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Department of Agricultural, Food and Environmental Sciences

PRODUCTION OF AGRI-FOOD DERIVED COMPOST, SOIL AND PLANT RESPONSES FOLLOWING ITS APPLICATION

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This thesis is dedicated to my parents who sacrificed all their lives and loved their children unconditionally.

To my father, Seyed Reza Hoseini, who gave me the vision to trust my capabilities and taught me to work hard.

To my mother, Kobra Nouri, who cherished me with love and taught me the way of living and being independent.

Every success I achieved is because of them.

May Allah bless their souls.

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ABSTRACT

This research was about the production of agri-food composts derived from coffee husk (CH) and brewers' spent grain (BSG) in proportion 2:1 (Compost1), CH and cow manure (CM) in proportion 4:1 (Compost2), and a mixture of CH, BSG, and CM in proportion 5:3:2 (Compost3), and also determine the effects of temperature conditions on their properties. According to the results, we suggested the agri-food compost derived from coffee husk and brewers' spent grain in warm condition. Subsequently compost was added to the wheat and Cannabis Sativa L. field and it is well-documented that the achievement of a high yield of wheat and hemp depend on retaining the nutrients in the plant root environment, which is important for intended crop production. Due to the high cost and environmental side effects of chemical fertilizer, there is a growing tendency to utilize organic fertilizer like compost in agriculture. No main effects of composts were observed for fatty acids. Soil physic-chemical features were improved by the application of composts, especially the compost derived from coffee husk and brewers' spent grain. Moreover, this compost had positive influence on the microbial enzyme activate and biomass, probably due to the composition and higher input of carbon organic into the soil. As a result, it can be suggested to use this compost to reduce environmental pollution, improve soil fertility, and achieve high production of wheat crop.

Key Words: Food By-product, Compost, Wheat, Hemp.

SOMARIO

Questa ricerca ha riguardato la produzione di compost agroalimentari derivati dalla buccia del caffè (CH) e dal prodotto di scarto di un birrificio artigianale (BSG) in proporzione 2:1 (Compost1), CH e letame (CM) in proporzione 4:1 (Compost2) e una miscela di CH, BSG e CM in proporzione 5:3:2 (Compost3) e la valutazione degli effetti della temperatura esterna sul processo di compostaggio e sulle proprietà dei compost ottenuti. I risultati ottenuti suggeriscono che il compost di qualità migliore è quello ottenuto miscelando CH e BSG (Compost1) in condizioni di temperatura esterna più elevata. I diversi tipi di compost sono stati successivamente aggiunti a delle parcelle di suolo (schema a blocchi randomizzati) coltivate con frumento e Cannabis Sativa L. in considerazione del fatto che il mantenimento di un'elevata concentrazione di nutrienti nell'ambiente radicale della pianta può portare a rese più elevate delle colture considerate e che vi è una crescente tendenza in agricoltura a utilizzare fertilizzanti organici come il compost per ridurre l'utilizzo di quelli chimici. Non si sono osservati effetti dell'applicazione del compost sulla componente degli acidi grassi mentre le caratteristiche fisico-chimiche del suolo oggetto di studio sono risultate migliorate, in particolare dopo l'applicazione del compost derivato da CH e BSG che ha avuto effetti positivi sia sul contenuto di C-biomassa del suolo che sull'attività enzimatica del suolo, probabilmente in relazione alla sua composizione e al maggiore apporto di carbonio organico. Alla luce dei risultati ottenuti, si può suggerire l'utilizzo di questo compost (Compost1) per contribuire alla riduzione dell'inquinamento ambientale, migliorare la fertilità del suolo e ottenere un'elevata produzione di frumento.

Parole chiave: sottoprodotto alimentare, compost, frumento, canapa.

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CHAPTER1

Production of agri-food derived compost, soil and plant responses following its application.

ABSTRACT

This research was about the production of agri-food composts derived from coffee husk (CH) and brewers' spent grain (BSG) in proportion 2:1 (Compost1), CH and cow manure (CM) in proportion 4:1 (Compost2), and a mixture of CH, BSG, and CM in proportion 5:3:2 (Compost3), and also determine the effects of temperature conditions on their properties. According to the results, we suggested the agri-food compost derived from coffee husk and brewers' spent grain in warm condition. Subsequently compost was added to the wheat and *Cannabis Sativa L.* field and it is well-documented that the achievement of a high yield of wheat and hemp depend on retaining the nutrients in the plant root environment, which is important for intended crop production. Due to the high cost and environmental side effects of chemical fertilizer, there is a growing tendency to utilize organic fertilizer like compost in agriculture. No main effects of composts were observed for fatty acids. Soil physic-chemical features were improved by the application of composts, especially the compost derived from coffee husk and brewers' spent grain. Moreover, this compost had positive influence on the microbial enzyme activate and biomass, probably due to the composition and higher input of carbon organic into the soil. As a result, it can be suggested to use this compost to reduce environmental pollution, improve soil fertility, and achieve high production of wheat crop.

Key Words: Food By-product, Compost, Wheat, Hemp.

1. INTRODUCTION

1.1. Composting

Composting is “the controlled aerobic biological decomposition of organic matter into a stable, humus-like product. It is essentially the same process as natural decomposition except that it is enhanced and accelerated by mixing organic wastes with other ingredients to optimize microbial growth [1][2]. Therefore, such waste management system turns a waste into a resource by creating a recycled product made up of stabilized organic matter, carbon rich, and free of most pathogens and weed seeds. It constitutes series of techniques towards organic waste treatment that is in total agreement with sustainable agriculture [1]. The composting process is conducted by a series of different microorganisms aiming to degrade organic matter. Therefore, the monitoring of these microorganisms in succession is key for effective management of the composting process, rate of biodegradation, and compost quality given that the appearance of some microorganisms reflects the maturity of the compost [1].

Compost is from classical Latin *compositus* root. The ancient Akkadian Empire in the Mesopotamian Valley referred to the use of manure in agriculture on clay tablets about 4300 years ago [3]. There is evidence that Romans, Greeks and the tribes of Israel knew about compost too. During the Neolithic period, human beings for the first time began to live in urban settlements, changing their habits from essentially hunters and gatherers to farmers and breeders. Since the establishment of these settlements, waste pits became commonly used. The first waste pits made out of stone and built outside the houses were found in Sumerian cities about 6000 years ago. In these pits, organic urban waste was stored for eventual application on agricultural fields [3].

1.2. Raw materials

The topic of this study was to produce efficient composts under two different sets of temperature conditions. The experiment was done in the experimental greenhouse of Università Politecnica delle Marche (UnivPM), Ancona, Italy. Raw materials of experiment were coffee husk (CH), brewer's spent grain (BSG), and cow manure (CM).

Fig. 1: raw materials from left to right (coffee husk, Brewers' spent grain and manure)

Coffee Husk: In the processing from the coffee fruit to the exposed bean, called green coffee beans, there are two methods primarily used: the dry and the wet method. During the different processes to obtain the beans, large amounts of by-products are generated as approximately 50% of the coffee fruit is not used for the production of green coffee. The by-products are mainly used as fertilizers and animals' food. The main by-product from the dry method is the coffee husk which is composed of the dried skin, pulp and parchment. Of each ton harvest coffee fruit, 0.18 ton of coffee husk are produced. Component analysis showed that coffee husk is rich in organic matter (cellulose, hemicelluloses, pectins, and lignins), chemical nutrients such as N and K, and secondary compounds such as caffeine, tannins, and polyphenols [4][5].

Brewers' spent grain: Malted barley is the brewers' preferred grain for making beer. In its most basic form, it is barley that has been allowed germinating by soaking the grain in water. This prepares the starches to be converted into fermentable sugars. BSG is a lignocellulosic material containing about 17% cellulose, 28% non-cellulosic polysaccharides, and 28% lignin. BSG is available in large quantities throughout the year, but its main application has been limited to animal feeding or energy production [6].

Cow Manure: Manure is composed of animal feces and urine and may contain livestock bedding, additional water and wasted feed. It is a valuable fertilizer that contains a broad range of nutrients such as N, P, and K as well as micronutrients such as Cu, Mn, and Zn. Manures with added bedding are also an excellent source of organic matter which improves soil quality when applied to land. The water, nutrient and organic matter contents of manures, however, vary greatly making them more difficult to manage than synthetic fertilizers [7].

1.3. Compost making methodology

Raw materials were mixed in three piles with the ratio of 2:1 (CH+ BSG), 4:1(CH+ CM), and 5:3:2 (CH+ BSG+ CM). Every plastic pile had around one cubic meter volume with some installed pipes in the bottom for aeration. After mixing 300 kg of materials in every pile, they were covered by plastic bags to avoid of humidity and heat losing. Compost was made by aerated static pile method, and aeration was done by compressor every ten days for 15 minutes, for better air movement in the piles wood chips were put in several layers between materials to increase the porosity and penetration capability. The temperature of piles measured every day and tried to keep the humidity around 60%. All the conditions for two composts were kept the same, except temperature differences.

Fig. 2: Installed compost piles in greenhouse and aeration pipe

Only coffee husk compost was made before experiment and due to the high pH (more than 9), decided to make a mixture with different organic waste to make a balance in pH. *Salmonella* test on all of compost samples and raw materials was negative. Sampling was done every 30 days of composting process, and the samples immediately kept in -20ºC fridge.

1.4. Field experiment

The field preparation and soil descriptions for T0 (time zero) were performed in Agugliano, Marche region (Coordinates: 43°33′N 13°23′E; Elevation: 203 m), Italy, and around 10 kg of compost distributed in any plot with 18 m^2. Wheat and hemp seeds cultivated in the arranged field by Completely Randomized Design (CRD) method separately in three replications. The total dimension of every field was 216 m^2, with margin and corridor was 513 m^2. Totally, 12 blocks with 18 m^2 dimensions were considered.

The soil morphological descriptions were carried out three times i) before compost distribution and cultivation, time zero (T0), ii) after compost distribution and cultivation (T1), and iii) after harvesting, (Th). Soil samples were collected within 50 cm-depth from three horizons (Ap1, Ap2, and Ap3), which were air-dried at room temperature, ground, and sieved at 2 mm, and stored at 4 °C for further soil analysis.

Fig. 3: soil sampling by horizons.

We scheduled to obtain four separated papers from this Ph.D. thesis work.

1. Marziyeh Hoseini, Stefania Cocco, Cristiano Casucci, Valeria Cardelli, Giuseppe Corti, Coffee by-products derived resources. A review, 2021, Journal of Biomass and Bioenergy 148, 1-10. https://doi.org/10.1016/j.biombioe.2021.106009

2. Producing Agri-Food Derived Composts at Different Temperature Conditions (under preparation)

3. Agri-food derived composts improve the soil physicochemical features and wheat yield (under preparation)

4. Effect of Coffee-husk based compost on yield and lipid fraction of *Cannabis sativa L.* (under preparation)

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CHAPTER2

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BIOMASS & BIOENERGY

Coffee by-products derived resources. A review

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ABSTRACT

Coffee is the most common beverage and there are two main different methods to process coffee cherries. Coffee husk is the main by-product of coffee processing by dry method and is available in large quantities throughout the years, but its main application has been limited to animal feeding or energy production. Most of the coffee husk is disposed in landfills or arable land, usually with no care of its fate and changes to the source of pollution, especially in developing countries. Coffee husk can have several re-uses, but it is important to have environment friendly methods to change it into useable material or material to be recycled in nature because of its important content of organic matter, chemical nutrients, and secondary compounds. The aim of this review is to recollect the amounts and uses of the coffee industry by-products, giving emphasis to its transformation into compost because of their large content of nutrients and the need to introduce high valuable organics into the soil.

Key Words: Coffee-Husk, Compost, Win-win Solution, Soil Fertility, Waste-Management.

1. INTRODUCTION

Demand for coffee in the last 150 years was more than in the past not only because of the increased population and urban development but also because coffee has become one of the most consumed beverages in the world [1]. Nowadays coffee trade is economically at the second place of the world rank after petroleum [2]. According to the International Coffee Organization (ICO), in the 2016/2017 crop year the world coffee production was ≈152 million of 60-kg bags (for a total of ≈9 million tons), for an economic value of ≈90 billion dollars [3]. The world population of ≈7.6 billion in 2017 is expected to reach 8.6 billion in 2030, and 9.8 billion in 2050, with one third of the population concentrate in urban areas [4]. Because of this, also production and consumption of coffee are expected to increase concerning the actual levels.

Even though the first plantation of coffee was done in Yemen by Arab people in 13th century with seeds transferred from Ethiopia [5], nowadays Brazil, Vietnam, Indonesia, Colombia, Ethiopia, India, and Mexico are the major producers of coffee, with Brazil producing half of the world production [6]. The coffee plant belongs to the Rubiaceae family and, among the numerous species present in nature, currently only *Coffea arabica L.* (known as Arabica) and *Coffea canephora L.* (known as Robusta) have an important economic value [3]. Coffee processing industry produces huge amount of by-products since from 30 to 50% of coffee fruit weight is waste [6]. Due to the high amount of coffee seeds production, several re-using solutions have been proposed, but a win-win solution to manage the considerable amount of coffee husk is needed [7].

Because of this, the aims of this review were to report of 1) different processing methods of coffee cherries, introduce the main by-products of coffee processing, and emphasize on the needed detoxification of coffee husk obtained by dry method; and 2) the several usages of coffee husk in industry and agriculture. All the themes referring to processes and uses were discussed also by synthesizing advantages and disadvantages reported in the literature.

2. COFFEE SEED ANATOMY

The economic lifespan of coffee tree is maximum of 30 years. The shrub is perennial and can reach a height of 10 m. The first flowers are produced when plants are 3–4 years old, and are creamy white and sweetly scented, appearing in clusters in the axis of the leaves. Two fertilized ovules of coffee flower ovary start to grow up two months after fertilization. Adequate water supply is important to break the dormancy in the third month. The ovary size increases, and the embryotic sac grows and fills with endosperm. Till the end of the fifth month after fertilization, weight and volume of fruits increase significantly. Between sixth and eighth months after fertilization, the fruit reach to maturity, represented by an oval drupe of 18 mm in length and 10–15 mm in diameter. The ripe fruit has bright red or yellow color, and it is also called "cherry" [8]. [9]. [10].

The coffee fruit has four different layers protecting the seed that must be removed to collect the two beans forming the seed. The outer layer is the skin (epicarp or exocarp), with waxy substance and red color. The second layer is the pulp (mesocarp), that is a slim layer of pectinaceous materials. The third layer is the parchment (endocarp), with polysaccharide covering. The last layer, sticking to the seed, is named silverskin or chaff [11]. Fig. 1.

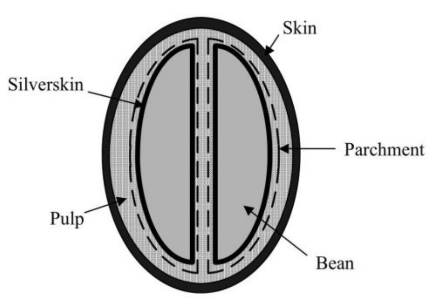

Fig. 1: Schematic picture of coffee fruit from [11].

3. COFFEE PROCESSING METHODS AND BY-PRODUCTS

To keep the quality of the seeds and preserve them from pathogens, they must be extracted from the four layers forming the other part of the fruit. The industrial process of coffee seeds preparation can be made following two main methods: dry and wet methods. The dry method is the traditional one, but it is also the simplest and environment-friendly since it produces less amount of solid and liquid by-product. Following this method, after having selected and cleaned the cherries, these are sun-dried with frequent turning to obtain a relatively homogenous drying. Thus, the outer layers of the cherries are removed by a hulling machine, and the coffee beans are roasted and bagged. With the wet method (or washed method) more equipment and water are needed in comparison to the dry method. With this method, the quality of the coffee beans is higher than with the dry method because the bean components are better preserved, and the number of defective seeds is less. Following this method, after the cherries are sorted and cleaned, the pulp is separated by a squeezing machine and the seeds are roasted [3]. Every ton of fresh coffee cherry produces 0.12–0.18 tons of coffee husk with the dry method and 0.5 tons of coffee pulp with the wet method [12]. Even though the quality of the obtained coffee seeds depends on the processing, the seeds produced with both methods have their own market since the beverage obtained with seeds submitted to the dry process is less acidic than those obtained with the wet method.

Each step of the coffee processing from coffee fruit to a cup of coffee, including separation of coffee seeds, roasting, packing, and making a drink, produces by-products.

