## SCHÓLARENA

#### **RESEARCH ARTICLE**

# Comparative study of the fertilizing powers and phytotoxic potential of compost juice and compost from olive industry wastes

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## Abstract

In this work, we aim to valorize the olive industry wastes as organic fertilizers, an integral part of the circular economy. The study is focused on the evaluation of the fertilizing powers and phytotoxic potential of the compost juice in comparison with those of compost resulting from the co-composting of various olive industry wastes.

Our results showed that the phytotoxicity of compost juice depends mainly on the extraction factors. For the germination index (GI) determined on various seeds as alfalfa (*Medicago sativa*), water cress (*Lepidium sativum*), sorghum (*Sorghum bicolor*) and tomato (*Solanum lycopersicum L*.), the compost juice provides the better results. However, for the morphological and physiological parameters of plants studied, we have found that compost gives better results than compost juice.

A multivariate analysis using Principal Component Analysis (PCA) was performed in order to evaluate the effects of soil physicochemical characteristics on the development of physiological and morphological parameters of different plants in the different mixtures C/S mediums. The two principal axes extracted about 60% of the total variance with 47.49% for PC1 and 13.52% for PC2 at P=0.05 level.

Keywords: Compost tea; plant; soil; correlation

## Introduction

The end product of composting is usually a humus-rich organic amendment that works in the long term to improve the physical, chemical and biological properties of the soil (Walter et al. 2006). The organic matter contained in the compost is able to absorb water and retain it so that the plants use this reserve between rains and waterings, thus improving the water retention capacity in the soil (Annabi et al. 2007). Compost efficiently supplies phosphorus, potassium (36-48% is available to the plant), and trace elements to the soil and the plant (Soumare et al. 2003; Zhang et al. 2006).

Until recently, compost juice was defined simply as a liquid extract of composted material containing soluble organic and inorganic nutrients, and a large number of organisms, including bacteria, fungi, protozoa and nematodes (Scheuerell 2003). Recent researches on compost juice still defines it as a "compost water extract" which contains populations of beneficial microorganisms (Scheuerell 2002; Ingham 2005). Some researchers have shown that the use of compost juice helps reduce the severity of diseases such as powdery mildew and grape powdery mildew, caused by *Uncinulanector* and *Plasmopara viticola* respectively; gray mold of strawberries and late blight of potatoes (Elad et Shtienberg 1994; McQuilken et al. 1994; Yohalem et al. 1994); buttercup infestation and tomato blight (Touart 2000); tomato bacterial spot (Al-Dahmani et al. 2003); *Septoria* leaf spot (Gangaiah et al. 2004) and damping-off caused by *Pythium ultimum* (Scheurell et Mahaffee 2004).

The application of compost juice also showed an increase in the fouling of the tight clay soil; to meet the fertilizer needs of seedlings and increase the number of grass roots (Grobe 2003b). Scheuerell et Mahaffee (2002) claim that a better understanding of the microbiology of compost juice and the survival of microbe interactions on the surface of plants should lead to changes in the production practices and technology of application of compost juice.

It is well known that the Mediterranean countries are the most concerned by the major environmental problems generated by the olive industry (IOC 2017; Ghidaoui et al. 2019). Among these countries, Tunisia occupies the fourth largest producer of olive oil (Mekki et al. 2018; Meftah et al. 2019). Olive industry generates immense volumes of agro-industrial residues (Mekki et al. 2013; Gargouri et al. 2014; Al-Imoor et al. 2017). Such residues are mainly composed of solid organic residues and olives mill wastewaters (Bargougui et al. 2019, 2020; Sdiri et al. 2019).

This work is part of a project developed by our Laboratory, the main objective of which is to develop solution for the treatment and valorization of olive bio-wastes generated by the olive industry which constitute the vital sector of the Tunisian economy (Bargougui et al. 2019; 2020). It is in this context that this study aims to evaluate the fertilizing power and the phytotoxic potential of the compost juice prepared during a previous work in the same Laboratory (Bargougui et al., 2019) and to compare the effects of compost and compost juice on soil characteristics as well as on germination and growth of some plants species.

## Materials & methods

## **Biological Material**

#### The compost

The compost used is made by co-composting olive industry organic wastes mixed with poultry manures. The composting process was carried out by windrow composting for 6 months at the Olive Tree Institute of Sfax (Southern Tunisia). Mature compost is sieved and stored dry at room temperature (Bargougui et al., 2019).

#### The compost juice

The compost juice tested comes from the compost described above. It was prepared in the Laboratory of the Olive Tree Institute of Sfax (Btissam et al.,2010) Four factors are taken into consideration during the preparation of the juice, which are: the extraction ratio (compost/water): 1/2, 1/5 and 1/10; the extraction temperature: 25°C and 40°C; the extraction time: 24 hours, 72 hours, 5 days and 10 days; the method of extraction (aerated (A)/unaerated (UA)). For each ratio, the compost/water mixture is prepared, homogenized and then placed in an incubation tank according to the incubation factor (temperature). At the desired incubation time, the mixture is filtered and the collected juice is put in a container and stored in the refrigerator until use.

#### The soil

The soil used in our work was collected from the "Taous" experimental station of the Olive Tree Institute of Sfax, in an uncultivated area that has not undergone any prior treatment. For soil intended for Laboratory analyses, sieved to 2 mm and dried in ambient air then it was stored in plastic bags. A quantity of soil for determining mineral nitrogen and soil water content was kept cool at 4°C.

#### Test plant seeds

In order to assess the phytotoxic potential of the compost and the compost juice, 4 types of plant seeds were chosen, namely alfalfa (*Medicago sativa*), cress (*Lepidium sativum*), sorghum (*Sorghum bicolor*) and tomato (Solanum lycopersicum L). All seeds were obtained from the company AGRIPROTEC, located in Tunis and specialized in the marketing of seeds. The seeds were stored under optimal conditions for use in germination tests.

