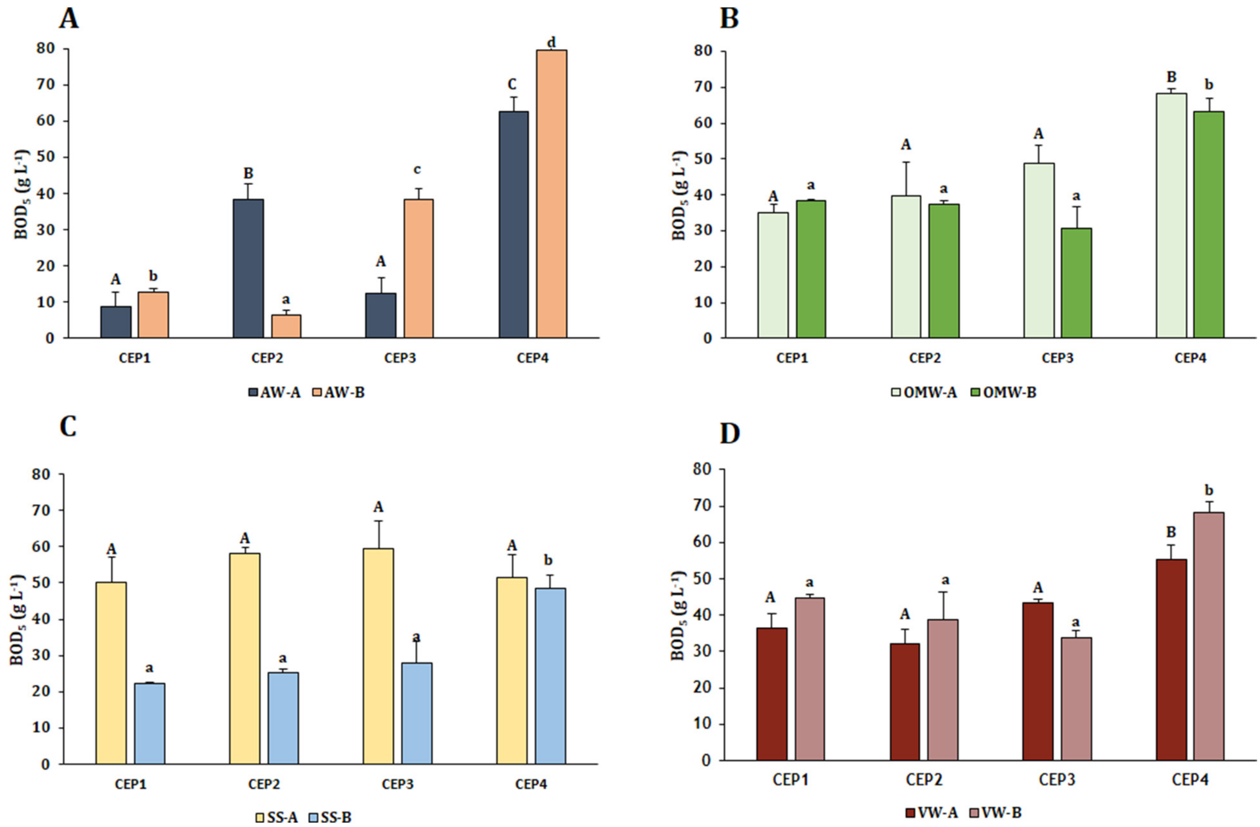

Fig. 2. Biological Oxygen Demand (BOD_5) of the aqueous compost extracts. Data are mean values ($n = 3$). those with the same letter in the same column are not significantly different from each other (LSD. $p < 0.05$). A: Biological Oxygen Demand (BOD_5 g L^{-1}) of Agri-food Waste (AW-A. B); B: Biological Oxygen Demand (BOD_5 g L^{-1}) of Olive Oil Mill Waste (OMW-A. B); C: Biological Oxygen Demand (BOD_5 g L^{-1}) of Sewage Sludge (SS-A. B); D: Biological Oxygen Demand (BOD_5 g L^{-1}) of Vegetable Waste (VW-A. B).