3.1. Spent coffee ground.

This byproduct is the result of coffee brewing from coffee making such as homemade coffee and coffee machines or indirect way like instant coffee and beverage factories. It has a dark brown color, coarse texture, and high moisture [11]. The content of lipids in the fresh spent coffee ground is around 2% on a weight basis, with palmitic and linoleic acids covering 35% of the total extractable oil [13]. This by-product is also rich of vitamin E since, in classic espresso coffee and in coffee machines coffee, only 1 and 5%, respectively, of this vitamin is extracted. Therefore, coffee ground cakes can be used as a source of liposoluble antioxidant vitamins [14].

3.2. Defective and premature coffee beans.

Both coffee harvesting and roasting process produce two types of by-products, respectively: immature and defective beans. These beans must be removed from the mass of the valuable beans since they might decrease the quality of the final products [15]. In fact, beans from defective cherries have higher amounts of free amino-acids and phenols and contain fewer sugars than normal beans because they did not reach proper maturity [16]. As an alternative use for these low-grade coffee beans, Alves et al. [11] suggested to use them for the extraction of chlorogenic acid or caffeine, for their potential applications in the food and pharmaceutical sectors.

3.3. Silverskin.

Coffee silverskin is a so thin layer sticking to the coffee seeds that detaches only during coffee roasting [17]. Coffee silverskin has antioxidant activity, because of the presence of melanoidins [18], prebiotic activity [19], and contains dietary fiber [20]. These valuable components have encouraged studies on the production of body weight control beverages, diet bread, biscuits [17]. [21], and cosmetic products [22].

3.4. Coffee pulp.

Coffee pulp represents ≈35% of the coffee fruit [11] and is a by-product of the wet method in coffee hulling process. In the coffee pulp, the content of phenolic acids is slightly higher than in coffee husk, 1.5% vs. 1.2%. Among the phenolic acids comprising the coffee pulp, flavan-3-ols, hydroxycinnamic acids, flavonols, and anthocyanidins are the most represented [23].

3.5. Coffee husk.

Coffee husk is the main by-product of the dry method and is formed by all the layers at once, including dried skin, pulp, mucilage, and the parchment [24]. When the coffee cherry is dried, ≈12–18% of the dried fruit weight is coffee husk [5]. In general, amount of components and indexes of coffee husk vary according to the coffee species, the geographical origin of the cherries, and the chosen method of processing [25], which explains the differences in the composition reported by many authors [26]. [2]. [27]. [11]. [6]. [15]. In Table 1 we reviewed and synthesized composition and physicochemical properties of coffee husk. However, Alves et al. [11] reported completely different amounts of lignocellulosic polymers, with 24.5% cellulose, 29.7% hemicellulose, and 23.7% lignin. It is desirable that in future studies coffee husks be classified according to their properties.

Table 1: Main composition and physicochemical properties of coffee husk.

	Value	References
Organic component (g kg^{-1})		
Carbohydrates	580-850	[27]. [6]. [11]
Cellulose	430	[2]. [27]
Hemicellulose	70	[2]. [27]
Lipids	5-30	[27]. [6]. [11]
Total fiber	240	[2]
Ash	25-62	[28]. [11]
Protein	80-110	[27]. [6]. [11]
Caffeine	10	[2]. [6]. [11]
Tannins	50	[2]. [6]. [11]
Chlorogenic acid	25	[2]
Pectic substance	16	[2]
Lignins	90	[2]
Sugar content (g kg^{-1})		

Reducing sugar	120	[29]
Total sugar	140	[29]
Sucrose	20	[29]
Physicochemical parameters		
pH (1:10)	5.35-6.63	[30]. [31]
EC (dS m^{-1})	2.24-3.1	[30]. [31]
Organic carbon (g kg^{-1})	545	[30]
Organic matter (g kg^{-1})	815	[31]
C/N ratio	29.8-40	[30]. [31]

3.5.1. Macro- and micro-nutrients.

Coffee husk is rich of macro and micro-nutrients, with considerable amount of N (1720-1830 mg kg^{-1}), P (80 mg kg^{-1}), K (20 600 mg kg^{-1}), and others (Table 2). Positively enough, it contains small amounts of Na.

Table 2: Elemental content of coffee husk. Values are expressed on a dry matter basis.

Element (mg kg^{-1})	Coffee Husk	References
Total content of inorganic elements	5000-30 000	[32]
N	1720-1830	[32]. [30]
P	80	[32]
K	20 600	[32]
Ca	2210	[32]
Mg	790	[32]
Fe	260	[32]
Cu	20	[32]
Mn	60	[32]
Zn	10	[32]
B	91.4	[33]
S	1100	[33]
Se	0.19	[32]
Na	40	[32]

3.5.2. Amino acids.

The coffee husk contains a protein content ranging from 8 to 11% on a dry matter basis [34], with a relatively high content of amino acids such as glutamic acid (7.7% of the total protein content) and aspartic acid (7.1%) [34]. Glutamic acid is responsible for the transport of glutamine and other amino acids through the blood, and its presence decreases the need to consume sugar and alcoholic beverages. The aspartic acid is involved in the metabolism of DNA and RNA, but also in protecting the liver and boosting the immune system. So, coffee industry by-products are a source of amino acids that could be evaluated as dietary phytochemicals useful for human beings. Dietary supplements and/or food fortification based on coffee by-product production may be feasible too [35]. [36]. Table 3 shows a comprehensive view of the main amino acids present in the coffee husk.

Table 3: Content of protein and of the main amino acids in coffee husk and pulp. From [27]. [34]

Protein content	8-11%, on a dry matter content
Amino acid	% with respect to the total protein content
Glutamic acid	7.7
Aspartic acid	7.1
Leucine	4.7
Glycine	4.2
Proline	3.7
Valine	3.7
Alanine	3.5
Lysine	3.4
Serine	3.3
Isoleucine	3.3
Threonine	3.1
Phenylalanine	3.0
Arginine	2.8
Histidine	2.5
Tyrosine	1.9
Methionine	0.3

Cystine	0.3

3.5.3. Volatile oils.

Al-Yousef and Amina [37], working on coffee husk from *Coffea arabica L.*, reported of the content of a volatile oil made of at least 55 molecules. As reported in Table 4, the main chemical compositions of volatile oil in the essential oil of coffee husk, as determined by gas chromatography-mass spectroscopy (GC-MS), is mainly represented by butylatedhydroxy (65.83%), with much smaller content of 1,2-benzenedicarboxilic acid (7.28%), phenylethyl alcohol and octanoic acid (1.69% each), and 2,3-isopropylidene-6-decoxyhexo (1.63%). According to the mass spectra observation, 30% and 40% of the compounds present in the oil are hydrocarbon and oxygenated constituents, respectively, while aromatic compounds dominates. Volatile components showed antibacterial, antifungal, and antioxidant potentiality that are helpful in the treatment of infection diseases. In Table 4, the total time required to analyse a single sample was 58 min and the components were identified on the basic of GC-MS retention time. M^+ represents molecular ions, which are important for determining the molecular weight by GC-MS.

Table 4: Content of the main volatile oils in the essential oil of coffee husks. From [37].

	Required time (min)	Area (%)	M^+ (g)
Butylated hydroxytoluene	24.2	65.83	220.18
1,2-benzenedicarboxylic acid	31.92	7.28	278.34
Phenylethyl alcohol	13.66	1.69	122.09
Octanoic acid	15.76	1.69	144.21
2,3-isopropylidene-6-deoxyhexo	26.12	1.63	220
Decane, 1,1’-oxybis-	47.9	1.59	298.54
Nonanoic acid	18.4	1.58	158.16
1,2-benzenedicarboxylic acid	33.78	1.37	278.35
Beta-d-arabino-2-hexulopyran	24.66	1.17	234.00

Oxalic acid, 2-ethylhexyl tetr	44.78	1.11	398.61
Hexatriacontane	49.36	1.00	506.97

M^+ is the molecular ion, expressed as the ratio between mas and charge number of ions (M/Z); since Z is almost always 1 in GC-MS, M^+ is mainly generally the mass (g) of the ionic molecule.

Al-Yousef and Amina [37], evaluated the volatile oil and total alcohol extract of coffee husk for their antimicrobial activity with respect to three well-known antibiotics like ampicillin and doxycycline, used as positive control against bacteria, and nystatin, used as the control antifungal drug. In the experiment, minimum inhibitory concentration of ethanol extracts as well as volatile oil of coffee husk against drug resistant clinical strains was determined. The results are shown in Table 5. Both volatile oil and total alcohol extract of coffee husk reduced the growth of *Staphylococcus aureus*, *Escherichia coli*, *Pseudomonas aeruginosa*, and *Candida albicans* colonies by 50–104% with respect to three tested antibiotics, with the alcohol extract being more efficient than volatile oil for *Staphylococcus aureus* and *Pseudomonas aeruginosa*.

Table 5. Antimicrobial activity and minimum inhibitory concentration of essential oil and total alcohol extract of coffee husk from *Coffea arabica* L. on the growth of four infective microbes [37].

Samples	*Staphylococcus aureus*	*Escherichia coli*	*Pseudomonas aeruginosa*	*Candida albicans*
	Zone of inhibition (mm)			
Coffee husk essential oil	14.0 ± 1.3 (-66.7%)	17.0 ± 1.9 (-68.0%)	13.0 ± 2.0 (-50.0%)	15.0 ± 1.3 (-5.2%)
Total alcohol extract	22.0 ± 0.5 (-104.8%)	17.0 ± 2.5(-68.0%)	20.0 ± 1.1 (-83.3%)	14.0 ± 1.2 (-0.9%)
Ampicillin	21	-	-	-
Doxycycline	-	25	24	-
Nystatin	-	-	-	23
	Minimum inhibitory concentration (mg mL^{-1})			
Samples	*Staphylococcus aureus*	*Escherichia coli*	*Pseudomonas aeruginosa*	*Candida albicans*
Coffee husk essential oil	0.8	0.8	0.8	0.8
Total alcohol of coffee husk	3.2	>3.2	3.2	>3.2

*Values of Minimum inhibitory concentration is given in % v/v for dry oils

3.5.4. Phenolic acids.

The main phenolic acids of fresh coffee husk are caffeine, tannins, and chlorogenic acids (for quantities see Table 1), whose presence prevents various uses of fresh coffee husk because of their ecotoxicological concerns [38]. For example, fresh coffee husk is not suitable for animal feeding because of its anti-nutritional properties due to the excess of phenolic acids [39]. Thus, chlorogenic acid has phytotoxic effects able to decrease seed germination and plant growth and, because of this, it cannot be distributed in soil as soil fertilizer [40]. [38]. In addition, caffeine and tannins negatively affect aquatic organisms like algae, sea urchin, and fishes, which develop morphological and behavioral abnormalities [41].

Caffeine. The alkaloid caffeine (1,3,7-trimethylxanthine) has been found in more than 60 plant species, with the highest levels in coffee beans, tea, and cocoa. Other two alkaloids of the xanthine derivative group are theobromine (3,7-dimethylxanthine) and theophylline (1,3-dimethylxanthine) [42]. Generally, caffeine has positive effects on humans as it has chemical structure like that of adenosine, so it is well-known as adenosine receptor. Because of this, caffeine may help to be relaxed and sleep [43]. Caffeine can be also able to contrast obesity and diabetes [44], as well as Parkinson's [45] and Alzheimer's symptoms [46]. However, caffeine has negative effects on the environment as it is toxic to aquatic organisms and mammalians, and has negative effects on animals, plants, fungal and bacterial growth [47].

Tannins. Tannins are commonly found in the bark of vascular plants and, to a lesser extent, into leaves, fruit, flowers, and seeds [48]. Tannins are considered as anti-nutritional compound, and this aspect limits the use of coffee husk in animal feed [49]. Benefits of tannins for human health include antibacterial and antifungal activity [50], antimicrobial activity, being effective against parasites and some viruses [51], anti-inflammatory [52], and anti-allergy [53] activities. Tannins are also known for their low biodegradability; because of this reason they tend to remain for long time in the environment and accumulates in the food chain [54].

Chlorogenic acid. The esterification of caffeic acid with quinic acid produces chlorogenic acid, which is a soluble polyphenol [55] that plays many human health benefits, including neuronal cell death protection [56] and anticancer activity [57]. [58]. Chlorogenic acid plays positive roles also in plant functions including cell wall synthesis, wound healing, and root hair formation [59]. However, depending on its concentration, it may play a negative role especially in roots [60]. Villarino et al. [61], reported the inhibitory effect of chlorogenic acid on fungal growth, due to its role on plant defence. The content of other phenolic acids extracted from coffee husk is reported in Table 6.

Table 6. Phenolic components of coffee husk (modified from [62]).

Method	Condition/Solvent	Epicatechin	Gallic acid	Tannic acid	Protocatechuic acid	Vanillic acid
		µg of gallic acid equivalent per kg of coffee husk (dry matter)				
Ultrasonication	Ethanol	-	-	-	-	2346.7
Soxhlet	Ethyl acetate	47.6	3869.2	-	-	-
	Ethanol	-	-	3859.2	-	-
SFE*	200bar/40^{0}C	-	14.85	-	-	-
CO_2	300bar/40^{0}C	32.55	-	-	12.4	-

* SFE: supercritical fluid extraction.

Secondary metabolites in coffee husk such as caffeine and other phenolic compounds are good source of antioxidants. Table 7 shows the antioxidant capacity of aqueous extract of coffee husk evaluated following DPPH (2,2-diphenyl-1-picryl-hydrazyl-hydrate) radical sequestration method and inhibition of co-oxidation of β-carotene and linoleic acid percentage [63]. For this experiment, coffee fruits were randomly collected at four different farm locations, from two plants in the northern (husk 1 and grains 1), southern (husk 2 and grains 2), eastern (husk 3 and grains 3), western (husk 4 and grains 4) and central region (husk 5 and grains 5) of the plantation [63]. Results indicated that micro-environmental conditions present in the plantation affect the antioxidant capacity of aqueous extract of coffee husk.

For each column, means followed by different letters differed for P < 0.05, by the Tukey test.

Table 7. Antioxidant capacity of aqueous extracts of coffee husk evaluated by the free DPPH radical sequestration method and inhibition of co-oxidation of β-carotene and linoleic acid [63].

Sample	DPPH EC50* (mg mL^{-1})	Inhibition of co-oxidation of β-carotene and linoleic acid (%)
Coffee Husk 1	4.71^{f}	40.78^{bc}
Coffee Husk 2	3.57^{h}	34.88^{c}
Coffee Husk 3	4.44^{g}	43.74^{abc}
Coffee Husk 4	2.73^{j}	44.55^{abc}
Coffee Husk 5	3.44^{i}	40.80^{bc}
Coffee Grain 1	15.09^{a}	68.58^{a}
Coffee Grain 2	11.48^{b}	66.43^{ab}
Coffee Grain 3	10.44^{c}	58.22^{abc}
Coffee Grain 4	10.10^{d}	64.65^{ab}
Coffee Grain 5	7.53^{e}	68.22^{a}

* EC50 = half maximal effective concentration.

For each column, means followed by different letters differed for $P < 0.05$, by the Tukey test.

3.5.5. Lignocellulosic materials.

Cellulose, hemicellulose and lignin are the principal lignocellulosic components forming plant cell walls. Lignocellulosic compounds like phenolic acids often prevent coffee husk usage and degradations, so it is necessary to find techniques able to break down these substances [64]. [65]. According to Oliveira et al. [66], the lignin of coffee husk represents a significant resource for enabling use of coffee husk as raw material for biorefineries where lignin can be separated from other coffee husk components with a pre-treatment by diluted acid followed by soda extraction. The extracted lignin can be then wet-oxidized under aqueous and alkaline conditions, in order to produce valuable products such as low molecular weight biochemicals.

Many factors, like lignin content, crystallinity of cellulose, and particle size, limit the digestibility of hemicellulose and cellulose. Pretreatments improve the digestibility of the lignocellulosic material. Each pre-treatment has its own effect(s) on cellulose, hemicellulose, and lignin. Many thermal, acid, alkaline, and oxidative pre-treatments have been evaluated for improving biodegradability of lignocellulose substrates [64]. For example, Baˆeta et al. [67] pre-treated coffee husk by a steam explosion technique that increased the bioavailability and biodegradability of cellulose, broke down the lignocellulose structural components, and produced soluble organic compounds. This method is effective for increasing the anaerobic biodegradability too.

3.5.6. Flavorings

The most important characters of coffee as a beverage are acidity, aroma, and taste. Without acidity, the coffee is approximately tasteless [68]. Sampaio et al. [69] reported that coffee husk is a valuable by-product due to its aroma and presence of sugars that can be converted to ethanol. Table 8 shows flavor and aroma inside beverages made by different concentration of coffee husk and pineapple juice.