## Methods

#### Preparation of compost juice

For the preparation of the compost juice we used 20g, 40g and 100g of compost for 200 ml of distilled water, respectively for the ratios 1/10, 1/5 and 1/2 (compost/water). The preparation was carried out in Erlenmeyer flasks and well homogenized using a glass ring. Eight repetitions were performed for each report: four aerated and four unaerated. For aerated repetitions, the mixtures were taken daily and turned over using a glass ring then left under aeration for 10 to 15 minutes in a hood before returning them to the adequate temperatures (25 and 40°C). The unaerated ones were covered with parafilm to prevent oxygen penetration. For each repetition four different incubation times were used 24 hours, 72 hours, 5 days and 10 days and for each incubation time two incubation temperatures were used 25°C and 40°C. After the end of the incubation period, the mixture is collected and then filtered through a 2 mm sieve to obtain the compost juice.

#### Physicochemical characterization of the compost juice

The pH and the EC were measured using the method of Sierra et al. (2007). Suspended matter (SM) was determined by the filtration technique. Dry matter (DM) was evaluated by drying at 105°C. Organic matter (OM) and mineral matter (MM) were expected after calcinations at 550°C for 4 h. Kjeldahl total nitrogen was assessed using a standard method (Kandeler, 1995).

#### Soil characterization

The pH and the EC were measured using the method of Sierra et al. (2007). Suspended matter (SM) was determined by the filtration technique. Dry matter (DM) was evaluated by drying at 105°C.Organic matter (OM) and mineral matter (MM) were expected after calcinations at 550°C for 4 h. Kjeldahl total nitrogen was assessed using a standard method (Kandeler, 1995). Soil texture was determined using the pipette method (Thomas et al., 2012). Soil water retention capacity (SWRC) was evaluated gravimetrically by

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saturating the soil overnight. The pH and EC were measured using a pH meter and an EC meter, respectively. Dry matters (DM), organic matters (OM) and minerals matters (MM) were determined according to Sierra et al., (2007). The various soils nitrogen forms as total nitrogen (TN), ammoniacal nitrogen ( $NH_4$ -N), and nitrate nitrogen ( $NO_3$ -N) were expected according to the Kjeldahl method (Kandeler et al., 1995).

## **Experimental set-up**

The collected soil was well homogenized, then distributed in plastic pots(at the rate of 2 kg soil/pot). The pots used have a diameter of 14 cm, a depth of 13 cm.

Each pot is sown with 10 seeds. The seeds were sown on February 14, 2020. The experiment was carried out in four repetitions for each compost/soil ratio and two repetitions for the compost juice in comparison to a control batch, irrigated with tap water (i.e. 40 plants per batch of pots for control and compost/soil ratios and 20 plants per batch for compost juices). The plants were irrigated manually using a beaker every two days at the rate of 50 ml per pot. For the monitoring of plant growth in vivo, we chose to test the compost juice extracted at a single constant temperature (25°C), extracted during a period of 5 days for the three extraction ratios: 1/2, 1 / 5 and 1/10 (aerated (A) and unaerated (NA)). The doses used for irrigation of the plants are 50 ml/pot.

## Germination tests and monitoring of plants growth

The phytotoxicity potential of compost and compost juice were evaluated by measuring the germination index (GI %) of the four seeds mentioned above in comparison with distilled water as control medium (Zucconi et al., 1981).

In addition, and in order to follow the effects of the different ratios of compost/soil and compost juice on the growth of the different plants, some morphological parameters (as stems elongation, sheets number, leaves number and leaf area) and biochemical parameters (as the root part/shoot part ratio, the leaves chlorophyll content, the membrane permeability and the dry matter yield) have been realized the length of the cultivation period.

## Statistical analyses

For each parameter analyzed three replicates were made. All data obtained were analyzed by the SPSS statistical software (Statistical Package for the Social Sciences, version 20). Differences between mean values were calculated by analysis of variance (ANOVA), with a Duncan's test at probably level P=0.05. To study the correlation between soil and plants parameters, the Pearson correlation procedure was performed at P=0.05 probability levels.

## **Results and Discussion**

## Physicochemical and granulometric characteristics of the used compost and soil

The compost used has a pH (6.7) close to neutral, an elevated EC (7.3 mS cm<sup>-1</sup>), an important organic matter content (OM = 40.6 % DM) and with a C/N ratio of around 8.7 which testifies to its stability. Indeed, the C/N ratio is widely used as an indicator of the maturity of the compost (Sellami et al. 2007, Mekki et al. 2014; Bargougui et al. 2019). Pathak et al. (2011) have shown that a C/N ratio of around 10 is the ideal ratio for well matured compost. Wang et al. (2017) recorded higher values of the C/N ratio in the compost obtained from manure and straw. According to French standards (NF U 44-051, 1981), specifying and defining organic conditioners, this compost could be specified as vegetable compost since their total nitrogen expressed as a percentage on the basis of dry matter did not exceed 3% (2.7) and their organic matter content was less than 55 on the basis of dry matter (Table 1). Indeed, compared to French standards, the composition of minerals and fertilization confirmed the beneficial effect of this product as organic fertilizer.

Parameters	Compost	Soil
рН (25 °C)	6.7±0.1	8.56±0.2
EC (mS cm-1)	$7.3 \pm 0.00$	0.32±0.02
OM (%DM)	$40.6\pm0.00$	1.15±0.05
MM (%DM)	$59.4\pm0.01$	98.85±0.05
CEC (meq %DM)	$42.3\pm0.01$	3.2±0.1
TOC (%DM)	$23.54\pm0.01$	$0.66 \pm 0.01$
TN (% DM)	$2.7\pm0.03$	$0.07 \pm 0.01$
C/N ratio	$8.72\pm0.1$	9.42
K (g Kg-1)	$3.7 \pm 0.01$	0.12±0.01
P (g Kg-1)	$0.25\pm0.01$	$0.02 \pm 0.00$
Na (g Kg–1)	$0.27 \pm 0.02$	0.11±0.01
Phenolic compounds (g Kg–1)	$0.32\pm0.00$	ND
Sand (%)	ND	84.4
Silt (%)	ND	13.6
Clay (%)	ND	2.0

Table 1: Description of the parameters of model (1)

Table 1 recapitulates also the main granulometric and physicochemical characteristics of the soil studied. Analysis of the results shows that the soil used has a sandy texture (84.4% sand), with an alkaline pH (8.56). The OM content is very low (1.15% DM), which is in agreement with previous studies which confirm that Tunisian soils suffer from a deficiency in organic matter whose content does not exceed the 2% (Mekki et al. 2013; Chaari et al. 2014; Gargouri et al. 2014). The EC is of the order of 320  $\mu$ S cm<sup>-1</sup>, reflecting a relatively low mineral content. The total nitrogen content is around 70 mg Kg<sup>-1</sup>, which shows that the soil studied is poor in nitrogen. In addition, a low cation exchange capacity (CEC = 3.2 (meq % DM)) that our soil possesses is also reported by previous work (Magdich et al. 2012).