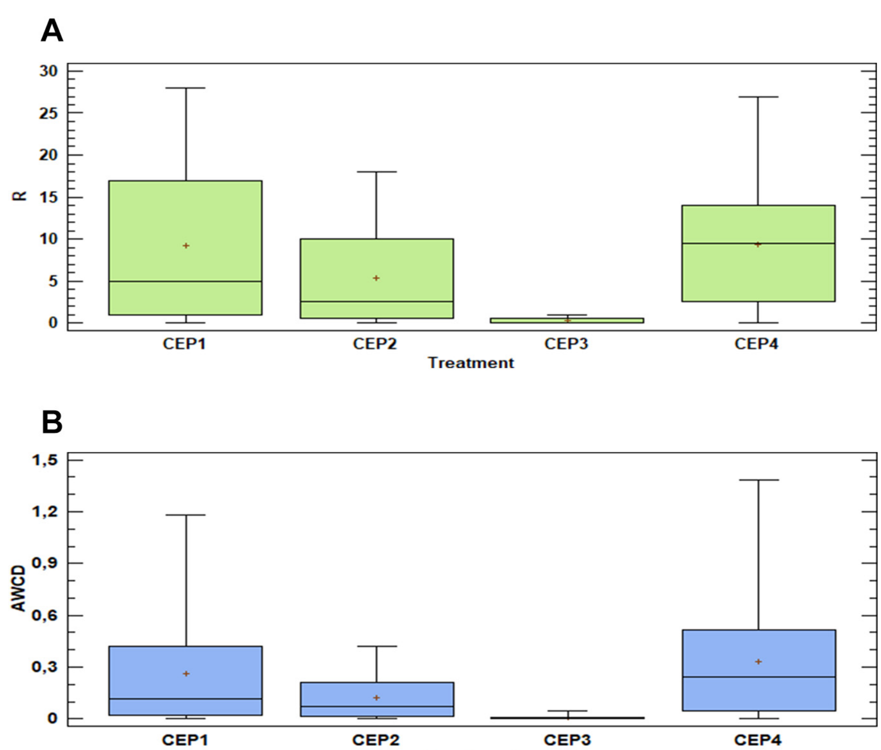

Fig. 3. Results of the functional biodiversity of the microbiota present in the aqueous compost extracts. A: Richness (R); B: Functional Intensity (AWCD).

carried out with the Biolog Ecoplates technique, which studies microbial communities based on their ability to oxidize 31 organic substrates (Sofo and Ricciuti, 2019). The two parameters represented were the functional richness (R), which refers to the number of substrates oxidized by the microorganisms and the intensity of functional activity, AWCD (Martínez-Gallardo et al., 2021). In this work, the extracts obtained by the CEP1 and CEP4 protocols showed the highest functional biodiversity values. Specifically, the extract belonging to OMW-B gave the highest value via CEP1 and CEP4 for both R, 25 and 22.33, and AWCD, 0.97 and 0.94, respectively. As expected, CEP3 showed the worst result for the two parameters analyzed, being zero for most of the measurements taken. This was due to the fact that a lot of organic matter was extracted with this treatment, but the high temperatures applied reduced the microbial load of the extracts (Qiu et al., 2021).