Table 8. The sensorial analysis of flavor, aroma and overall appearance of the beverage developed with the rinds of coffee with different concentrations and pineapple juice [63].

Sample	Flavor	Aroma	Overall impression
	Score		
F1*	2.96 ± 1.88^{b}	4.25 ± 2.06^{b}	3.75 ± 1.90^{b}
F2**	4.86 ± 2.02^{a}	5.44 ± 1.91^{a}	5.05 ± 2.01^{a}
F3***	5.48 ± 2.11^{a}	5.44 ± 1.95^{a}	5.48 ± 2.05^{a}

For each column, means followed by different letters differ for $P < 0.05$, by the Tukey test.

*100% coffee husk extract; **90% coffee husk extract+10% concentrated pineapple juice; ***80% coffee husk extract+20% concentrated pineapple juice. The evaluation was assessed by non-trained 52 judges who used a structured 9-point hedonic scale (1 = I greatly dislike, 9 = I enjoyed it very much).

According to Tables 7 and 8, aqueous extracts of coffee husk represent a promising natural source of bioactive phytochemicals, also because of their low levels of antinutrients [63]. Neves et al. [63] noticed that the beverage incorporated with concentrated pineapple juice presented the greatest acceptability, besides increasing the antioxidant capacity of the product. Thus, the formulated beverages constitute a promising alternative for the beverage market, given the meaningful content of phenolic constituents derived from coffee husk.

3.5.7. Detoxification

Phytotoxic compounds like caffeine, chlorogenic acid, and tannins (Table 1), if released into the environment from coffee waste, can have severe ecotoxicological effects on several organisms [70]. Therefore, detoxification of coffee husk from phytotoxic compounds and antinutritional factors, or at least degrading them to a plausibly safe level for reusing or recycling, is necessary. Detoxification of coffee husk with physical, chemical, or biological methods were studied by Ref. [71]. [72]. [42], while a general review of enzymatic and microbial methods to remove caffeine is reported by Ref. [73].

Some physical (percolation), chemical (alcohol extraction), or microbial (fermentation with fungi) treatment can reduce the phenolic content in coffee husk. Treatments with bacteria and/or fungi and composting are the most used treatments for coffee husk and for other coffee by-products like coffee pulp and silverskin because they are more efficient and economic for controlling huge amount of waste. High concentrations of bacteria are required for caffeine detoxification since caffeine has a toxic effect for bacteria, and 0.1% concentration of caffeine inhibits protein synthesis in bacteria and yeast [71]. [74]. However, some microorganisms can grow in presence of caffeine and survival is due to their capacity to degrade it [75]. [76]. Several studies were carried out to investigate the use of purines, including caffeine, as a source of energy for microorganism growth [77]. Although fungi growing on caffeine have been isolated, most of the studies were done with bacteria isolated from soil, mainly belonging to the Pseudomonas group, with emphasis on Pseudomonas putida [78]. Yamaoka-Yano and Mazzafera [79] used P. putida strain and, after a short incubation periods of 9 days, observed 40% reduction of caffeine. Brand et al. [71] tested biological detoxification of coffee husk by filamentous fungi (Rhizopus, Phanerochaete, and *Aspergillus spp.*) using a solid-state fermentation system in which coffee husk was used as the sole source of C and N. Rhizopus arrizus LPB-79 strain showed great results on caffeine and tannins degradation (87% and 65%, respectively), which were obtained in 6 days at pH 6.0 and at 60% moisture.

The toxicity of coffee leachate were studied in laboratory by standardized tests on aquatic organisms [80]. [7], and results showed that the half maximal effective concentration (EC50) of coffee leachate was 6.02% v/v on the bacterium Vibrio fischeri, lower for the bacterium Daphnia similis (1.5%), and even less for the microcrustacean Ceriodaphnia dubia (0.12%). The reduced EC50 values from bacteria to water fleas was explain as the result of increased exposure to ingestion. There are good studies on caffeine toxicity, but no toxicity test has been performed on leachate from coffee by-products. Furthermore, there are several paths through which coffee can enter the environment such as processing/roasting or the retail consumption, suggesting that there is a major gap in toxicity data for coffee industry by-products that requires urgent attention [7].

4. USES OF COFFEE HUSK

Coffee husk is the main coffee by-product that has been the topic of several studies in order to use it in industrial activities, to produce biofuel, as contaminants sorbent, dietary fiber, and bioactive compounds, for the extraction of enzymes, or in agriculture as animal feeding or for making compost, silage, biochar, or mushroom substrata.

4.1. INDUSTRIAL USES

4.1.1. Coffee husk in ceramic industry

Generally, coffee producing countries use coffee husk as solid fuel and this method produces huge amount of ash that has environmentally side effects. Ash obtained by coffee husk combustion (that collected from ash dumps) are rich in alkaline and alkaline-earth metals that are a candidate for replacing the scarce and expensive feldspars traditionally used as fluxing component in clay based ceramic formulation. Results shows that adding 25–40% of ashes in common clay-based ceramic formulation had the best result in ceramic quality [81].

4.1.2. Coffee husk in particleboards production

Bekalo and Reinhardt [26] and Nuamsrinuan et al. [82] studied theuse of coffee husk for partial replacement of wood (up to 50%) in the production of particleboards. The results of particle sheet from milling process passed the standard tests of mechanical properties, while swelling and water absorption did not. The coffee husk-wood board showed great promise for its use in structural and nonstructural panel products based on superior flexural and internal bond properties.

4.1.3. Flavor production

Food flavoring compounds can be produced by chemical synthesis or extracted from natural materials. Nowadays the second way is highly demanding since the obtained products are considered safer and healthier than those obtained via synthesis. Plants are acceptable source of essential oils and flavors, but their value depends on factors like weather conditions and plant diseases. Due to the presence of antinutritional factors such as caffeine and tannins, coffee husk cannot be used directly as a flavour source [83]. Instead, when coffee husk is treated by steam to remove caffeine and chlorogenic acids, it can be used for aroma production by using fungi of the genus Ceratocystis [83]. Soares et al. [83] tested coffee husk as raw material for fruits flavor production by solid state fermentation and found that different dosages of glucose can determine the production of different flavors such as banana and pineapple.

4.2. fuel production

4.2.1. Coffee husk as a solid fuel

The use of coffee husk as solid fuel is the simplest way to manage problems due to disposal or accumulation in nature, even though the production of ash is also raising concerns. In fact, the ash derived from coffee husk combustion is often the object of illegal covert disposal and the source of environmental impacts [81]. About 70% of the coffee husks produced in Kenya is used as solid fuel [84]. The coffee husk is carbonized in a kiln and then ground, coagulated, and molded in form of briquettes prior to being packed into bags. The obtained coffee charcoal briquettes have better quality than wood charcoal [29]. [68] but, as for other agricultural residues, carbonization is not the best choice to recover energy from coffee husk, as its combustion efficiency is minimal because of not exactly suitable physicochemical properties such as low bulk density, low ash melting point, and high volatile matter content [85].

4.2.2. Gasification of coffee husk

In order to find solutions to improve energy recovery from coffee husk, gasification is a possibility to increase energy recovering by producing ignitable gas through a partial incineration at elevated temperatures and moderate heating rates. The obtained gas is a mixture of CO, H_2, CH_4, CO_2, and N_2, and temperature level is key to improve gas quality [86]. In coffee producing countries, biomass energy has potential to be the most abundant sustainable renewable energy but, to reach this goal, there is the necessity to develop and sustain contemporary technologies that increase the biomass-to-energy conversion. One way can be the high temperature air/steam gasification of biomass [87]. Wilson et al. [87] studied coffee husk experimental gasification under high temperature conditions by batch facility and found positive influence of high temperature on increasing the gasification process. Experiments carried out at 4% O_2 concentration obtained the highest gasification rate (96% of the coffee husk), while 82.80% and 71.29% of the husk was gasified with gasification conditions at 2 and 3%, respectively. Miito and Banadda [86] found that the 46.6 million tons per year of coffee husk produced in Uganda, with a heating value of 18.34 MJ kg^{-1}, will address a 0.7% of the total energy consumed in the country, while protecting the environment too. The same use could be feasible in the countries where coffee is produced and processed, namely where coffee husk abounds.

4.2.3. Ethanol production from coffee husk

Coffee husk has also good potential to be used for bio-ethanol production and, as for gasification, temperature and yeast concentration are key to control the quality of the production with batch fermentation method [88][27]. The availability of cellulose, hemicellulose, and lignin in coffee husk is similar to that of other agricultural residues such as sugarcane bagasse, barley and wheat straws, rice husk, and others. However, because of the high amount of coffee husk generated, the toxic nature of coffee husk, the high percentage of fermentable sugar, and the presence of high concentration of carbohydrates, it could be a good source of raw material for bio-alcohol production [89].

4.3. Adsorption of contaminants

Cu, Cr, Cd, Ni, Hg, Pb, and Zn are the most abundant pollutants in industrial wastewater. Common methods for the removal of heavy metals from wastewaters include ion-exchange, filtration, electrochemical treatment, chemical precipitation, and adsorption. Since the activated charcoal used to remove organic and inorganic pollutants from aqueous effluents is expensive, and the activated charcoal produced from coffee husk showed high specific surface and porosity [90], it is a valid solution to reduce costs for wastewater treatment [91][92]. In addition, the adsorbed metals can easily desorb and the biomass be ready for final disposal [93]. Adsorption of Pb [94], Ni [95], cyanide [96], dye contaminants [97], and antibiotic norfloxacin [98] by coffee husk in batch mode is used to decontaminate aqueous solution. Berhe et al. [93] studied the efficiency of coffee husk to adsorb Pb(II) from industrial effluents using batch experiment and found that, at optimum adsorption conditions (pH 5 and 90 min of contact time at 200 rpm), there was the maximum adsorption efficiency of 95.14%.

4.4. Products obtained by fermentation

4.4.1. Organic acids

Coffee husk is a cheap and available substrate to produce organic acids like gibberellin and citric acid by fermentation techniques. Shankaranand and Lonsane [99] produced citric acid from coffee husk by using *Aspergillus niger* under solid state fermentation method, and by every 10 g of coffee husk they produced 1.5 g of citric acids. Machado et al. [100] evaluated the feasibility of employing coffee husk as a substrate to produce gibberellic acid in both solid-state fermentation and submerged fermentation tests.

4.4.2. Enzymes

Coffee by-products can be also used to produce enzymes like pectinase, tannase, and caffeinase by two main industrial enzyme production methods: solid-state fermentation and submerged fermentation [101]. However, Battestin and Macedo [102] produced tannase from coffee husk by using *Paecilomyces variotti,* while Murthy and Naidu [101] studied the production of amylase, protease, and xylanase by fungal organisms.

4.5. Bioactive compounds

Bioactive compounds are an extra nutritional factor typically present in small quantities in foods that have been intensively studied to evaluate their effects on health. Some of those are phenolic compounds or antibiotic molecules, while other have an anti-inflammatory, hepatoprotective, and antioxidant activity, or the ability to improve cognitive capabilities [7]. Agro-industrial by-products are good sources of bioactive compounds and have been explored as sources of natural antioxidants [103].

4.5.1. Dietary fiber

Agro-wastes are great sources of dietary fiber, which include cellulose, hemicelluloses, lignin, pectin, gums, and other polysaccharides. The soluble and insoluble dietary fibers have a wide range of health benefits, such as reduction of the risks of gastrointestinal diseases, cardiovascular diseases, and obesity [104]. The kind of coffee and the degree of roasting and extraction method influence the dietary fiber content and structural characterization of coffee husk and other coffee by-products, as determined by the Association of Official Agricultural Chemistry (AOAC) methods [105].

4.5.2. Anthocyanins

Anthocyanins are flavonoid compounds responsible for the red/blue color of many fruits and flowers. By using concentrated methanol as extractant, Prata and Oliveira [106] reported cyanidin 3-rutinoside as the dominant anthocyanin in coffee husk, so this latter could be used as a source for anthocyanin pigments as natural food colorant.

4.6. Agriculture

Uses of coffee husk in agriculture can be many, but the high content of phenolic acids and the mutagenic effect of caffeine suggest that recycling of coffee husk in agriculture should be preceded by detoxification process(es) able to decrease the concentration of these components. Information about detoxification of coffee husk is given at point 3.5.7.

4.6.1. Animal food

Agricultural industry by-products like coffee husk as livestock food is important to reduce the food competition and help to environment sustainability. Yearly coffee roasting industry produces million tons of coffee husks that contain valuable nutrients like proteins, carbohydrates, and minerals. Huge amount of production and good nutrients content make coffee husk a good material for animal food [107]. However, the idea of using coffee husk for ruminants, pigs, chickens, fishes, and rabbits was released several decades ago, but the result was not so bright and acceptable. In fact, National Dairy Board [108] reported that coffee husk is not a delicious food for cattle, which can tolerate only small portion of it because of the content of phenolic components. Fishes and poultries are even more sensitive than cattle and pigs, so the quantity of coffee husk in their diet must be small [34], unless to submit the husk to a detoxification process.

4.6.2. Mushroom bed

Coffee husk is an appropriate bed for mushroom growth because of its availability and cheap price and, due to its fragmented nature, no grinding is needed before application, but it needs disinfection. Coffee husk is a good substrate for mushroom bed, especially for *Lentinula edodes* (shiitake) and *Flammulina velutipes* species [109]. Fermentation of coffee husk by the fungus *Pleurotus ostreatus* increased protein and cellulose contents and decreased the proportion of lignin, tannins, and caffeine. Further, when fermentation of coffee husk increased, the volatile fatty acid and digestible dry matter decreased [109].

4.6.3. Biochar

Biochar is charcoal produced by pyrolysis of organic materials and can be used as soil amendment and fertilizer [110]. The biochar quality depends on the nature of the raw material and temperature. Acid soils, which abound in tropical areas, have deficiencies in plant nutrients like N, P, K, Ca, and Mg, and consequently have low crop production rates. Adding biochar reduces the soil acidity due to the alkalinity of biochar and increases the availability of nutrients and water [111]. Studies on coffee husk biochar showed that it improved soil chemical properties by increasing pH, electrical conductivity, cation exchange capacity, organic matter, total N, and available P [112]. Dume et al. [113] reported that the application of 15 tons ha^{-1} coffee husk biochar that had been produced at 500^0C temperature had positive result on soil fertility and yield. Deal et al. [114] compared the performance of biochar in five different feedstocks (coffee husk, maize cobs, eucalyptus wood, groundnut shells, and rice husks) in the humid tropics. Results showed that biochar from coffee husk were the most productive in the maize field. The soil pH in tropical area are so acidic (pH=4.7) and pH increasing because of soluble coffee husk biochar improve soil quality and efficiency for crop production.

4.6.4. Silage and composting

Silage is the direct usage of organic residues on soil surface without any treatment, while composting is the biological decomposition of organic waste promoted by bacteria, fungi, worms, and other organisms under controlled aerobic conditions to obtaine~~d~~ a partially decayed organic matter [115]. The chemical composition of coffee husk in terms on nutritive elements like N, K, P, and others (Table 2) makes it suitable to be used as amendment in agricultural soils.

Silage. Coffee husk silage can be a good option for K depleted soils, but there is the risk of phytotoxic production [116]. Application of raw coffee husk in the field inhibits the plants, specially the roots, while anaerobic decomposition increases the emission of greenhouse gases [30]. It was observed that addition of coffee husk on soil provided an increase in dry matter content, but also decreased the buffering capacity responsible for maintaining soil pH [15]. However, coffee husk spread at the soil surface may decrease the soil erosion, temperature, and evapotranspiration. So, notwithstanding the problems due to its phytotoxic activity, coffee husk can help in land reclamation [116].

Composting. One of the most important problem in coffee industry is by-products accumulation and, subsequently, economic and environmental costs for their management due to their potential contamination effect caused by the leaching of phenolic compounds. In fact, notwithstanding the many different uses to which coffee husk can be addressed, in coffee producer countries every year huge amounts of coffee husk are produced and, especially in developing countries, much of this husk is released in the land without any pre-treatment. Instead, phenolic acids content and mutagenic effects of caffeine require to treat coffee husk before land distribution to reduce its environmental concern. As reported at point 3.5.7, there are several ways to remove inhibitors from coffee husk, but composting is the most affordable, environmentally friendly, and efficient system. Because of this, different investigations aimed to improve waste management and ecosystem sustainability have been done on coffee husk so to transform a disposal problem into a valuable product for agriculture. Composting of coffee husk with other organic materials or alone is one of the best ways to profitably manage coffee husk since the process has capacity to solve management problems like mass accumulation and detoxification. Composting by oxygen-driven biological methods allows easily recycling great amounts of agricultural by-products and producing high-quality fertilizers [117][30]. Coffee husk has characteristics that make it suitable to be composted; for instance, it has a C/N ration around 30 and is rich in lignocelluloses materials, which makes it an ideal substrate for microbial processes [2]. Inoculation of lignocellulosic waste materials with lignin-degrading microorganisms accelerates the composting process and improves compost quality and the humification process [118].