#### Effects of the various extraction factors on the compost juice characteristics

The effects of the various extraction factors on the compost juice parameters as the pH, the electrical conductivity (EC), the dry matter (DM) and the organic matter (OM) contents were investigated.

## Effects on the pH

The study of the effect of different factors on the compost tea pH shows that this parameter was not affected neither by the aeration conditions, nor by the extraction ratio nor also by the incubation time. However, the pH was slightly influenced by the temperature. A moderate difference (P<0.05) was noted between pH of compost tea extracted at 25°C and 40°C (Table 2). Indeed, all the pH values registered are close to neutral or slightly basic. Besides, neither the extraction ratio (1/2, 1/5, 1/10), nor the incubation time (24 hours, 72 hours, 5 days, 10 days) have influenced the acidity of the juice obtained and just slight insignificant differences are noted. Note that only the aeration factor has a slight influence on the pH. In fact, it is observed for the same extraction ratio having the same incubation period, the pH of the aerated repeats is greater than that of the unaerated ones except for the unaerated 1/5 ratio at 25°C, whose pH is higher than its aerated counterpart under the same conditions. The pH is practically the same to a few decimal places for the different ratios and this for the majority of the incubation times. In this context, Gorliczay et al. (2019) noted that the pH values of compost juice with an extraction time of 24 hours are neutral and slightly alkaline. By increasing the duration of the extraction, the pH values of the solutions decreased and shifted towards the acidic direction (table 2). A previous study showed a similar phenomenon where the pH values of compost juice after 56 h of extraction was 6.08, while after 112 h of extraction it was 5.97 (Martin et al. 2012). This process can be explained by the fact that atmospheric CO<sub>2</sub> or CO<sub>2</sub> produced by microbial activities (anaerobic bacteria, fungi) dissolves in the compost juice forming carbonic acid, which is a weak acid, thus causing a decrease in pH over time (Zumdahl 1993).

Temperatur	е	25°C				40°C				
Parameter	Incubation time Extraction ratio	24H	72H	5D	10D	24H	72H	5D	10D	
	1/2 4	8.19±0.02	8.54±0.2	7.24±0.01	8.58±0.12	8.44±0.01	8.21±0.03	7.71±0.2	7.9±0.0	
	1/2 A	А	А	А	А	В	В	В	В	
	1/5 A	8.19±0.04	8.57±0.01	7.34±0.33	7.95±0.04	8.21±0.15	8.10±0.12	7.87±0.01	7.85±0.02	
		А	А	А	А	В	В	В	В	
	1/10 A	8.23±0.01	8.63±0.12	7.44±0.01	7.89±0.13	8.25±0.0	7.78±0.22	7.91±0.02	7.62±0.01	
pН		А	А	А	А	В	В	В	В	
	1/2 114	6.97±0.01	7.98±0.2	6.82±0.01	$7.48 \pm 0.04$	7.91±0.5	7.87±0.05	6.93±0.2	7.93±0.01	
	1/2 0/1	А	А	А	А	В	В	В	В	
	1/5 114	7.56±0.12	8.23±0.01	6.97±0.11	7.47±0.03	7.81±0.01	7.58±0.12	7.14±0.31	7.80±0.0	
	1/5 UA	A	А	A	А	В	В	В	В	
	1/10 114	8.15±0.0	8.28±0.1	7.41±0.04	7.25±0.0	7.78±0.12	7.35±0.01	7.29±0.33	7.78±0.02	
	1/10 UA	А	А	А	А	В	В	В	В	

Average values of five replications ± SD; ND: not determined.

**Table 2:** Effects of different extraction factors (extraction ratio, temperature, time incubation and aeration) on the compost tea pH Values represent the means of 3 replications per treatment ± SD. For each parameter, capital letter (A, B) in the same line indicate difference between temperatures. A: aerated. UA: unaerated.

## Effects on the electrical conductivity (EC)

The electrical conductivity (EC) makes it possible to obtain an estimate of the overall content of soluble salts in the compost juice. The study of the effects of the compost juice extraction factors on its EC is summarized in table 3. Based on the results obtained, we notice that the incubation temperature greatly influences this parameter. Indeed, the EC values of juices incubated at 40°C are much higher than those incubated at 25°C. This may be due to the evaporation of water at 40°C which will cause a salt concentration situation. Likewise, the juice extraction ratio influences the EC. The more the extraction ratio decreases, the more the EC declines and vice versa. This is because, by using a larger amount of compost, the extract becomes more concentrated, so the amount of dissolved anions and the conductivity values of which increase. The 1/2 report shows the highest EC values for the different incubation times and for the different temperatures regardless of the aerated or unaerated approach. This is due to the decrease in the salt content of the juice implied by the decrease in the amount of compost used for juice extraction.

Temperatur	e	25°C				40°C				
Parameter	Incubation time Extraction ratio	24H	72H	5D	10D	24H	72H	5D	10D	
	1/2 4	3.71±0.01	5.78±0.01	5.38±0.2	7.09±0.02	7.70 ±0.1	8.60±0.11	9.14±0.3	12.75±0.01	
	1/2 A	A a	Aa	Aa	Ab	Aa	Aa	Aa	Ab	
	1/5 A	1.91±0.2	2.81±0.02	2.36±0.01	4.06±0.04	5.59±0.03	6.80±0.7	6.27±0.2	10.43±0.41	
		Ва	Ва	Ba	Bb	Ba	Ва	Ва	Bb	
	1/10 A	1.22±0.01	1.57±0.1	1.60±0.12	2.67±0.3	4.89 ±0.2	5.30±0.1	5.00±0.0	8.93±0.2	
EC (mS.		Ba	Ba	Ba	Bb	Ba	Ba	Ba	Bb	
cm <sup>-1</sup> )	1/2 UA	1.92±0.2	5.64±0.02	5.10 ±0.0	5.80 ±0.01	7.88±0.13	7.69 ±0.3	8.74±0.01	12.20±0.1	
		Aa	Aa	Aa	Ab	Aa	Aa	Aa	Ab	
	1/5 114	1.75 ±0.0	2.61±0.1	$2.48 \pm 0.01$	$3.41 \pm 0.11$	3.86±0.2	5.64±0.4	$5.40 \pm 0.0$	10.10±0.0	
	1/5 UA	Ва	Ва	Ва	Bb	Ba	Ва	Ba	Bb	
	1/10 114	$1.15 \pm 0.01$	$1.77 \pm 0.01$	$1.46 \pm 0.01$	2.33±0.3	3.95±0.0	4.62±0.02	3.70±0.1	8.38 ±0.01	
	1/10 UA	Ba	Ba	Ba	Bb	Ba	Ba	Ва	Bb	