The metabolic diversity of aqueous compost extracts as a function of substrate chemical category are shown in Fig. 4 (A-D). The parameter analyzed was the Substrate Average Well Color Development (SAWCD) for each substrate category (Martínez-Gallardo et al., 2021). According to the results, the major metabolic activities were those related to carboxylic acids, polymers, carbohydrates, and to a lower level, amino acids, since they were present in almost all the extracts. CEP1 and CEP4 were the most balanced treatments in terms of metabolic diversity (Fig. 4A and Fig. 4D). That is, they were the ones that showed practically all the categories of substrates analyzed. Among these two, CEP1 was the protocol with the highest metabolic diversity due to the low presence or absence of amine metabolism-related activities in CEP4. Extracts derived from the CEP protocol, in contrast, were the least metabolically diverse. In fact, only four of the eight samples extracted with the CEP3 protocol showed positive results for all substrates. From this same treatment, the OMW-A extract was the one that exhibited only two metabolic activities, namely those related to carbohydrates and polymers (Fig. 4C). The high incubation temperature corresponding to the CEP3 protocol could be the cause of the lower microbial load of the extracts and thus of their functional metabolic diversity. Moreover, this agrees with the data corresponding to the values obtained for R and AWCD.

3.4. Correlation analyses

Various statistical analyses were performed on the basis of the results obtained. In particular, Table 3 (A-D) shows the Spearman correlations between the parameters studied as a function of the extraction protocols applied. Regarding protocols CEP1 and CEP4, a positive correlation was detected between of the BOD_5 and the germination index. Therefore, it seems evident that at mild extraction temperatures and incubation times equal to or >48 h, it is possible to obtain liquid organic amendments from compost that improve plant growth. In addition, it was also seen in Table 3A and B, that a high electrical conductivity reduces the microbiota present in the extracts. According to Zhang et al. (2019), a high concentration of salts, causes an increase in EC, and consequently a decrease in microbial load. In the CEP3 protocol, the parameters that negatively correlated with the germination index were determined. These were AWCD, pH, EC and TOC. Except for AWCD, all of them were physicochemical parameters that directly influence the survival of microorganisms. This correlates with the low AWCD values obtained for this treatment and fits with the microbial restriction implied by the application of aggressive temperatures.

In order to classify the different extracts obtained according to the variables studied, a discriminant function analysis was carried out (Fig. 5). In this case, the strong influence of the nature of the composts used to obtain the extracts was amply reflected. The discriminant analysis carried out explained 92.92 % of the variation observed. The first discriminant function explained a 55.07 % of variation, resulting in the differentiation of two groups of samples. One of them was formed by the VW samples (VW-A and VW-B), probably due to the unusual EC or pH values they showed. The other group included the rest of extracts. The second discriminant function explained the remaining 37.85 % of variation. It distinguished the extracts from OMW-B on the one hand, and those obtained from AW-B on the other. In these cases, both samples were separated from OMW-A and AW-A mainly by the high TOC values reached. A third group included extracts from the remaining raw materials.

Due to the rising price of fossil fuels, inorganic fertilizers have become more expensive, representing a significant monetary outlay for farmers