Dzung et al. [119] studied coffee husk supplemented with cow manure and lime. The mixture was composted for 3 months and then was supplemented with 0.1% (w/w) effective microorganisms like N_2-fixing *Azotobacter* sp. and *Bacillus megaterium*; the authors found that the quality of the obtained compost was better than some bio-organic fertilizers present on the agriculture market. This compost was applied on coffee field and the results showed that soil fertility, nutrient content in the coffee leaves, and the growth of the coffee plants were improved in comparison with the control. Sekhar et al. [120] applied different dosages of coffee husk compost with NPK fertilizers in various amounts in the paddy field and found that applications of 4 ton ha^{-1} of coffee husk compost plus 80 kg ha^{-1} N, 60 kg ha^{-1} P, and 50 kg ha^{-1} K gave the highest grain and straw yield production. Kassa and Workayehu [31] evaluated the quality of composts comparing the quality of only coffee husk compost with mixtures made of coffee husk+cow dung, coffee husk+*Millettia ferruginea*, coffee husk+cow dung+*Millettia ferruginea*, and coffee husk+effective microorganism, and concluded that the mixtures coffee husk+*Millettia ferruginea* and coffee husk+cow dung+*Millettia ferruginea* gave the highest quality composts. In the coffee husk composting experiments run by Bidappa [121][108] and Tuan [122], as we may deduce from the fact that the use of these composts improved soil fertility and crop yield, a strong reduction of phenolic compounds was obtained. Shemekite et al. [30] used cow dung and green wastes as co-substrates in the composting of coffee husk and monitored the physicochemical changes and the microbial community dynamics during the composting process. While at the beginning of the process the microbial communities of all the compost piles differed, they were similar at the end, as shown by DGGE fingerprints and microarray analysis.

Improving soil fertility and plant growth is one of the benefits coming from compost application in agriculture. Other helpful impacts are the decrease of soil erosion and evapotranspiration, which may contribute to land reclamation. Thus, since composting process disinfects organic wastes from pathogens and weed seeds and stabilized C, N, and other nutrients in the organic fraction, applying compost to the field can help to maintain or increase the soil organic matter content, biological activity, and porosity, so helping water, air, and plant roots to penetrate easily the soil [123][117].

4.7. Resuming of Coffee Husk Applications

Table 9 shows a comprehensive view of the possible uses of coffee husk obtained by processing coffee cherries by dry method.

Table 9. Possible uses of coffee husk in industrial, fuel, agri-food, and agriculture activities.

Application	Reference
Industrial use	
Ceramic	[81]
Particleboard	[26]. [82]
Flavor extraction	[83]
Fuel	
Solid fuel	[84]. [85]
Gasification	[86]. [87]
Ethanol production	[88]. [89]. [27]
Contaminants adsorption	
Lead (Pb)	[94]. [93]
Nickel (Ni) cyanide	[95]. [96]
dye contaminants	[97]
norfloxacin	[98]
Fermented products	
Organic acid	[99]. [100]
Enzymes	[102]

Bioactive compounds	
Dietary fiber	[104]
Anthocyanin	[106]
Agriculture	
Animal food	[34]
Mushroom bed	[109]
Biochar	[112]
Silage	[116]
Compost	[30]. [120]. [121]. [122]

5. CONCLUSIONS

Coffee consumption in the world increases every year and the same happens for its by-products. Coffee husk is the main by-product of coffee roasting process by dry method and is one of the most abundant by-products that are spread in the land, giving rise to some environmental concerns. Nonetheless, coffee husk components make this material suitable to be used in several ways in many industrial, fuel, agri-food, and agriculture activities. Because of its high content of phenolic compounds, the use of coffee husk may require detoxification, and many systems have been identified to reduce the toxic effect of coffee husk; all these systems are reported in this review. The use of coffee husk as direct or indirect fuel is one of the most practiced way to recycle it but, because of its content in nutritive elements, the use in agriculture should be promoted, especially in acid soils, possibly after composting instead to be directly used as soil silage. However, the lack of local application and performing of the scientific results obtained at a global scale is a challenge that should be the topic of future studies in order to improve recycling of these valuable materials and increase soil fertility.

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CHAPTER3

Producing Agri-Food Derived Composts at Different Temperature Conditions

ABSTRACT

The composting process has been attracted to the tendency of researchers since it is an environmental-friendly, cost-effective, and efficient process. The lack of optimum environmental conditions during the process may lead to producing unhealthy and immature composts, thus we choose identifying the influence of temperature conditions variation on the composting. Therefore, this work was focused on producing agri-food composts derived from coffee husk (CH) and brewers' spent grain (BSG) in proportion 2:1 (C1), CH and cow manure (CM) in proportion 4:1 (C2), and a mixture of CH, BSG, and CM in proportion 5:3:2 (C3), and also determine the effects of temperature changes on their properties. The "warm" composting led to a rise in temperature much faster than "cold", providing an opportunity to the composts, especially C1, for being matured in a faster manner. The pH remarkably increased up to 60th day for C1 (both temperature conditions), however those of C2 and C3 (warm) remained in neutral-phase values or sequentially decreased (cold), and on the final day, it was decreased. The C/N ratio in "warm" condition was lower than in "cold". Throughout three months, the C/N ratio decreased from 13.5 to 10.6 for treatment C1 followed by C3 and C2 (9.7 and 9.4) for warm condition. Microbial biomass carbon and enzymatic activity in this work indicated an overall increment for both temperature conditions from the 0th to 30th day and a decrement from the 30th to 60th day of composting. The nutrient and trace elements have a high variation in the different composting conditions of the year. The highest GI % was observed in C1 (77%) followed by C3 and C2 (70 and 66%) in the cold conditions. Of the parameters studied, pH indicated the closest

relationship with GI%. Consequently, we suggested the agri-food compost derived from coffee husk and brewers' spent grain in warm condition.

Key Words*:* Maturity; Compost; Temperature Comparing; Coffee Husk.

1. INTRODUCTION

Composting is a bio-transformation process in which the microbial activities lead to pathogen-free, durable products for improving the fertility of the soil [1]. The main section of the waste produced is compostable (~55%), suggesting an opportunity to take a look at composting as a promising approach for the use of natural waste ([2][3]. Therefore, composting has been attracted to the tendency of researchers since it is an environmental-friendly, cost-effective, and efficient process [4]. It consists of three phases: *i*) a mesophilic stage or primary decomposition; *ii*) a thermophilic stage of severe microbial degradation; *iii*) a maturation stage that lasts for a couple of months [5]. During the three phases, microbes use accessible organic resources to support their reproduction and energy requirements [6] and overcome each other depending on the environmental and nutritional situations occurring at each phase. Because of the recent challenge to produce compost faster, a solution is augmenting cow dung or effective microbial communities to wastes [7]. Such an amendment can decrease the time taken for waste decay and enhance the compost stability. Thus, tracking of microbial activity established in the composts is an important tool for efficient management of natural wastes in composting [8].

As a natural waste, cow manure consists of a large amount of K, P, and N thereby has become an environmental challenge in our surroundings due to improper use [9]. However, aerobic composting can stabilize organic materials in the waste and keep down their detrimental potential through microbial communities [10]. As a by-product of the brewing industry that makes up 80% of brewing waste, brewer's spent grains can be utilized to fertilize the soil [11].

The compost derived from brewer's spent grains can admiringly compete with chemical fertilizers in terms of plant/crop yield in the field [11]. Coffee husk consists of tannin and caffeine, which can make it slow degradation and toxic, causing the waste disposal challenge. Of course, coffee husk is rich in lignocellulose, which makes it a desired substrate for microbial communities [12]. Besides, this improves the biological and physic-chemical properties of soil, enhances soil organic matter, and decreases the depletion of natural sources [12]. Consequently, the composting gives a promising opportunity for converting natural wastes into nutrient sources in the farming.

For well-organized composting, an optimum aeration rate (0.006‑0.3 L air kg^{-1}), C/N ratio (25‑30), moisture content (40‑60%), and suitable bulking agents (to make possible better air circulation) have been known as important indicators [13]. In full scale application the role of aeration turns to be fundamental, because it should ensure: i) the oxygen supply for the microbial activity; ii) the control of temperature iii) the removal of the excess moisture. The required airflow for temperature and moisture control is usually much higher than that needed for the biological oxidation of the organic substrate. The airflow rate is depending on the characteristics of the organic substrate, and so as the aeration mode [14].

Moreover, the temperature achieved in the compost piles influences remarkably its oxidation rate [1]. In the course of compost production, the environmental temperature affects the temperature inside the waste mass, leading to better waste humification [15]. Thus, higher temperature condition facilitated the microbial activities and waste decomposition caused more heat inside the waste piles [16], whereas less heat prevents the piles from establishing the most favorable composting temperature [16]. The lack of such optimum temperature may lead to producing unhealthy and immature composts. As a result, we require identifying the influence of temperature on the composting process.

Being aware of this, we compared the efficiency of three types of composts produced under two temperature conditions. The hypotheses of this research were: i) composts quality differs because of the different temperature conditions during composting; ii) coffee husk-derived compost obtained at the highest temperature has a better quality in comparison to other composts.

The current study revealed the influence of different temperature conditions on the composting process, and in producing valuable composts.

2. MATERIAL AND METHODS

2.1. Study area

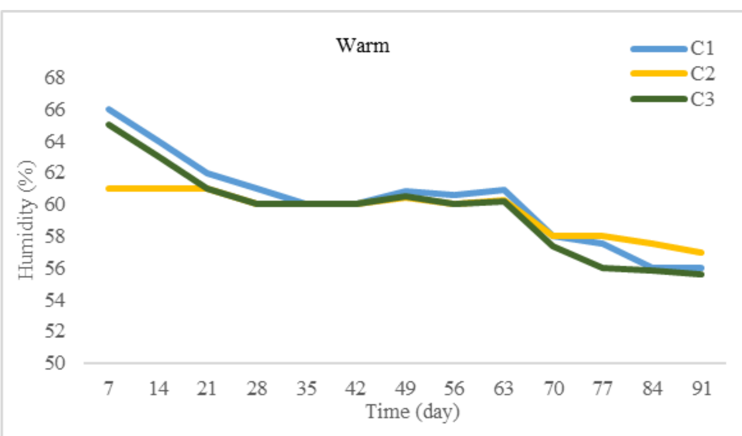
The experiment was accomplished in the experimental greenhouse of Università Politecnica delle Marche, Ancona, Italy under two different temperature conditions.

2.2. Composting environmental conditions

The composting process was run under two different temperature conditions, “warm” and “cold”. In both cases, we selected to change temperature day by day to mimic the situation of an outdoor composting period made in warm or cold situations, respectively. In Table 1 the environmental temperature recorded for each day of composting is reported. Except for temperature, all the other composting conditions such as dimension of the piles, raw materials weight and volume, humidity, and oxygen input for two composts were kept the same. Humidity of compost piles was maintained at ~60%, even though during the last two weeks it drop slightly lower than 60% due to the increasing environmental temperatures (Fig. 1).

Table 1. Greenhouse temperature during composting (ºC)

”warm” condition													
Week	**Week 1**	**Week 2**	**Week 3**	**Week 4**	**Week 5**	**Week 6**	**Week 7**	**Week 8**	**Week 9**	**Week 10**	**Week 11**	**Week 12**	**Week 13**
1st day of week	16,82	16,36	15,57	17,60	16,23	15,04	15,28	19,58	17,66	15,07	27,99	32,35	27,78
2nd day of week	17,49	17,98	16,68	15,33	14,96	13,83	14,05	18,60	18,00	27,28	27,98	29,81	30,27
3rd day of week	17,03	16,48	18,26	16,90	19,42	16,90	13,76	19,40	19,70	27,19	29,56	30,87	31,67
4th day of week	16,31	14,70	18,03	19,95	18,14	17,50	15,13	20,18	17,99	25,73	31,20	31,03	32,55
5th day of week	14,72	14,87	18,61	20,18	21,44	17,47	16,83	23,07	17,97	24,54	29,91	30,98	34,35
6th day of week	16,08	14,12	19,08	17,83	20,24	19,41	17,45	23,16	20,70	24,92	30,48	30,31	33,96
7th day of week	16,08	15,57	18,36	20,28	17,48	20,36	17,65	22,20	23,05	26,80	31,90	26,18	32,97
Mean	**16,36**	**15,73**	**17,80**	**18,29**	**18,27**	**17,22**	**15,73**	**20,88**	**19,30**	**24,50**	**29,86**	**30,22**	**31,94**
”cold” condition													
Week	**Week 1**	**Week 2**	**Week 3**	**Week 4**	**Week 5**	**Week 6**	**Week 7**	**Week 8**	**Week 9**	**Week 10**	**Week 11**	**Week 12**	**Week 13**
1st day of week	14,18	10,22	12,79	5,20	8,65	12,76	15,68	18,30	16,57	16,06	13,65	12,21	13,79
2nd day of week	6,78	8,86	14,70	8,05	9,30	14,17	14,30	16,39	18,76	15,87	11,66	15,78	14,38
3rd day of week	11,28	8,65	13,17	10,51	9,75	14,78	14,67	15,98	11,76	16,58	14,38	15,05	15,27
4th day of week	6,83	11,01	10,70	7,58	12,65	13,71	14,41	14,65	17,42	16,38	17,75	15,95	15,23
5th day of week	8,92	12,73	10,03	10,88	12,80	15,67	15,96	15,02	17,53	16,60	19,25	16,55	16,30
6th day of week	10,61	12,58	9,85	11,50	13,54	15,36	17,30	14,23	17,63	16,74	16,63	18,05	16,88
7th day of week	10,62	12,79	6,64	10,43	13,23	15,95	18,11	15,47	17,41	15,86	14,63	19,80	16,79
Mean	**9,89**	**10,98**	**11,12**	**9,17**	**11,41**	**14,63**	**15,78**	**15,72**	**16,72**	**16,30**	**15,42**	**16,20**	**15,52**



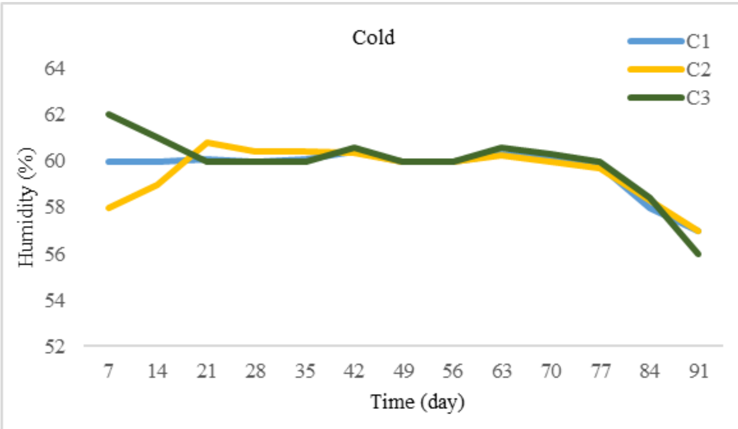


Fig. 1: The humidity percentage graph of three agri-food derived composts within 90 days. Abbreviations: C1, coffee husk (CH) and brewers' spent grain (BSG) in proportion 2:1; C2, CH and cow manure (CM) in proportion 4:1; and C3, a mixture of CH, BSG, and CM in proportion 5:3:2.

2.3. Composting and experimental design

The composts used in the recent work include coffee husk (CH) and brewers' spent grain (BSG) in proportion 2:1 (C1), CH and cow manure (CM) in proportion 4:1 (C2), and a mixture of CH, BSG, and CM in proportion 5:3:2 (C3) in plastic piles, which prepared in the course of 90 days. Every plastic pile had around 1 m^3 volume with some installed pipes in the bottom for aeration. After mixing 300 kg of the above-mentioned materials in every pile, they were covered by plastic bags to avoid humidity and heat loss. Composts were made by using the aerated static pile method, and aeration was performed by compressor every ten days for 15 min. For better air movement in the piles, the wood chips were put in several layers between materials to increase the porosity and penetration capability. The temperature of piles was measured every day and tried to keep the humidity approximately 60%. All the conditions for two composts were kept the same, except temperature differences. Only coffee husk compost was made before the experiment and due to the high pH (more than 9), we decided to obtain a mixture with different organic waste to make a balance in pH. *Salmonella* test on all of the compost samples and raw materials was negative.