**Table 3:** Effects of different extraction factors (extraction ratio, temperature, time incubation and aeration) on the compost tea electric conductivity. Values represent the means of 3 replications per treatment  $\pm$  SD. For each parameter, capital letter (A, B) in the same colon indicate difference between extraction ratios. Tiny letter (a, b) in the same line indicate difference between time incubation. Means followed by the same letter are not significantly different at *P*=0.05 level. Different letters indicate significant differences between treatments (*p*≤0.05, Duncan Test). A: aerated. UA: unaerated.

The incubation time also influences EC. Overall, the incubation time tends to increase electrical conductivity (Kim et al. 2015). There is a slight fluctuation between certain extraction times. We first note an increase in EC around 72 hours and then a decrease around the 5<sup>th</sup> day. Subsequently this conductivity will increase and reach a maximum value on the 10<sup>th</sup> day. In addition, aerated compost juices have the highest salinity. Thus our results are consistent with those of Gorliczay et al. (2019) who showed that the values of the EC decrease with the decrease in the compost/water extraction ratio.

## Effects on the dry matter (DM) content

Concerning the effects on the DM content, results showed that this parameter is greatly influenced by the extraction ratio. Indeed, the higher is the extraction ratio, the higher is the DM content. Temperature also influences the DM content. This later is elevated for juices incubated at 40°C than those incubated at 25°C. For the same volume, we found a higher DM concentration in the juice incubated at 40°C (table 4). In addition, we have found that the aeration factor decreases the DM content for juice incubated at 40°C. For the first three incubation times, the aerated juice (A) has a significantly higher DM content than the unaerated juice (UA). However, for juice incubated for 10 days at 40°C, we see the opposite effect and the unaerated juice has a higher DM content. For juice incubated at 25°C the aeration factor influences the DM content of the juice for the incubation times of 5 and 10 days where it is noted that aerated juices have a higher DM content than the unaerated ones (table 4).

Values represent the means of 3 replications per treatment  $\pm$  SD. For each parameter, capital letter (A, B, C) in the same line indicate difference between temperatures. Tiny letter (a, b, c) in the same clone indicate difference between extraction ratios. Means followed by the same letter are not significantly different at *P*=0.05 level. DM: dray matter. FM: fresh matter. A: aerated. UA: unaerated.

Temperature		25°C				40°C				
Parameter	Incubation time Extraction ratio	24H	72H	5D	10D	24H	72H	5D	10D	
	1/2 A	1.03±0.001 A a	1.62±0.02 A a	0.95 ±0.1 A a	2.33±0.1 A a	3.55 ±0.03 B a	7.00±0.0 B a	4.80±0.01 B a	2.6±0.0 B a	
	1/5 A	0.47±0.01 A b	0.75±0.04 A b	0.82±0.2 A b	1.06±0.0 A b	1.36±0.11 B b	5.67± 0.02 B b	1.66±0.03 B b	1.03±0.01 B b	
DM (%)	1/10 A	0.17±0.003 A b	0.49±0.02 A b	0.45±0.1 A b	0.64±0.11 A b	1.13±0.02 B b	1.46±0.04 B b	1.69±0.2 B b	0.75±0.0 B b	
FIVI	1/2 UA	1.30±0.2 A a	2.14±0.11 A a	0.82±0.04 A a	1.21±0.1 A a	1.60±0.0 B a	2.34±0.05 B a	3.89±0.33 B a	5.08±0.2 B a	
	1/5 UA	0.34±0.01 A b	0.52±0.12 A b	0.46±0.05 A b	0.69±0.03 A b	0.79±0.2 B b	2.78±0.12 B b	0.67±0.10 B b	2.97±0.3 B b	
	1/10 UA	0.17±0.02 A b	0.48±0.01 A b	0.32±0.02 A b	0.36±0.0 A b	0.72 ±0.1 B b	0.78±0.21 B b	0.47±0.1 B b	1.23±0.01 B b	

**Table 4:** Effects of different extraction factors (extraction ratio, temperature, time incubation and aeration) on

 the compost tea dry matter (DM) content.

Values represent the means of 3 replications per treatment  $\pm$  SD. For each parameter, capital letter (A, B) in the same line indicate difference between temperatures. Tiny letter (a, b) in the same clone indicate difference between extraction ratios. Means followed by the same letter are not significantly different at *P*=0.05 level. DM: dray matter. FM: fresh matter. A: aerated. UA: unaerated.

#### Effects on the organic matter (OM) content

Regarding the organic matter (OM) content, we didn't notice an exact proportionality between the extraction ratio and the OM content of the juice. However, in terms of incubation time, OM tends to decrease for juice incubated at 25°C and to increase for juice incubated at 40°C. The temperature does not greatly influence the OM content. The incubation time predominates more over the temperature factor. Alternatively, for juice incubated at 40°C, the aeration factor greatly influences the OM content. Indeed, aerated juices have a greater evaporation which will lead to a concentration of the juice and an increase in the OM content (table 5).

Values represent the means of 3 replications per treatment  $\pm$  SD. For each parameter, capital letter (A, B, C) in the same line indicate difference between temperatures. Tiny letter (a, b, c) in the same clone indicate difference between extraction ratios. Means followed by the same letter are not significantly different at *P*=0.05 level. A: aerated. UA: unaerated.