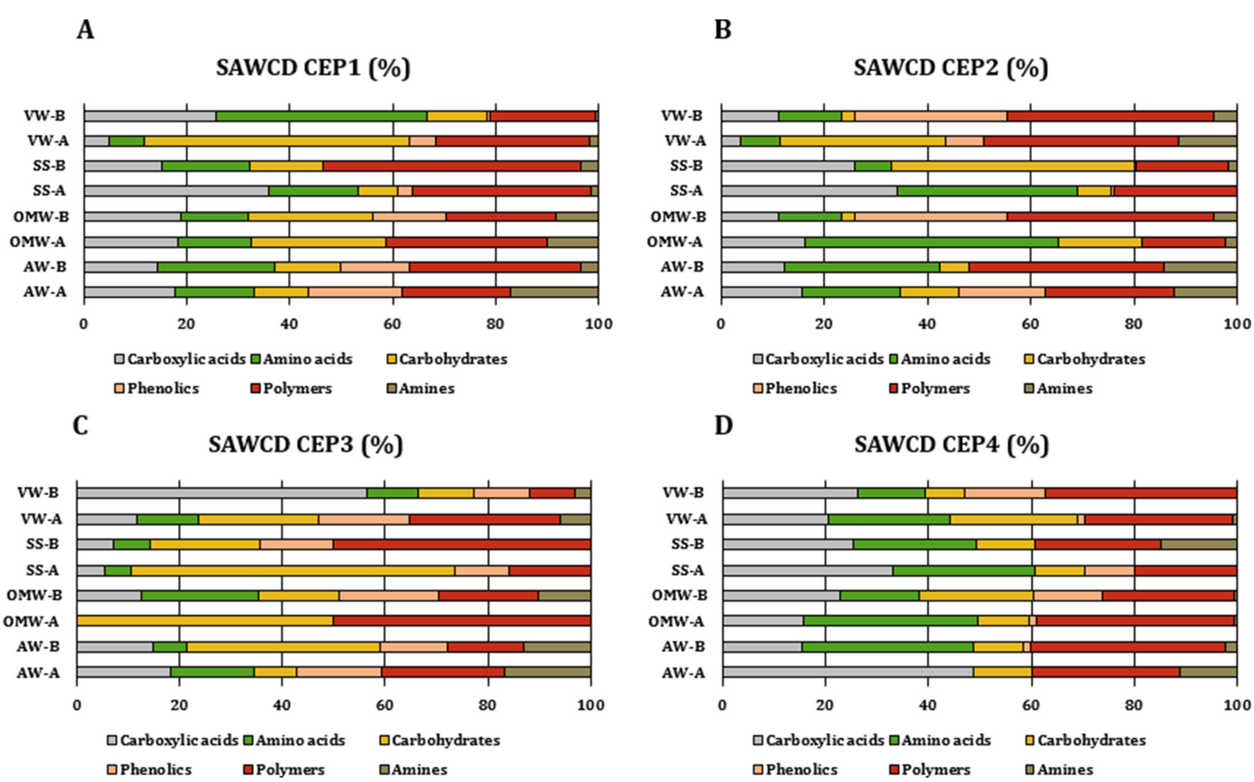

Fig. 4. Results of the metabolic diversity according to the Substrate Average Well Color Development (SAWCD) in percentage. A: SAWCD of Agri-food Waste (AW-A. B); B: SAWCD of Olive Oil Mill Waste (OMW-A. B); C: SAWCD of Sewage Sludge (SS-A. B); D: SAWCD of Vegetable Waste (VW-A, B).

Table 3
Spearman correlation of the four extraction protocols.

A	ph	EC	TOC	BOD_5	GI	R
AWCD	0.58	0.57	X	X	X	0.85
pH		X	0.49	X	X	0.5
EC			X	X	X	−0.64
TOC				X	X	X
BOD					0.47	X
GI						X
R						

B	ph	EC	TOC	BOD_5	GI	R
AWCD	X	X	X	X	−0.43	0.93
pH		X	0.43	X	X	X
EC			X	X	X	−0.41
TOC				X	X	X
BOD					X	X
GI						X
R						

C	ph	EC	TOC	BOD_5	GI	R
AWCD	0.69	X	0.6	X	−0.6	0.64
pH		0.51	0.65	−0.45	−0.63	X
EC			0.65	X	−0.56	X
TOC				X	−082	X
BOD						X
GI						X
R						

D	ph	EC	TOC	BOD_5	GI	R
AWCD	X	X	X	X	X	0.85
pH		0.54	X	X	X	X
EC			0.61	X	X	X
TOC				X	X	0.62
BOD					0.44	X
GI						X
R						

A: Spearman Correlation of Compost Extraction Protocol 1 (CEP1); **B:** Spearman Correlation of Compost Extraction Protocol 2 (CEP2); **C:** Spearman Correlation of Compost Extraction Protocol 3 (CEP3); **D:** Spearman Correlation of Compost Extraction Protocol 4 (CEP4). Abbreviations: EC: Electrical Conductivity; BOD_5: Biological Oxygen Demand; TOC: Total Organic Carbon; GI: Germination Index; Richness (R); Functional Intensity (AWCD).