2.4. Sampling

Sampling was carried out every 30 days of the composting process, and the samples were immediately kept in a -20 °C fridge. In this research, we compare the same sampling times in the terms of compost parameters between the “warm” and “cold” temperature differences (T1-T4) to reach the best compost quality.

2.5. Analyzed parameters

Microbial biomass carbon (MBC) was estimated through the fumigation extraction procedure by [17] and hydrolysis of fluorescein diacetate (FDA) was determined by [18]. Germination index (GI %) of compost was evaluated by the germination test of Chinese cabbage [19]. The soluble extract of compost was used to measure nutrient and trace elements in the compost. To make a solution, the ratio of compost to distilled water was 1:10, and it remained overnight in the lab, then centrifuged for 10 min, and the solution was filtered by Whatman paper, and finally, the Inductively Coupled Plasma mass spectroscopy ICP-MS test was performed on the solution.

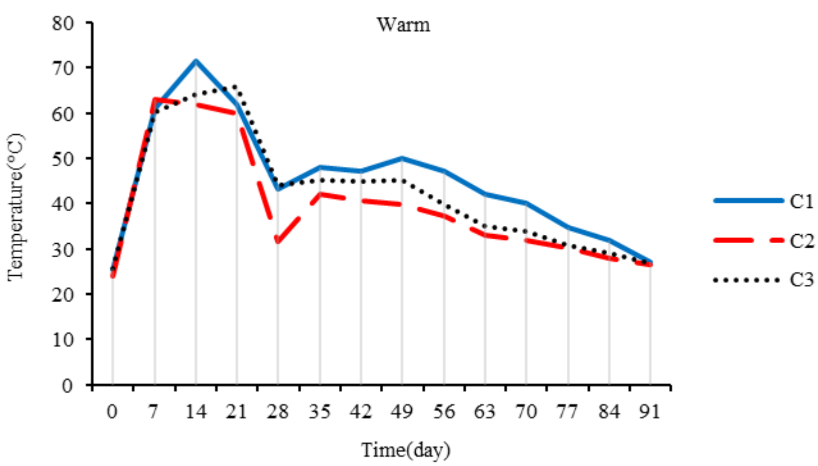
2.6. Statistical analysis

A correlation test was performed with the R software and corrplot package. To investigate the relationship between temperature and compost variables, the repeated measure analysis was used along with Tukey's range post hock test ($P< 0.05$) in the SPSS V. 25 software. The analysis of nutrient and trace elements was performed with PCA by using R software and factoextra package.

3. RESULTS AND DISCUSSION

3.1. Graph of composting temperature throughout the study

Temperature is an important indicator that affects microbial activities throughout the time of the composting process. The temperature shifts within this process happen in three phases: the mesophilic (20ºC–45ºC), thermophilic (45ºC–70ºC), and cooling step [16]. Temperature displayed an increment in the initial stage of composting for both temperature conditions, from mesophilic to thermophilic (14^{th} day) phase followed by a decrement in later steps of compost production (90^{th} day) (Fig. 2). The "warm" (higher temperature) composting led to a rise in temperature much faster than the "cold" (lower temperature) (especially C1 treatment). The time-temperature profile of the composting process was mostly in the thermophilic range for the "warm" with the highest temperature recorded in the C1 compost (~70 °C) by the 14^{th} day followed by the C3 and C2 (63 and 66 °C). However, the "cold" treatments registered an increment in temperature (66 °C) within the severe microbial degradation phase of composting, i.e., the 14^{th} day. Thermophilic microorganisms, which facilitated oxidative reaction, are presumably responsible for the heat released throughout "warm" composting, exhibiting an effect of temperature [1]. The appropriate aeration and higher temperature in the C1 treatment may have affected microbial activities that led to better stability of compost and higher mineralization of organic matter [13]. Consequently, these concepts verified that the "warm" composting has obtained a better degree of improvement. A lower temperature (18 °C) was recorded around 90^{th} day (at maturation time) in "cold" treatments than those of "warm" (28 °C), which stated partial composition that in turn leading to immature compost in the cold conditions. In line with our findings, Khalil *et al.* (2001) [20], tracked the shifts in temperature of solid waste compost. They demonstrated that the compost mass temperature in all seasons of the year reached the max level after 21 days of the composting process and then declined by the end of the process but did not touch ambient temperature. The authors and other researchers [16] suggested that the harmer season provides an opportunity for the compost for being matured in a faster manner. Cheng *et al.* (2019), reported that composting temperature should be kept over 50 °C for 10 days or over 60 °C for 5 days, and below 70 °C to guarantee that the composting process runs normally. The high temperatures (> 45 °C) help to shift the $NH4^{+}$ to NH3 equilibrium towards ammonia and inhibit nitrification at the same time, both of which would increase ammonia volatilization [21].



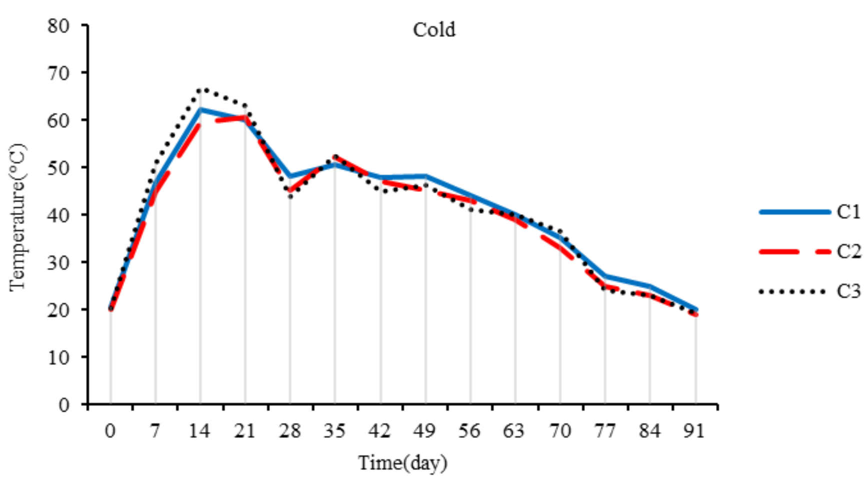


Fig. 2: The temperature graph of three agri-food derived composts within 90 days. Abbreviations: C1, coffee husk (CH) and brewers' spent grain (BSG) in proportion 2:1; C2, CH and cow manure (CM) in proportion 4:1; and C3, a mixture of CH, BSG, and CM in proportion 5:3:2.

3.2. General characters of compost (C/N, pH)

As observed in the recent work, pH clearly differed for both "cold" and "warm" treatments (Fig. 3). The pH value for each compost pile on the 0^{th} day was between 7.5 and 7.8. The pH remarkably increased up to 60^{th} day for C1 (both temperature conditions), however, those of C2 and C3 ("warm") remained in neutral-phase values or sequentially decreased ("cold"). The C1 treatment displayed a max pH registered ~9 and ~8.5 for "cold" and "warm", respectively, on the 60^{th} day of study. The initial pH (~7.5) was increased in the C1 treatment (both temperature conditions) up to the 60^{th} day with a subsequent decrement at the end of compost production (90^{th} day). The increased pH from the 0^{th} to 60^{th} day revealed protein breakdown in the organic wastes with the release of NH_3, which was in line with [22]. Besides, this alkaline pH maybe because of the faster biosynthesis of phenolic metabolites within the degradation time [13]. However, pH was decreased in the 90^{th} day, i.e., at the end step of decomposition. All the accounts, this is suggestive of a fast degradation of organic materials in "warm" treatments (C1, C2, C3) with regard to the course of composting. In the cold condition, the C2 and C3 treatments exhibited decreasing pH at the same time with increasing days of composting, revealing higher organic acids and slower degradation in these treatments. As stated by [16], for efficient composting, natural wastes with low pH values in the 90^{th} day of composting (such as C2 and C3 in this work) must be blended with other materials achieving suitable situations so that the initial pH of the substrate for compost production can be considered in the ~7.5 value.

In compost production, the C/N ratio is important so that if this ratio being high at the onset of the process, the decomposition of organic materials will be slower. The soil generally comprises a C/N ratio of 10:1. Data for the effect of temperature variations on the C/N ratio within the composting process has been shown in Fig. 3. The findings demonstrated that the C/N ratio for the "warm" condition, especially C2 and C3, was better than all the other treatments. Throughout three months, the C/N ratio decreased from 14.5 (0^{th} day) to 12.8 for the treatment C1 followed by C3 and C2 (11.9 and 11.4) for "cold". Moreover, throughout three months, the C/N ratio decreased from 13.5 (0^{th} day) to 10.6 for treatment C1 followed by C3 and C2 (9.7 and 9.4) for "warm". Similarly, [20] documented that decrement in the C/N ratio in "cold" was higher than in "warm". Our observations are indicative of an acceptable maturity (C/N ratio of 10 up to 15), as stated by [23]. The lower reduction of the C/N ratio from 30^{th} up to 60^{th} day in the C1 revealed more evolution of CO_2 owing to C transformation within "warm" [16]. Besides, the drop in moisture level in the "cold" conditions may reduce the metabolic reaction of microorganisms, leading to an increased C/N ratio. Interestingly, we observed that although the highest decrement in the ratio of C to N was recorded on the last day of the experiment in "cold", a primary decrement can be indicated in "warm". It is recommended that for efficient composting with a lower period of time, the primary acclimatizes must focus on the initial steps of composting activity [24]. The finding is in line with that of [25][8], where "warm" compost stabilizes faster than "cold", demonstrating the potency of the composts for fast degradation of organic matters.

3.3. Microbial biomass carbon (MBC)

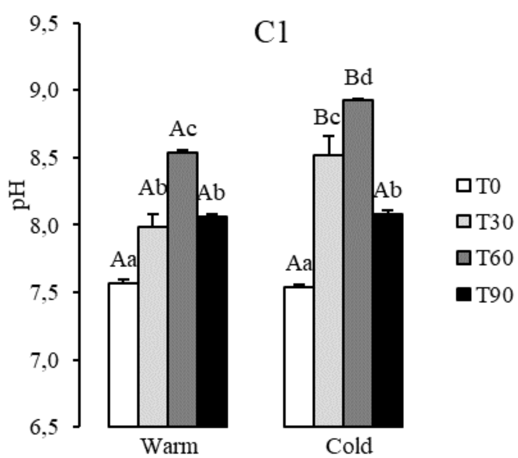
From our observations, it seemed that microbial biomass carbon (MBC) was increased from 0^{th} to 30^{th} day followed by a decrease from 30^{th} to 60^{th} day, and an increase from 60^{th} to 90^{th} day in both temperature conditions (Fig. 3). It appeared that with time, the max reduction in MBC from 8.5, 7.9, and 7 (0^{th} day) to 6.3, 4.4, and 4.8 (90^{th} day) in the C3, C1, and C2, respectively, for "warm" treatments, more stabilized remarkably than "cold". Further decrease in the C2 was attributed to the existence of readily obtainable nutrients in cow manure, sourcing effective microbial degradation and high temperature. This is in line with findings observed by [26], stating that the decomposition was stopped, and the compost obtained maturity by the 30th day in “warm”. However, the favorable output for MBC could not be achieved in “cold”, likely owing to an inadequate rise in temperature and low microbial multiplication. Similarly, it was reported a considerable increase in the microbial biomass carbon attained in spring and autumn seasons compared to winter and summer [20].

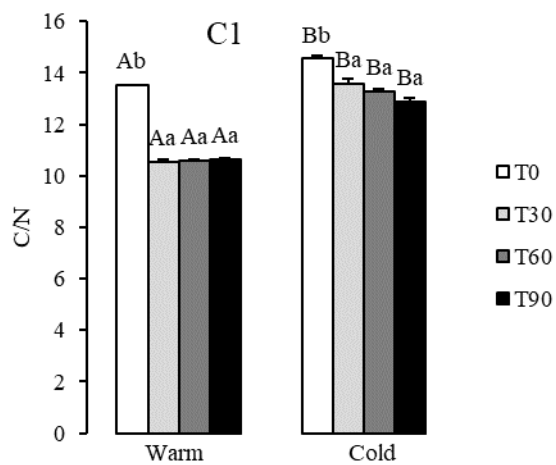
3.4. Hydrolysis of fluorescein diacetate (FDA)

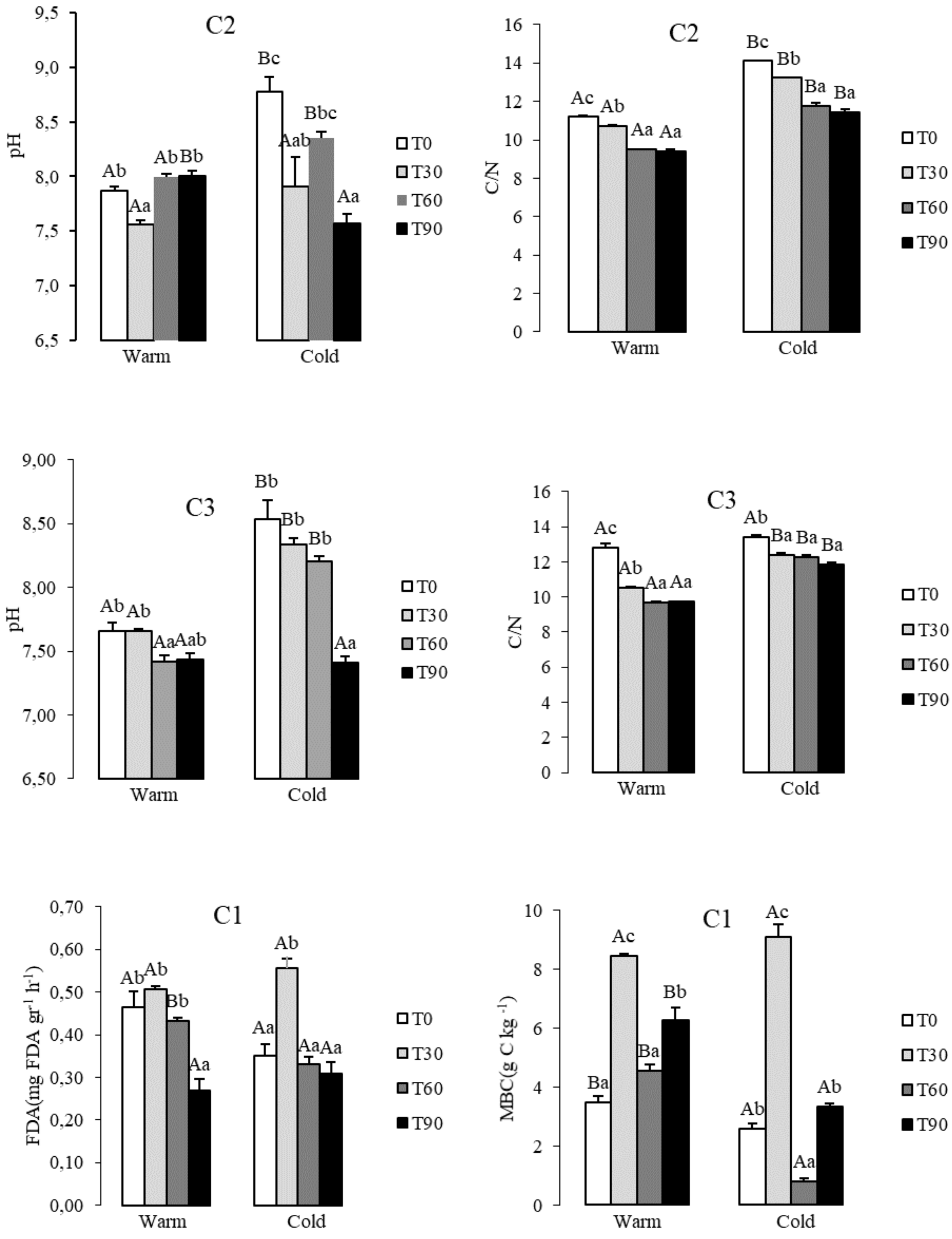
The tracking of enzymatic activities is imperative for obtaining insight into microbial transformations happening throughout the composting process [27] and, since a wide range of hydrolytic enzymes are produced by microorganisms during the composting process, FDA hydrolysis was used as an indicator of enzyme activities. The FDA-enzyme activities indicated an overall increment for all the treatments (“warm” and “cold”) from the 0th to 30th day and a decrement from the 30th to 90th day of composting (Fig. 3). Among the treatments, FDA was recorded to be the highest in the treatment C3 (0.61 mg FDA g^{-1} h^{-1}) followed by C2 (0.61 mg FDA g^{-1} h^{-1}) and C1 (0.55 mg FDA g^{-1} h^{-1}) in the 30th of composting under “cold” conditions, attributed to accelerated enzymatic activities in the treatments by readily available nutrients in cow manure. The mixture of coffee husk, brewers’ spent grain, and cow manure may stimulate the enzymatic synthesis owing to excess accessibility of substrates. At later steps (from 30th to 90th day), the enzymatic activity dramatically decreased for both temperature treatments. As previously reported, this decrement is associated with a decline in the C/N ratio and pH when mineralization is happening within composting [15], which was in line with our observations.