Tempera	ture	25°C	·			40°C					
Para meter	Incuba tion time/ Extrac tion ratio	24H	72H	5D	10D	24H	72H	5D	10D		
OM (%DM)	1/2 A	53.60±0.55 Aa	49.62±0.2 Aa	56.34±1.2 Aa	57.79±0.2 Aa	45.54 ±0.22 Ba	50.86±0.66 Ba	61.41±0.1 Ba	69.53±0.15 Ba		
	1/5 A	51.10±0.25 Aa	46.12±0.2 Aa	51.13 ±2 Aa	52.24±0.3 Aa	49.76±0.11 Ba	56.57±0.25 Ba	67.26±0.1 Ba	75.15±0.22 Ba		
	1/10 A	30.02±0.1 Ab	30.53±0.2 Ab	31.04±0.55 Ab	30.61±1.1 Ab	39.46±0.05 Bb	35.24±0.22 Bb	37.49±0.3 Bb	36.42±1.2 Bb		
	1/2 UA	55.45±0.1 Aa	53.50±0.2 Aa	49.67±0.3 Aa	47.81 ±0.1 Aa	43.39±0.02 Ba	46.18±0.65 Ba	60.66 ±0.6 Ba	65.53±0.01 Ba		
	1/5 UA	54.56±0.22 Aa	48.81±0.35 Aa	49.67±0.04 Aa	49.36±0.22 Aa	39.59± 0.1 Ba	46.45 ±0.02 Ba	49.29±0.55 Ba	43.23±0.13 Ba		
	1/10 UA	39.67±0.12 Ab	30.03±0.01 Ab	33.83±0.02 Ab	31.12±0.2 Ab	38.39±0.35 Bb	35.96±0.04 Bb	35.09±0.6 Bb	39.33±0.1 Bb		

**Table 5:** Effects of different extraction factors (extraction ratio, temperature, time incubation and aeration) on

 the compost tea organic matter (OM) content.

Values represent the means of 3 replications per treatment  $\pm$  SD. For each parameter, capital letter (A, B) in the same line indicate difference between temperatures. Tiny letter (a, b) in the same clone indicate difference between extraction ratios. Means followed by the same letter are not significantly different at *P*=0.05 level. A: aerated. UA: unaerated.

## Effects of the various extraction factors on the phytotoxicity of compost juice

The effects of the different extraction factors on the phytotoxicity potential of the compost juice were investigated. The germination of watercress (*Lepidium sativum*) showed germination index (GI) for the most part lower than that of the control ones (figure 1). The 1/10 A (aerated) and UA (unaerated) ratios showed the highest germination index when the juice is incubated at 40°C for three days. There is no exact proportionality between the extraction ratio and the germination seeds number. It can be seen that the germination index tends to decrease when switching from juice incubated at 25°C to one incubated at 40°C. At 25°C, we notice that the germination index varies depending on the incubation time. Initially high for juice incubated for 24 hours, these indices will drop for juice incubated for 72 hours and subsequently increase for juice incubated for 5 days. These values decrease to low values for the juice incubated for 72 hours compared to that of 24 hours and then decrease for the juice incubated for 5 days. The lowest values were observed for the juice incubated for 10 days.



**Figure 1:** Germination index (GI %) of different tested species (water cress (*Lepidium sativum*), alfalfa (*Medicago sativa*), sorghum (*Sorghum bicolor*) and tomato (*Solanum lycopersicum L.*) in function of the compost juice extractions factors.

Intended for alfalfa (*Medicago sativa*), it is noted that the longer the incubation time of the juice increases, the more the germination index tends to decrease, regardless of the incubation temperature. We also observe that the 1/10 ratio gave the best germination indices for all temperatures and incubation times. However, the extraction factor plays an important role in the phytotoxicity of the juice, so the more the ratio decreases, the more the germination index increases.

The sorghum germination test (*Sorghum vulgare*) reveals that for the juice incubated at 40°C almost all the values of the germination indices are lower than that of the control, which could reflect a phytotoxic potential of this juice. A few values slightly exceed the value of the germination index of the control. They are 1/10 A, 1/5 A incubated for 3 days and 1/10 A incubated for 10 days. For juice incubated at 25°C, a good germination index is observed for juice incubated for 10 days except for the 1/2 A ratio. So temperature plays a key role in lowering the germination index. Indeed, the germination indices decrease considerably for the juices incubated at 40°C compared to the juice incubated at 25°C.

The majority of tomato (*Solanum lycopersicum L*.) germination indices for all the juice ratios tested are lower than those of the control. The 1/5 and 1/10 ratios show the best germination indices for juice incubated at  $25^{\circ}$ C and that incubated at  $40^{\circ}$ C. The incubation time greatly influences the phytotoxicity of the juice. It is noted that the lowest germination indices are observed at the

level of the juice incubated at 10 days. The extraction ratio slightly influences the germination indices. Neither the aeration factor nor the temperature influences the germination indices of the tomato. In this context, Ghidaoui et al. (2019) have shown that the dilution of olive mill waste waters can reduce its toxicity and improve the faba bean (*Vicia faba*) seeds germination. Others researchers have confirmed that olive wastes salinity and their phenolic compounds content constitute the major elements of their phytotoxicity (Mekki et al. 2007; Saadi et al. 2007; Mechri et al. 2011; Bargougui et al. 2019).

#### Effects of compost and compost juice on plants morphological parameters

A comparative study of the effects of compost and compost juice on the morphological development of the plants mentioned above was carried out in vivo. The morphological parameters monitored were; the plant total length, the aerial part length, the root part length, the leaves number and the leaf area. For the C/S mixtures ratios, we found a higher growth of the aerial part than that of the control for sorghum and tomato while for alfalfa there was no significant difference compared to that of the control. For the root part, there was no significant difference compared to the control (figure 2). Indeed, due to its mineral content, the compost stimulated the growth of tomato and sorghum. For sorghum, the growth peak was seen for the 1/2 C/S ratio, while for tomato it was at the 1/10 C/S ratio (figure 2). Olivier et al. (2019) studied the effect of different C/S ratios on the growth of three varieties of plantains. Their results showed that the lowest growth values were observed at the level of the controls. In the case of aerated juice, the effect of the juice varies from one species to another. It was observed that the growth of alfalfa was approximately equal to that of the control. In the case of sorghum this growth was slightly greater than that of the control. For the tomato, this growth was lower than that of the control. These results were consistent with those of El-Tantawy et al. (2009) who obtained results showing that with the application of compost juice on tomato plants the control plants had the most growth. Indeed, previous researches registered that compost juice significantly improves plant size (Azza et al. 2010; Gharib et al. 2008).