(Walsh et al., 2012). From an economic point of view, our results therefore support the growing interest in the use of aqueous compost extracts especially under intensive growing conditions (Ye et al., 2020). In contrast, the application of extracts only involves mixing water and compost using simple infrastructures (Wang et al., 2018). This facilitates the scaling up of the production process. It also allows agriculture to be self-sufficient through the use of agricultural residues (Pane et al., 2016). In fact, two residues used in this work, VR-A and VR-B, come from horticultural residues. This is in addition to those composts that maximize the fertilizing capacity when converted into aqueous extract, especially with CEP1. Moreover, the use of this alternative is part of the circular economy. This work is the preamble for the use of aqueous compost extracts in protected agriculture through fertigation. With this, it will be possible to evaluate and compare efficacy with respect to agrochemicals. It will even be possible to analyze to what extent an extract can replace chemical fertilizers commonly used in fertigation.

4. Conclusions

In this study, the main factors affecting the quality of the compost extracts are determined. This study has been achieved by applying four different protocols in terms of temperature, agitation and incubation time. Thus, it was determined that protocols using mild temperatures, such as CEP1 and CEP4, allow obtaining liquid organic amendments with a suitable biofertilizing capacity, even increasing the phytostimulant character of the starting compost. In contrast, the most aggressive protocol, CEP3, compromises the survival of the microbiota and the functional biodiversity of the extracts, since it favors the extraction of phytotoxic substances. Additionally, scaling the obtaining process of aqueous compost extracts does not require high cost nor complex infrastructures. Thus, compared to agrochemicals, these extracts constitute a sustainable, effective and promising alternative to chemical fertilizers.

CRediT authorship contribution statement

R. Lerma-Moliz: Conceptualization, Investigation, Methodology, Data curation, Writing- Original draft preparation. **Juan A. López-González:** Investigation, Methodology, Writing- Reviewing and Editing. **F. Suárez-Estrella:** Investigation, Methodology. Project administration, Supervision. **María R. Martínez-Gallardo:** Investigation, Methodology. **Macarena M. Jurado:** Investigation, Methodology. **María J. Estrella-González:** Investigation, Metodology. **Ana J. Toribio:** Investigation, Metodology. **R. Jiménez:** Investigation, Methodology. **María J. López:** Conceptualization, Supervision, Visualization, Formal analysis, Writing- Reviewing and Editing.

Data availability

Data will be made available on request.

Declaration of competing interest

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

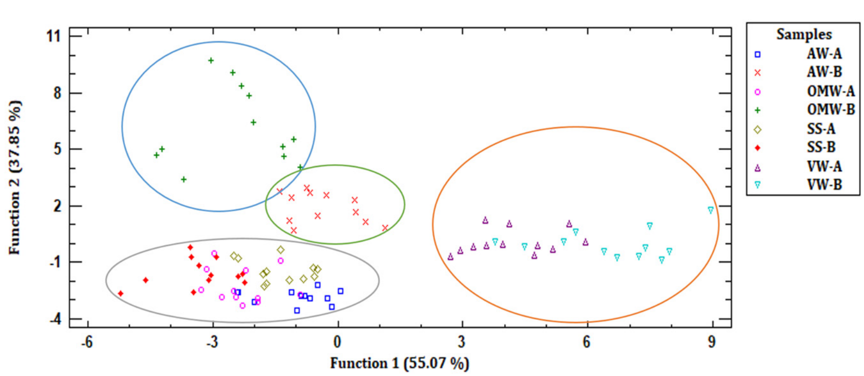

Fig. 5. Discriminant analysis loading plot of the different parameters analyzed in the aqueous compost extracts obtained. Abbreviations: AW-A. B: Agri-food Waste; OMW-A. B: Olive Oil Mill Waste; SS-A. B: Sewage Sludge; VW-A. B: Vegetable Waste.

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