The enzyme activity of the 60th day was higher in the "warm" than in "cold" conditions, as also reported by [20].

Muscolo et al. (2018) findings evidenced many differences between compost properties highlighting that stability/maturity doesn't means quality. In some case these properties may overlap, but in the majority of the situation can be different and need to be discriminated. Maturity and stability are mainly linked to composting parameters and can be assessed by measuring C/N, TN values and carbon loss. While the quality of composts is mainly linked to chemical composition of raw material and can be assessed evaluating the compost effects on soil ecosystem functioning by monitoring fungi/bacteria ratio, FDA, activity of dehydrogenase (DHA) and nutrient amount. FDA and DHA activities have been identified as markers for assessing the quality of amended soils [28]. Researchers findings showed an increase in microbial biomass, bacteria, actinomycetes, DHA and FDA in soil amended with compost suggesting an intense biological activity driving a mineralization processes with a greater release of nutrients, increase in EC and pH [28].






C2
pH
9,5
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Aab
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Aa
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Aa
Warm
Cold
T0
T30
T60
T90
C2
C/N
16
14
12
10
8
6
4
2
0
Bc
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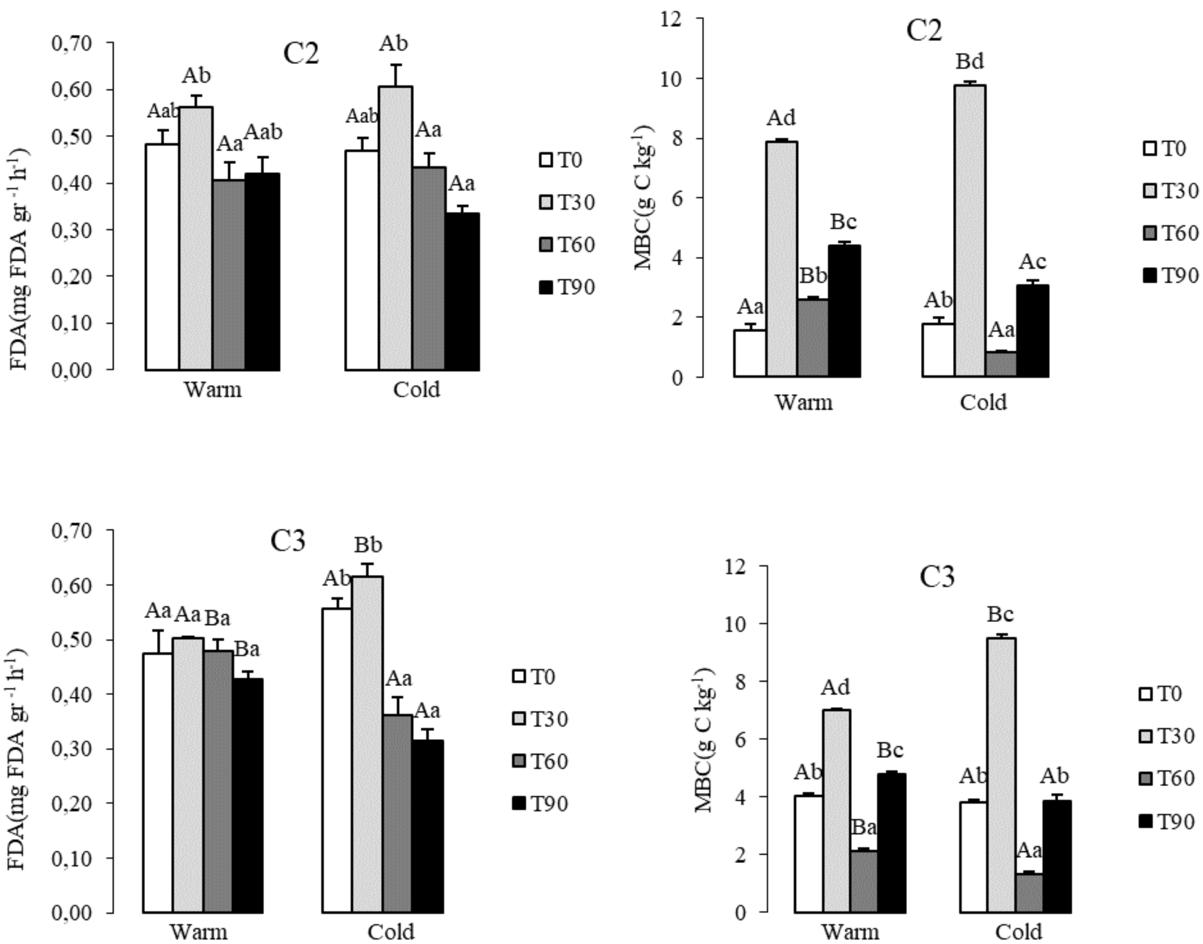


Fig. 3: The analyzed parameters of three agri-food derived composts. Abbreviations: C1, coffee husk (CH) and brewers' spent grain (BSG) in proportion 2:1; C2, CH and cow manure (CM) in proportion 4:1; and C3, a mixture of CH, BSG, and CM in proportion 5:3:2.

3.5. Nutrient and trace elements analysis by PCA

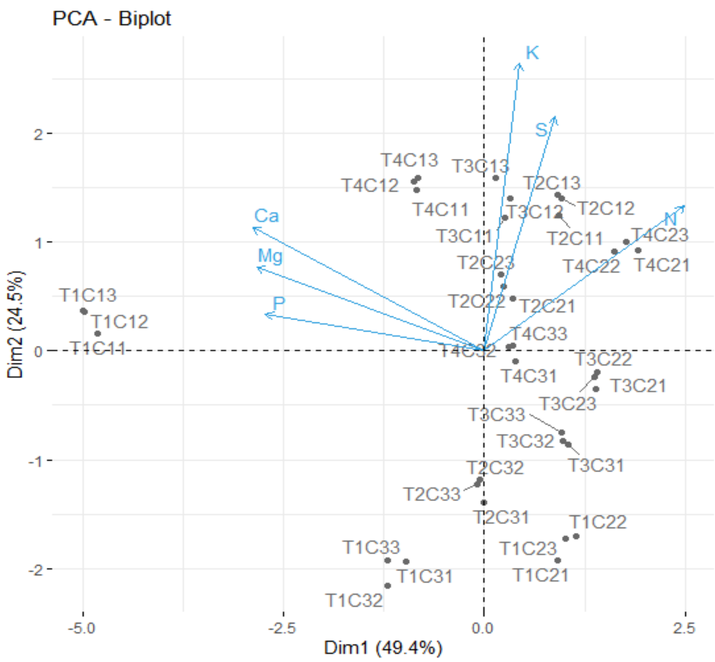
The output of PCA analysis for different nutrient and trace elements of the compost was represented in Tables 2 and 3, Fig. 4, 5. This statistical tool illustrates the degree of association among the components. From the findings obtained from nutrient elements, two principal components with an eigenvalue -5.0 to +2.5 were obtained, where PC1 and PC2 demonstrate the major conjunctions among the nutrient elements. For the "warm" conditions, variation for PC1 and PC2 corresponds to 49.4% and 24.5% as compared to 34.9% and 25.7% of the "cold" conditions. The indicators presented far away from the center display more correlation, suggesting higher variation in the process. In the "warm", the PC1 corresponds to K, S, and N, while PC2 corresponds to P, Ca^{+2}, and Mg^{+2}. The C1 and C2, which had the highest values for the first and second components, were determined as the most effective treatments. However, the PC1 corresponds to P and Ca^{+2}, while PC2 corresponds to N, S, K, and Mg^{+2} in the "cold". The C1 and C3, which had the highest values for the first and second components, were determined as the most effective treatments.

From the findings attained from trace elements, the most significant indicators falling within PC2 were Al, Ba, Mn, Zn, Fe, Cu, Pb, Cd, and Ni under "warm" conditions. On the other hand, only Na was recorded leading to variation in PC1. The C2 and C3, which had the highest values for the first and second components, were determined as the most effective treatments. Our observations relative to "cold" conditions were the opposite of the "warm" ones, so that only the element Na was in the PC2 and the rest of the elements were in the PC1. The C1 and C2, which had the highest values for the first and second components, were determined as the most effective treatments. From our results, it can be concluded that the nutrient and trace elements have a high variation in the different temperatures of the year, as described by [13] who studied compost nutrients in four seasons of the year by the PCA analysis.

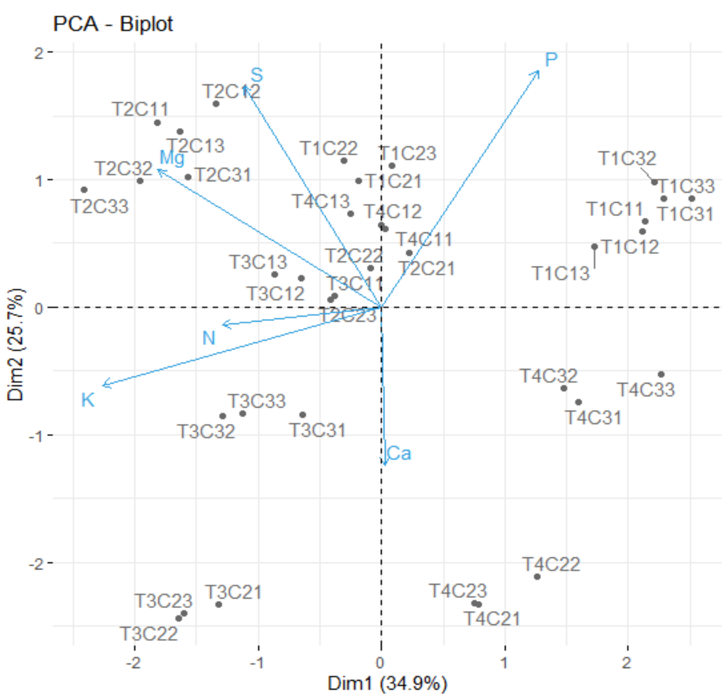
Table 2. Principal component analysis (PCA) for nutrient elements over different temperature conditions.

Nutrient elements	"warm"		"cold"	
	PC1	*PC2*	*PC1*	*PC2*
Ca (mg L^{-1})	-0.892^{**}	0.351^{*}	0.010^{ns}	-0.499^{*}
Mg (mg L^{-1})	-0.876^{**}	0.237^{ns}	-0.730^{**}	0.432^{*}
K (mg L^{-1})	0.133^{ns}	0.818^{**}	-0.909^{**}	-0.247^{ns}

S (mg L^{-1})	0.272^{ns}	0.665^{**}	-0.451^{*}	0.695^{**}
P (mg L^{-1})	-0.842^{**}	0.104^{ns}	0.512^{*}	0.745^{**}
N (%)	0.772^{**}	0.411^{*}	-0.517^{*}	-0.055^{ns}



Nutrient Elements
("warm" condition)

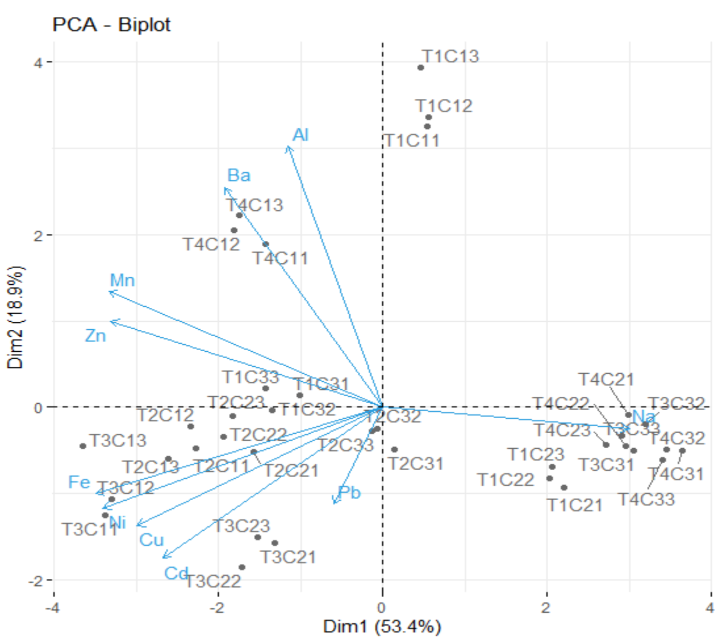


Nutrient Elements
("cold'' condition)

Fig. 4: Principal component analysis (PCA) for nutrient elements over different temperature conditions and times. Abbreviations: C1, coffee husk (CH) and brewers' spent grain (BSG) in proportion 2:1; C2, CH and cow manure (CM) in proportion 4:1; and C3, a mixture of CH, BSG, and CM in proportion 5:3:2. T1-T4 indicate 0th, 30th, 60th, and 90th day of composting process, respectively.

Table 3. Principal component analysis (PCA) for trace elements over different temperature conditions.

Trace elements	"warm"		"cold"	
	PC1	*PC2*	*PC1*	*PC2*
Al (mg L^{-1})	-0.307*	0.801**	0.853**	-0.031^{ns}
Cd (mg L^{-1})	-0.707**	-0.463*	0.876**	0.103^{ns}
Fe (mg L^{-1})	-0.922**	-0.265^{ns}	0.954**	-0.036^{ns}
Ni (mg L^{-1})	-0.901**	-0.308*	0.888**	0.192^{ns}
Pb (mg L^{-1})	-0.155^{ns}	-0.29*	0.281^{ns}	0.911**
Mn (mg L^{-1})	-0.882**	0.355*	0.949**	-0.097^{ns}
Zn (mg L^{-1})	-0.874**	0.264^{ns}	0.873**	-0.377*
Na (mg L^{-1})	0.794**	-0.064^{ns}	-0.826**	0.269^{ns}
Ba (mg L^{-1})	-0.509*	0.673**	0.841**	0.054^{ns}
Cu (mg L^{-1})	-0.791**	-0.364*	0.934**	0.152^{ns}



Trace Elements
("warm" condition)

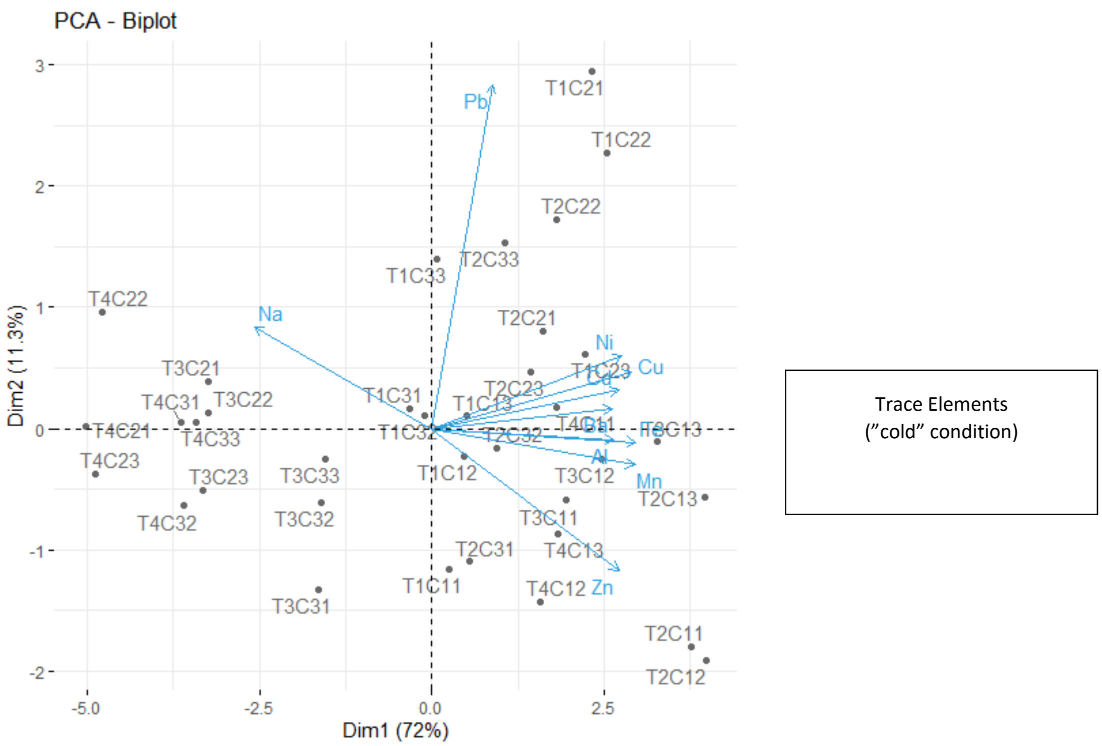


Fig. 5: Principal component analysis (PCA) for trace elements over different temperature conditions and times. Abbreviations: C1, coffee husk (CH) and brewers' spent grain (BSG) in proportion 2:1; C2, CH and cow manure (CM) in proportion 4:1; and C3, a mixture of CH, BSG, and CM in proportion 5:3:2. T1-T4 indicate 0^{th}, 30^{th}, 60^{th}, and 90^{th} day of composting process, respectively.