**Figure 2:** Effects of various mixtures compost/soil and compost tea extraction ratios on plants growth (TL: total length, RL: root length, SL: shoot length).

For C/S mixtures, there was an increase in the number of sheets depending on the ratio. The more the ratio decreases, the more the number of sheets increases (figure 3). The highest number of sheets was registered for the 1/10 C/S ratio. We have found that compost juice does not have a great influence on the leaves number in sorghum but decreases considerably for alfalfa and moderately for tomato. In this context, Zraibi et al. (2015) found that compost improves the leaves number of the plants, which was in part consistent with our results because for alfalfa there was a considerable decrease in the leaves number.







Figure 3: Effects of various mixtures compost/soil and compost tea extraction ratios on plants leaves number.

The effect of compost on the leaves number was identical to that of compost juice except for the tomato where it is noted that this number is higher compared to the control. These results diverge from those obtained by Mrabet et al. (2011). Compost juice influences the number of leaves differently depending on the species. Thimothy (2017) had shown that compost juice did not greatly influence the number of leaves. For the aerated juice the values are lower than those of the control and decrease when the concentration of the juice decreases. As for the unaerated juice, the 1/2 ratio has a value slightly higher than that of the control and tends to decrease when the concentration of the juice decreases. The leaf area of the tomato is for the majority of the reports superior to that of the control except for the 1/2 UA report (figure 4). The highest leaf area values were measured for juice for the C/S ratio. Compost and compost juice have a beneficial effect on the leaf surface of the tomato. The lowest value of leaf area was observed at the level of 1/2 UA. Olivier et al. (2019) investigated the effect of different compost ratios on the leaf area of three varieties of plantains. Their results showed that compost improves the surface area of two of the varieties studied and has no significant effect on the third. We can deduce that the effect of compost on the leaf surface varies depending on the variety and probably the species, which agrees with our results obtained. Btissam et al. (2010) carried out trials of ferti-irrigation of tomato plants with compost juice. Their results showed an increase in the leaf area of the tomato plants compared to the control plants.







Figure 4: Effects of various mixtures compost/soil and compost tea extraction ratios on plants Leaf area.

#### Effects of compost and compost juice on plants physiological and biochemical parameters

The comparative effects of compost and compost juice on some physiological and biochemical parameters of the plants tested were studied. All the parameters relate to the contents of chlorophyll pigments, the leaf membrane permeability, the leaf water content as well as the fruit yield were determined. For Chl a, Chl b and carotenoids, it can be seen that the values for all the plants were for the most part greater than those of the controls except for a few values approximately equal to those of the control (figure 5). El Kadiri Boutchich et al. (2016) performed chlorophyll assays on several plants grown on soils with different compost/soil ratios. Their results showed that all the plants tested have a higher chl a and chl b contents than the control, which was in agreement with our results. However, Toundou O. (2016) obtained total chlorophyll contents approximately equal to those of the control. Chaichi and Djazouli (2017) performed chlorophyll measurements on the irrigated bean with different ratios of compost juice. Their results showed that the bean plants irrigated with the compost juice have total chlorophyll much higher than the controls. Such results were consistent with those we obtained.







Figure 5: Effects of various mixture compost/soil and compost tea extraction ratios on plants chlorophyll and carotenoids content.

The membrane permeability does not present a lot of variation either for the compost/soil ratios or for the compost juices. It is practically equal to that of the controls for the different species. In some reports, it is found that this membrane permeability is slightly low compared to that of the control. This is the case of 1/2 A of alfalfa, 1/2 and 1/5 C/S of sorghum and 1/2 C / S and 1/10 UA of tomato (data not shown). For the 3 species, the highest value was observed for 1/2 A of sorghum and the lowest for 1/10 UA of tomato (data not shown). Indeed, the study of SOME et al. (2014) on cowpea (Vigna unguiculata «bean») has shown that the compost does not significantly influence membrane permeability. Compost juice contains a significant amount of humic and fulvic acids. Several authors attribute to humic acids an influence on membrane permeability, thus facilitating the transfer of mineral elements (Pinton et al. 1997, 1999). Our results for the C/S ratio were correlated with those of Toundou (2016) who demonstrated that whatever the treatment considered, the water content does not vary over time and is approximately equal for all compost/soil treatments.

The water content does not show significant changes from the control. For tomato, for 1/5 A we observed that the water content of the aerial part is much lower than that of the control (figure 6). This is partly explained by the delayed germination of the plant which mean that at the time of the measurements there was a significant morphological difference between 1/5 A and the other reports. The water content of the plants of the different ratios does not differ greatly from that of the control, whether for the aerial part or the root part. However, some values are lower than that of the control, namely 1/5 A of alfalfa and tomato, and 1/2 C/S of sorghum. The origin of this decline could be related to external factors. Our results for the C/S ratio are correlated with those of Toundou (2016) who demonstrated that whatever the treatment considered, the water content does not vary over time and is approximately equal for all compost/soil treatments.

Regarding the effects on yield, for sorghum, we noted the appearance of ears in all the pots except for the control and the 1/2 ratios (C/S, A and UA). For the tomato only the compost/soil ratios produced fruit. For alfalfa there is no fruit but we observed flowering for the compost/soil ratios. For all the plants, we noticed that the 1/5 C/S ratio gave the best result with higher blooms or a higher number of fruits or ears. It is followed closely by the 1/10 C/S ratio and finally comes the 1/2 C/S ratio (data not shown). Toundou O. (2016) through their results highlighted the beneficial effects of compost for the yield of a plant. Having tested different composts on tomato and corn, they found that all the composts used had a higher yield than the control. For the compost juice only the ratios 1/5 and 1/10 for the sorghum one produce seeds. For the other species there was no difference from the control.







Figure 6: Effects of various mixture compost/soil and compost tea extraction ratios on plants shoot and root water content.

## Effects of compost and compost juice on soil physicochemical characteristics

In order to know the effects of amendments added on the soil, a set of physicochemical parameters such as the pH, the EC, the OM, the total nitrogen Kjeldahl (TNK) and the phenolic compounds content of soils receiving compost or compost juice and planted by the plant species mentioned above were investigated. Such parameters were monitored in comparison to the uncultivated control soil at the end of the cultivation period (table 6).

	Soil Co	ntrol	-	Soil + A	Soil + Alfalfa			orghum	-	Soil +Tomato		
	pН	EC	OM	pН	EC	OM	pН	EC	OM	pН	EC	OM
С	8.92±	306.5±	0.41±	8.35±	287±	1.58±	8.71±	432±	2.09±	7.98±	338±	2.92±
	0.001	0.45	0.003	0.03	2.1	0.004	0.2	0.25	0.001	0.02	0.0	0.01
	a	a	a	a	a	a	a	a	a	a	a	a
1/2 C/S	7.96±	1647±	29.58±	7.79±	1199±	10.19±	7.86±	3090±	8.91±	7.63±	2600±	10.13±
	0.2	2.5	0.25	0.2	0.75	0.12	0.01	4.5	0.2	0.13	0.0	0.1
	b	b	b	b	b	b	b	b	b	b	b	b
1/5 C/S	7.89±	775±	11.23±	7.86±	453±	4.05±	7.87±	355±	3.93±	8.11±	485±	4.07±
	0.10	0.55	0.01	0.01	1.5	0.01	0.33	0.0	0.04	0.01	0.2	0.32
	c	c	c	b	c	c	b	c	c	c	c	c
1/10 C/S	7.77±	500±	7.34±	7.53±	308±	2.92±	7.95±	336±	2.97±	8.23±	394±	2.75±
	0.03	0.0	0.2	0.04	0.0	0.01	0.0	0.45	0.01	0.02	0.5	0.025
	d	d	d	b	d	d	c	d	d	c	d	a
1/2 A	8.92±	306.5±	0.41±	8.69±	271±	1.28±	8.09±	512±	3.33±	8.39±	288±	1.43±
	0.01	0.45	0.003	0.21	0.66	0.2	0.01	0.2	0.0	0.01	0.0	0.01
	a	a	a	c	a	e	d	e	e	d	e	d
1/5 A	8.92± 0.01 a	306.5± 0.45 a	0.41± 0.003 a	8.41± 0.03 a	254± 0.2 e	1.86± 0.03 f	8.48± 0.1 e	503±3 f	1.63± 0.01 f	8.36± 0.22 d	328± 0.0 a	1.55± 0.0 d
1/10 A	8.92±	306.5±	0.41±	8.81±	380±	1.92±	8.69±	450±	2.28±	8.07±	302±	1.3±
	0.01	0.45	0.003	0.11	0.0	0.1	0.02	0.0	0.02	0.01	0.2	0.0
	a	a	a	c	f	f	a	a	a	a	d	e
1/2 UA	8.92±	306.5±	0.41±	8.40±	377±	0.94±	8.53±	305±	1.35±	8.44±	303±	1.61±
	0.01	0.45	0.003	0.0	0.14	0.001	0.04	0.0	0.1	0.3	0.75	0.01
	a	a	a	a	f	g	f	d	f	d	d	f
1/5 UA	8.92±	306.5±	0.41±	8.71±	294±	2.25±	8.76±	417±	1.26±	8.49±	256±	1.48±
	0.01	0.45	0.003	0.01	0.2	0.0	0.11	0.5	0.003	0.01	0.0	0.02
	a	a	a	c	a	h	g	a	f	d	e	d
1/10 UA	8.92± 0.01 a	306.5± 0.45 a	0.41± 0.003 a	8.60± 0.02 c	306± 0.0 d	1.37± 0.01 e	8.77± 0.04 g	321±1 d	1.61± 0.01 f	8.22± 0.02 c	237± 0.55 e	1.27± 0.1 e

**Table 6:** Physicochemicals characteristics of soil control and soils cultivated after six month of plants cultivation. Values represent the means of 3 replications per treatment  $\pm$  SD. For each parameter, Tiny letter (a, b, c, d, e, f, g) in the same clone indicate significant difference between treatments. Means followed by the same letter are not significantly different at *P*=0.05 level. A: aerated. UA: unaerated.

For the pH, we noted that the pH values of the planted soils were lower than the pH of the initial soil. For the compost/soil mixtures, a decrease in the pH was observed for all the C/S ratios compared to the control soil except for the 1/5 and 1/10 C/S cultivated by tomato. These results agree with those of Mrabet et al. (2011) and El-Gizawy et al. (2013) who demonstrated that whatever the applied dose of the compost, the pH undergoes a slight decrease. Alternatively, Toundou et al. (2014) suggested that compost increases soil pH. However compared to the control soil after treatment, for alfalfa and tomato we noted a slight increase in pH. These results diverge from those obtained by Nasef et al., (2009) and Enshrah et al. (2016) who demonstrated that compost juice decreases soil pH. In the case of sorghum, soils irrigated with A showed a decrease in pH while those irrigated with UA increase the pH except for the 1/2 UA ratio whose pH has decreased.

Concerning the EC values, for the control pots, there was no great difference in EC before and after the cultivation. In fact, for the soil cultivated by alfalfa, the EC decreased slightly compared to that of the control soil while it increased in soils cultivated by sorghum and tomato. The EC for the C/S ratio tends to decrease compared to the EC without crop except for the 1/2 C/S ratio of sorghum and tomato where the latter increases considerably. After treatment, it was found that the higher the C/S ratio, the higher the EC (table 6). Our results agree with those of Toundou et al. (2014) and Mrabet et al. (2011) who showed that soil salinity in terms of electrical conductivity gradually increases in the presence of compost. This increase was all the greater the higher the concentration of the compost. However, Nasef et al. (2009) found that the compost addition reduce the soil salinity.

At the level of the pots irrigated with the compost juice, there was not much difference compared to the values before treatment. Indeed, the values obtained after treatment were between 237  $\mu$ S.cm-1 and 512  $\mu$ S.cm-1. There was no exact proportionality in the case of the EC for soils irrigated with juice. However, we noticed that for tomatoes the value of the EC of the soil decreases compared to that of the control pot. Such findings agree with those of El-Gizawy et al. (2013) who explained that the stability of aggregates due to the addition of compost tea tends to change the pore size distribution, bulk density, water percolation and decrease in soluble salts. For alfalfa we found that only the soils irrigated with 1/2 and 1/5 A juice had a lower EC than the control soil. Then again, for sorghum, the soils irrigated by juice 1/2 and 1/5 A showed a higher EC than the control soil while the others showed a lower EC (table 6).