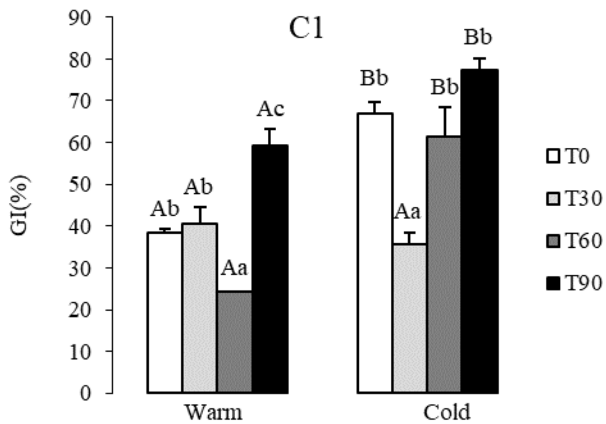
3.6. Maturity (GI %)

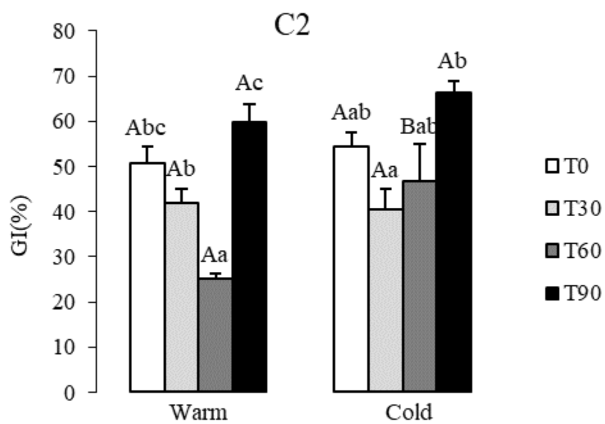
Quality control throughout the composting must ensure sufficient physical and chemical features, as well as a sufficient degree of maturity and stability [29]. An appropriate assessment of compost maturity is imperative for the successful use of agri-food wastes in agriculture. The available analyses differ in approach, duration, costs, simplicity, and precision. However, bioassays are concerned the most direct procedure for maturity because it displays the influence of compost maturity on plant growth [16]. The unstable and/or immature compost can have adverse effects on seed germination, plant growth and soil environment due to the decreased supply of oxygen and/or available nitrogen or the presence of phytotoxic compounds [30].

Stability is the resistance of the organic matter in compost against further microbial decomposition as long as there is no inhibition on the microbes by other factors not relevant to the organic matter, while maturity is an agronomic parameter that is clearly associated to the effect of compost on plant growth [31]. The indices of respiration and humification of compost are used to evaluate the stability and maturity of compost, respectively. Furthermore, the substances, including low molecular weight organic acids (e.g. phenolic acids), ammonium nitrogen ($NH4^+$-N), salinity, heavy metals, xenobiotics (e.g. antibiotics and agrochemicals), can cause damage to plants when they are in high levels. In general, many of these substances need to be evaluated via time consuming and expensive detection processes to determine whether their levels are beyond or within acceptable ranges. However, there exists the possibility for the unexpected factors that are not taken for analysis. Furthermore, there is a lack of analytical procedure to evaluate the joint effect of the toxic substances in compost. Consequently, as a bioassay, seed germination test has attracted a lot of attention to overcome these concerns. The seed germination test is an effective and economical bioassay to evaluate the potential toxicity and maturity of compost before it can be used [19]. The seed germination index (GI) was firstly proposed by Zucconi et al. (1981) [32] who used cress seeds in the germination test for evaluating the toxicity of compost. GI is calculated by the radicle length and germination percentage of the seeds in the sample (compost extract) compared to that in the control (e.g. deionized water). GI is correlated with some other biological and chemical indices for evaluating compost quality. Researchers showed that GI was positively correlated with the biological index of the *Artemia salina* cytotoxicity test for evaluating the toxicity of compost. In addition, GI is positively correlated with humification parameters while negatively correlated with the content of $NH4^+$. Therefore, the seed germination test has been broadly accepted for evaluating compost quality. In Italy, GI is listed in the quality assessment regulation of compost for commercialization [19].

Moreover, time courses of seed germination are usually several days under suitable conditions, which can be morphologically divided into three phases that consist of phase I (imbibition), phase II (radicle emergence) and phase III (radicle elongation). The uptake of water is the major process of seed germination during the phase I, which could be negatively affected by high salinity of compost. During the phase II, the low molecular weight organic acids of compost could be the primary inhibitor of radicle emergence after test a rupture. Radicle elongation could be inhibited by $NH4^+$ during the phase III. This speculation partly supports a viewpoint that seed germination can be used to examine the compost with high toxicity and radicle growth can be used to examine the compost with low toxicity [32]. Further studies are needed to validate the effectiveness and applicability of this conception in the seed germination test [19].

Overall, in this study, the index maturity decreased from the first to the 30th day, and then from the 30th day to the 90th day with an increase in all three composts under both temperature conditions (Fig. 6). The highest GI % was observed in C1 (77%) followed by C3 and C2 (70 and 66%) in the cold condition. At a lower level than “cold” conditions, the highest GI % was recorded in C2 and C3 (~60%) followed by C1 (59%) under “warm” conditions. This is in agreement with findings observed by [26], indicating that the decomposition had slow down, and thereby the composts obtained maturity by the 90th day under “warm” conditions. In line with [13], “cold” composting conditions cannot obtain a favorable result and generated immature composts even following three months. Totally, the germination index showed values greater than 80%, indicating absence of phytotoxins, and also compost stability and maturity [28] and germination index values lower than 60% indicate phytotoxic effects significantly different from the control [14].





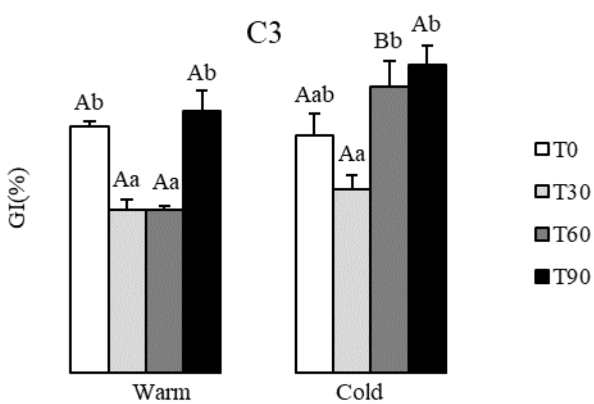
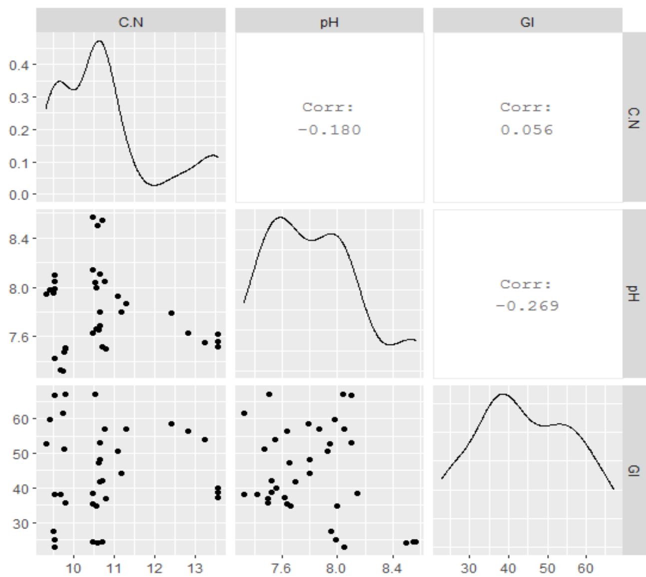


Fig. 6: Maturity (GI%) of three agri-food derived composts. Abbreviations: C1, coffee husk (CH) and brewers' spent grain (BSG) in proportion 2:1; C2, CH and cow manure (CM) in proportion 4:1; and C3, a mixture of CH, BSG, and CM in proportion 5:3:2.

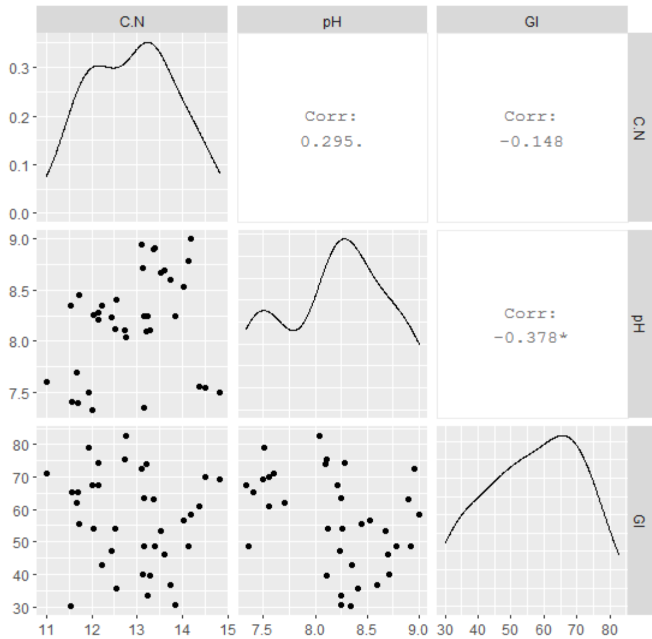
3.7. Correlation between GI%, C/N, and pH as maturity indexes

An understanding of the correlations among the maturity indexes that affect the final product of the composting process can decline the number of evaluations and thereby save money. Fig. 7 details the relationship between the GI%, C/N, and pH assayed in the final product of the composting process. The pH value negatively correlated with the GI% in both temperature conditions, while positively correlated with the C/N under "warm" conditions. It can be concluded that low pH accelerated the decomposition of low-weight carboxylic acids, leading to an increase in organic carbon [33]. However, it was found a negative correlation between pH and C/N under "cold" conditions, as demonstrated [33]. From our results, the C/N ratio negatively correlated with GI% under "warm" conditions. [34] evaluated the relationships between the stability and maturity indicators and various properties at different phases of the composting process. The authors demonstrated that the C/N ratio was correlated highly with several indicators thus these indicators can be utilized to evaluate the maturity of compost obtained by the mixing of various kinds of agri-food wastes. Of the indexes studied, pH in the composts indicated the closest relationship with GI%. Focusing on pH value, as maturity index, can be a useful procedure to decrease the number of assessments and therefore ensure that the compost is viable economically. As described by [35], the C/N ratio within composting process may also be another index by which maturity, only under "cold" conditions, can be evaluated cost-effectively.

According to CCQC Maturity Index [36] claimed, three components should be included in the maturity index, C/N ratio, followed by the other two aspects: Group A (CO2 production, or O2 consumption, or heat generation) and Group B ($NH4^{+}$-N/$NO3^{-}$-N, or CNH3, or GI), so that C/N ratio, heating and GI, together with the humification coefficient (HC), concentration of $NH4^{+}$-N, water-soluble organic carbon (WSOC) and organic matter (OM), to evaluate the nutrient availability in different treatments.



Correlation matrix for maturity indexes ("warm" condition)



Correlation matrix for maturity indexes
(”cold” condition)

Fig. 7: Correlation matrix for maturity indexes: A: “warm” condition, B: “cold” condition.

4. CONCLUSION

The quality and composition of the raw materials entering composting processes are affected by both temperature conditions, which led to remarkable effects on pH, C/N ratio, microbial biomass carbon, enzyme activity (FDA hydrolysis), nutrient and trace elements, and maturity (GI %) of agri-food composts produced. In the light of such conditions, coffee husk and brewers’ spent grain-derived compost prepared under “warm” conditions appeared the most favorable option for the composting process due to the relatively higher environmental temperature.

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CHAPTER4

Agri-food derived composts improve the soil physicochemical features and wheat yield

ABSTRACT

It is well-documented that the achievement of a high yield of wheat depends on retaining the nutrients in the plant root environment, which is important for intended crop production. Due to the high cost and environmental side effects of chemical fertilizer, there is a growing tendency to utilize organic fertilizer like compost in agriculture. Authors made three compost mixtures with coffee husk (CH) and brewers' spent grain (BSG) (Pile 1), CH and cow manure (CM) (Pile 2), and CH, BSG, and CM (Pile 3) then applied prepared compost on wheat field soil. Based on our observations, wheat grain yield increased from ~3.3 ton ha^{-1} (control) to ~5.2 ton ha^{-1} (Pile 1 treatment). Of the lipids extracted from compost-treated wheat in the recent work, C18:2 n-6, C18:1Δ9c, C16:0, and alpha-18:3 were the most abundant types, accounting for an average of 95% of the fatty acids analyzed. No main effects of composts were observed for fatty acids. Soil physic-chemical features were improved by the application of composts, especially the compost derived from coffee husk and brewers' spent grain. Moreover, this compost had positive influence on the microbial enzyme activities and biomass, probably due to the composition and higher input of carbon organic into the soil. As a result, it can be suggested to use this compost to reduce environmental pollution, improve soil fertility, and achieve high production of wheat crop.

***Key Words**:* Compost; Soil physic-chemical features; Grain yield; Soil fertility.

1. INTRODUCTION

It is proved that the achievement of a high yield of wheat depends on holding the minerals in the plant root environment, which is imperative for successful crop production. As observed by Ref. [1], the universal utilizations of P and N fertilizers have been elevated by 3.5- and 7-fold, respectively, in the past sixty years; both fertilizers are anticipated to elevate further 3-fold by 2050. Besides, the abundant application of fertilizers also raises the environmental pollution risk [2]. Moreover, due to the high cost of chemical fertilizers, there is a rising tendency to utilization organic fertilizers like compost in farming, particularly in arid and semi-arid areas of the world [1]. Thus, the implementation of compost to farming soil may be economical and sustainable.

Composting can be an environmentally safe, cost-effective, and efficient biological procedure for recycling residual biomass in agriculture [3]. Several advantages have been suggested for the application of compost as fertilizer, like enhancing microbial activity and organic content [4]. High content of nutrients such as K, P, and N can be obtained from compost usage [5]. Moreover, from Ref. [1], it has been recorded that natural compost implementation has a promoting influence on wheat yield, agronomical traits, and grain nutrient content than chemical fertilization. Therefore, it can be suggested to substitute chemical fertilizers with organic compost to achieve the high production of this crop.

As a natural product, coffee husk possesses tannins and caffeine, which can make it slow degradation toxic and, in the environment, leading to the disposal challenge. Nevertheless, this compost is rich in lignocellulose materials, which make it a favorable substrate for microbial activities [6]. Multiple, alternative applications of coffee husk have been suggested because it improves the biological and physic-chemical properties of soil, enhances soil organic matter, and decreases the depletion of natural sources [6]. Brewer's spent grains, a by-product of the brewing industry that makes up %80 of brewing waste, can be utilized as soil fertilizer alongside material recently known as wastes [7]. As reported previously, these materials in suitable mixtures by microorganisms can be added to soil. Moreover, the compost derived from brewer's spent grains can compete admiringly with chemical fertilizers in terms of crop yield in the field [7]. Cow manure, comprising a large amount of K, P, and N endangers human health and has become the main contaminant in environment due to inadequate application [8]. Aerobic composting can stabilize organic nutrients in manure and keep down their harmful influences through fungi and bacteria [9][10]. As above-mentioned, composting presents a promising tool for converting livestock manure into a nutrient source for horticultural and agricultural applications [8].