For the effects on OM content, we found that the OM content of C/S mixtures decreases compared to the initial soil. This can be explained by the mineralization of organic matter by microorganisms in order to provide the plant with the mineral elements essential for its growth. Compared to the control treatment after culture, we noticed an improvement in this OM content for the 1/2 and 1/5 C/S ratio. the 1/10 C/S ratio, this content was approximately equal to that of the control (table 6).

El-Gizawy et al., (2013) obtained similar results showing that the compost amendment of a soil improved the organic matter content of the soil, which could be due to the organic matter richness of the compost. For the soil irrigated with compost juice, we obtained values ranging between 0.94 and 3.33%. These values do not show a significant difference from the control treatment after culture. In this context, our results do not agree with the findings of Enshrah et al. (2016) who postulate that irrigation with compost juice improves the organic matter content of the soil. Regarding the total nitrogen Kjeldahl (TNK) content, high nitrogen content was noted in the soil on which alfalfa was grown compared to other plants. Indeed, alfalfa is known for its atmospheric nitrogen fixing effect, which explains these results (data not shown). Indeed, Aparicio-Tejo et al. (1981) showed the ability of alfalfa to store nitrogen in the nodules thanks to soil microorganisms and enzymes. In addition, our results showed that soil/compost mixtures have higher nitrogen content

Polyphenols were known by their negative role in the process of organic matter mineralization either by complexing protein nitrogen which becomes inaccessible to microorganisms or by inhibiting microorganisms (Duponnois et al. 2001; Diallo et al. 2006). We have measured the polyphenols content of cultivated soils. The analysis of the results obtained shows that the C/S ratios present the highest polyphenols contents for all soils with the highest value for the ratio 1/2 C/S comparatively to that of the control soil (data not shown). For the soils irrigated with the juice, their polyphenols content is slightly higher than that of the control. Likewise, we have observed that the soil cultivated by tomato has the highest polyphenol content. Such results were consistent with previous researches which confirmed that soil amendment with compost increased its polyphenols content (Hchicha et al. 2006; Said-Pullicino et al. 2010).

#### Correlation between soil characteristics and plants growth

A multivariate analyses PCA was performed in order to evaluate the effect of soil physicochemical characteristics on the development of physiological and morphological parameters of different plants studied. The two principal axes extracted about 60% of the total variance with 47.49% for PC1 and 13.52% for PC2 (figure 7). Axis 1 was mainly associated with soil characteristics (OM, TNK, Poly, EC, pH...). The principal group, which was positively correlated with PC1 included OMi, ECi, TNKi, Polyi, Polyt, ECt, OMt, ECa, Polya, TNKs, Polys, OMs and contributed significantly to this axis. All parameters in this group were highly and positively correlated. Indeed, after six month of plant cultivation, it's clear that the initial soil characteristics as his organic matter and mineral content highly influence its final characteristics independently of plant nature. The most important correlation was performed between ECi and ECa (0.965), ECi and ECs (0.921), ECi and ECt (0.962), OMi and OMa (0.985), OMi and OMs (0.960), Poly I and Polyt (0.987), TNKi and TNKs (0.947), TNK I and TNKa (0.929). This parameter was negatively associated to soil pH (pHi, pHa, pHs, pHt).

The axis 2 was related to leaf trait of alfalfa. LAa, Chlaa, Chlba, Carta, SLa correlated positively to axis 2. This group of parameter was negatively correlated to WCSa. The detailed analysis of relationships among variables showed that plant morphological and physiological parameters had a tendency to correlate with some soil characteristics. In fact, for alfalfa, TL, SL and RL tended to decrease with the increase of soil EC, OM and Polyphenols content. However, alfalfa growth was positively correlated to soil pH. On the other hand, leaf trait was highly and positively correlated with LA, Chla, Chlb Cart, and EL (figure 7).



**Figure 7:** Principal Component Analysis (PCA) of the morphological and physiological parameters from plant in relation with soil characteristics (plane representation); SL: shoot length; RL: root length; TL: total length; Chla: chlorophylla; Chlb: chlorophyll b; Cart: cartenoids; EL: electrolyte leakage; LN: leaf number; LA: leaf area; Poly: polyphenols; OM: organic matter; EC: electric conductivity; TNK: total nitrogen kjeldhal; WCR: water content root; WCS: water content shoots.

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Indeed, all these parameters were negatively related to water content of the shoot part of the plant.

For sorghum plants, a positive correlation was noted between LA and OM content (0.840) and EC (0.893). Also, chla, chlb and cart were highly and positively correlated to polyphenols content in soil (figure 7). On the contrary, water content on root part was negatively associated to TNK (-0.659), OM (0.666), Poly (-0.601). The same tendency of these parameters was reported in relation to water content in shoot part. Moreover, WCS and WCR were positively related to Electrolyte leakage. Concerning tomato, a positive correlation was reported between morphological parameters (TL, SL, and LN). Moreover, photosynthetic pigments have a tendency to decrease with the increase of soil polyphenols and organic matter content.

## Conclusions

In this study, we set out to study the fertilizing potential of a compost juice obtained from the olive industry bio-waste, within the framework of the circular economy. The likely effects of different compost/soil ratios and different extraction rates of compost juice on the plants development and productivity and on the soil characteristics were investigated.

The set of results showed that in most cases the compost juice effects on the seeds germination *in vitro* depends on the extraction factors. For the development of the plants, it was observed that the mixtures compost/soil give better results in comparison with the soil irrigated by compost juice and with control soil. Regarding the plants agro-physiologicol parameters, we found that there are no significant differences between plants amended compared to controls ones. However, for the chlorophyll pigments amounts we noted an improvement in the compost and the compost juice mediums compared to the control. Finally, we found that besides the effects on plants, the amendment with compost or irrigation with compost juice improved all the soil physicochemical characteristics.

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## **Conflict of Interest Statement**

The authors declare that they have no conflict of interest.

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