With such situations in mind, this work was aimed at the comparison of three types of composts [coffee husk (CH) and brewers' spent grain (BSG) in proportion 2:1 (Pile 1), CH and cow manure (CM) in proportion 4:1 (Pile 2), and a mixture of CH, BSG, and CM in proportion 5:3:2 (Pile 3)] on different soil properties and wheat yield to produce agri-food-derived compost, which improving the soil physicochemical features and wheat performance in the field. The research hypotheses were: i) there is a difference between composts and control in the soil properties and subsequently wheat yield and lipid fractions; ii) Pile 1 compost has the better quality in comparison of other composts because it has the most ratio of coffee husk possessing a high amount of C/N.

2. MATERIAL AND METHODS

2.1. Composting and experimental design

The current study was focused on the comparison of composts with coffee husk (CH) and brewers' spent grain (BSG) in proportion 2:1 (Pile 1), CH and cow manure (CM) in proportion

4:1 (Pile 2), and a mixture of CH, BSG, and CM in proportion 5:3:2 (Pile 3). The dimension of every pile was 1 m^3, and the weight of its materials was around 300 kg. All piles were covered with a plastic film in order to prevent excessive moisture loss and preserved in the greenhouse. The aerated static pile was followed as the composting method by Ref. [11]. To aerate the pile, layers of loosely piled bulking agents like wood chips were added so that air could pass from the bottom to the top of the pile.

The field preparation was performed in the first week of November 2018 in Agugliano, Marche region (Coordinates: 43°33′N 13°23′E; Elevation: 203 m), Italy, and around 10 kg of compost were distributed in any plot with 18 m^2 (corresponding to about 5.5 Mg ha^{-1}) on 27th November the wheat seeds cultivated in the arranged field by Completely Randomized Design (CRD) method in three replications. The total dimension of the field was 216 m^2, with margin and corridor was 513 m^2. Totally, 12 blocks with 18 m^2 dimensions were considered.

The soil descriptions were carried out three times i) in time zero (T0) before compost distribution and cultivation, ii) after compost distribution and cultivation (T1), and iii) time harvest (Th) after wheat harvesting. Soil samples were collected at T0 and Th in 50 cm depth in three horizons (Ap1, Ap2, and Ap3) and then air-dried at room temperature, grinded and sieved at 2 mm, and stored at 4 °C for further soil analysis.

2.2. Soil experiments methods

Soil pH was determined in water (1:2.5 weight/volume) using a combined glass-calomel electrode. On specimens ground to less than 0.5 mm, the organic C (OC) content was estimated by the Walkley-Black method without the application of heat [12], while the total N (TN) content was determined by the semi-micro Kjeldahl method. Available P was determined according to Ref. [13]. Exchangeable K and Mg were displaced by a 0.2 M $BaCl_2$ solution and determined b atomic absorption. Microbial biomass carbon (MBC) was estimated through the fumigation extraction procedure as described by Ref. [14]. Hydrolysis of fluorescein diacetate (FDA) was determined as reported by Ref. [15].

2.3. Statistical analysis

A correlation test was performed with the R software and corrplot package. To investigate the relationship between depths and soil variables, the repeated measure analysis was used along with Tukey's range post hock test ($P < 0.05$) in the SPSS V. 25 software. To decline data dimensionality as well as achieve a better picture of the effect of three composts on wheat yield and soil horizons physic-chemical features, PCA was accomplished on relationship between soil variables and depth (horizon) by R software and factoextra package. To compare wheat product, the one-way ANOVA followed by Tukey's range test ($P < 0.05$) were used in the R software. Before the ANOVA, the dataset was evaluated for variance homogeneity and distribution normality by Levene's and Shapiro-Wilk tests, respectively, both at a 5% significance level.

3. RESULTS

3.1. Soil physicochemical features

Based on the description of soil in the wheat field in the different horizons and times, at T0, there was a diffuse existence of the mesofauna at the soil surface. The soil surface was specified by an Oi horizon, three Ap horizons, and two Bw horizons, displaying hard rupture-resistance related to brittleness and firmness. The Ap horizons were 0- 29 cm thick, after which the Bw horizons were 29-81 cm thick. The structure was moderately developed, with friable platy aggregate in the Ap horizon and a friable angular block in the Bw horizon. The color of soil ranged from reddish yellow in the upper horizon to light brown in the lower one. Very few fine roots were found to the Ap and Bw horizons. Depth and thickness of Ap1 and Ap2 horizons had a tendency to reduce in the soils supplemented with the composts after the crop harvesting. Overall, our findings revealed that the application of composts, especially the compost derived from coffee husk and brewers' spent grain, had remarkable effects on the soil profile (Table 1).

Table 1. Description of soil in the wheat field in the different horizons and times

Name	Horizon [a]	Depth (cm)	Thickness (cm)	Boundary [b]	Matrix color	Structure[d]	Root [e]	Other observation
T0	Oi	-	-	-	-	-	-	-
	Ap1	0-6	6-9	C, W	2.5Y5/3	3, f, m, sbk, fr	1, vf, f	Skeleton 3% mm, mesophaona
	Ap2	6-18	10-12	C, W	2.5Y4/3	3, m, sbk, fr	1, vf, f	Skeleton 3% mm, mesophaona
	Ap3	18-29	-	C, W	2.5Y4/4	2, f, m, sbk, fr, co	-	Skeleton 3% mm,
	Bw1	29-40	12-16	C, W	2.5Y4/4	2, f, m, sbk, fr, co	1, vf	mesophaona
	Bw2	40-82	-	C, W	2.5Y4/3	2, f, m, sbk, fr, co	1, f	-
T1. Pile1	Ap1	0-7	6-9	C, W	2.5Y3/3	3, f, m, sbk, fr	1, vf, f	*Lumbricus* mesophaona
	Ap2	7-18	10-12	C, W	2.5Y3/2	3, m, sbk, fr	1, vf, f	*Lumbricus* mesophaona
T1. Pile2	Ap1	0-7	6-9	C, W	2.5Y4/3	3, f, m, sbk, fr	1, vf, f	*Lumbricus* mesophaona
	Ap2	7-18	10-12	C, W	2.5Y4/4	3, m, sbk, fr	1, vf, f	*Lumbricus* mesophaona
T1. Pile3	Ap1	0-7	6-9	C, W	2.5Y5/4	3, f, m, sbk, fr	1, vf, f	*Lumbricus* mesophaona
	Ap2	7-18	10-12	C, W	2.5Y4/4	3, m, sbk, fr	1, vf, f	*Lumbricus* mesophaona
T1.CTRL	Ap1	0-7	6-9	C, W	2.5Y5/3	3, f, m, sbk, fr	1, vf, f	*Lumbricus* mesophaona
	Ap2	7-18	10-12	C, W	2.5Y4/4	3, m, sbk, fr	1, vf, f	*Lumbricus* mesophaona
Th. Pile1	Ap1	0-3	2-3	A, W	2.5Y4/4	1 f, vf, gr	-	-
	Ap2	3-10	7-8	C, W	2.5Y4/2	3 f, vf, m, abk	2 vf	Dead vegetable, compost
	Ap3	10-28	18-19	C, W	2.5Y4/3	2 f, m, sbk	2 vf	Dead vegetable
	Bw1	28-38	6-8	C, W	2.5Y4/4	2 f, m, sbk	2 vf	-

	Bw2	38-52$^+$	-	-	2.5Y4/4	1-2 f, m, sbk	1-2 vf	Dead root, no rock, Dead vegetable
Th. Pile2	Ap1	0-4	1-4	A, W	2.5Y4/3	1 f, m, sbk, gr	-	Dead vegetable
	Ap2	4-14	10-15	C, W	2.5Y4/2	3 f, vf, m	1 f	Compost residue
	Ap3	14-24	10-12	C, W	2.5Y4/2	3 f, vf, m, abk, sbk	2 vf	Dead vegetable, shell fragment,
	Bw1	24-35	9-10	C, W	2.5Y4/3	3 f, vf, m, abk, sbk	2 vf	Dead vegetable, shell fragment,
	Bw2	35-50$^+$	-	-	2.5Y4/3	3 f, vf, m, abk, sbk	1 vf	Dead vegetable, shell fragment, mesophona
Th. Pile3	Ap1	0-5	3-5	A, W	2.5Y5/3	1 f, vf, gr, sbk	-	-
	Ap2	5-20	18-20	C, W	2.5Y4/4	3 f, vf, m, abk, sbk	3 vf	Dead vegetable
	Ap3	20-31	8-12	C, W	2.5Y4/4	2 f, vf, m, abk, sbk	3 vf	Dead vegetable, shell fragment
	Bw1	31-44	13-14	C, W	2.5Y4/4	2 f, vf, m, abk, sbk	1 vf	Dead vegetable, shell fragment
	Bw2	44-58$^+$	-	-	2.5Y3/3	1 f, m, sbk	1 f	Dead root, shell fragment
Th. Ctrl	Ap1	0-3	3-5	A, W	2.5Y5/3	1 f, vf, gr, sbk	-	-
	Ap2	3-15	11-13	C, W	2.5Y4/3	2,3 f. vf, m, sbk	2,3 vf	Dead vegetable
	Ap3	15-27	19-21	C, W	2.5Y4/4	2,3 f, vf, m, sbk	2 vf	Small snals
	Bw1	27-34	8-9	C, W	2.5Y4/4	2 f,m, sbk	1 vf	-
	Bw2	34-52$^+$	-	-	2.5Y4/3	2 f,m, abk sbk	1 vf	-

[a] Horizons’ designation according to Ref. [16]

[b] A=abrupt, C=clear, W=wavy

[c] Moist and crushed, according to the Munsell Soil Color Charts

[d] 0= Structureless, 1=weak, 2=moderate, 3=strong; vf= very fine, f=fine, m=medium, co=coarse, gr=granular, abk=angular blocky, sbk=sub-angular blocky, pl=platy, fr= friable

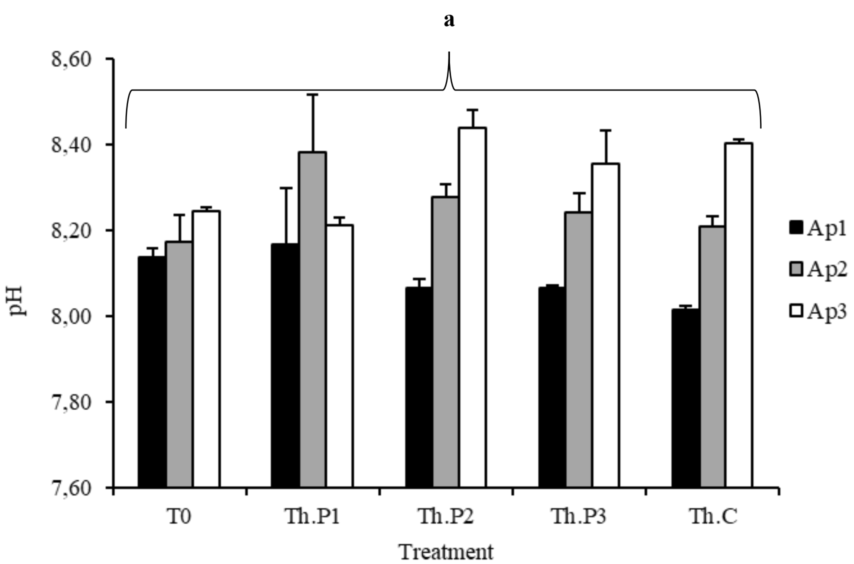
[e] 0=Absent, 1=very few, moderately few, 2=common, 3=many; vf=very fine, f=fine, m=medium, co=coarse.

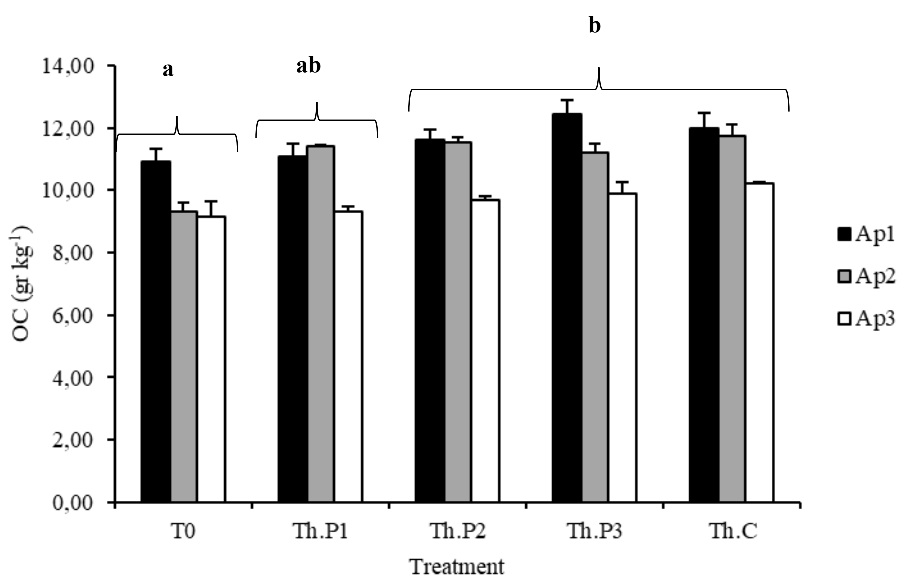
Abbreviations: Pile 1, compost with coffee husk (CH) and brewers’ spent grain (BSG) in proportion 2:1; Pile 2, compost with CH and cow manure (CM) in proportion 4:1; Pile 3, compost with a mixture of CH, BSG, and CM in proportion 5:3:2; Ctrl, control; T0, time before compost distribution and cultivation; T1, time after compost distribution and cultivation; Th, time harvest after wheat harvesting.

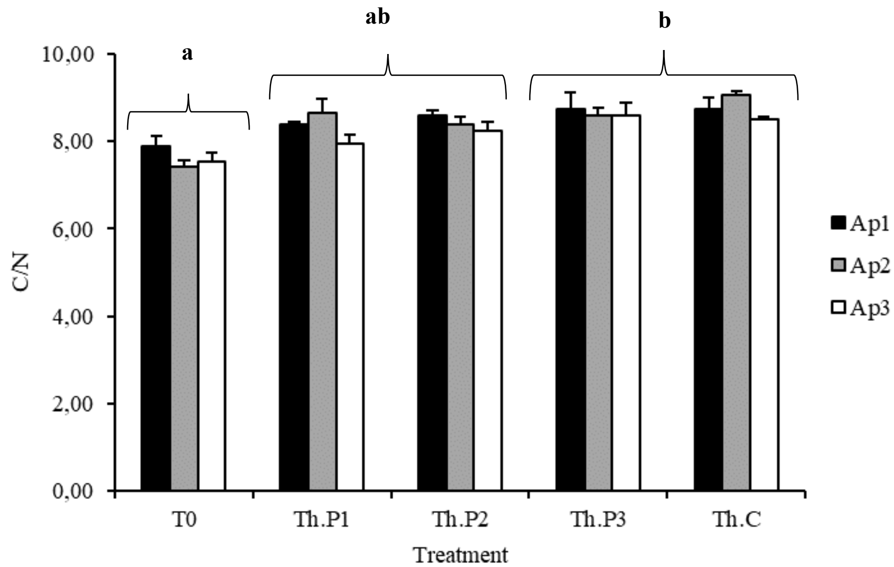
Our observations revealed that there are no significant differences among composts in the terms of pH, total N, and exchangeable Mg (EMg) in the various horizons and times. Pile 1 and Pile 3 composts significantly increased OC content in the Ap2 and Ap1 horizons, respectively, after wheat crops being harvested. The C/N ratio was affected by the application of composts on soils after wheat harvesting when compared to the conditions before compost distribution and cultivation. However, there was no significant difference between soils treated by composts and control soil after wheat harvesting. Available P (AP) content in all compost-treated soil horizons, especially Ap1, decreased significantly when compared to T0 so that the content of this element in the horizon Ap3 showed a tendency to zero. Interestingly, the exchangeable K (EK) level significantly raised in the horizon Ap2 of the soils supplemented with the compost derived from coffee husk and brewers' spent grain in proportion 2:1 (Figure 1).

The measured values of fluorescein diacetate hydrolysis can be compared from two points: the first one (a) is the situation at the onset of the study and the second one (b) the effect of composts throughout the research. FDA amounts were varying (min = 9 μg FDA g^{-1} h^{-1}; max = 26 μg FDA g^{-1} h^{-1}) at the onset of the work in the different horizons. After crop harvesting, the comparison of compost-treated soils displayed that a significant difference can be observed in the soils with the application of Pile 2 and Pile 1/ Pile 3 composts. These observations showed that Pile 2 compost had a positive influence on FDA value (Figure 1). Since the highest fluorescein diacetate value was found in the soils with coffee husk and cow manure, it can be concluded that the composition of compost influences the outputs of the experiment.

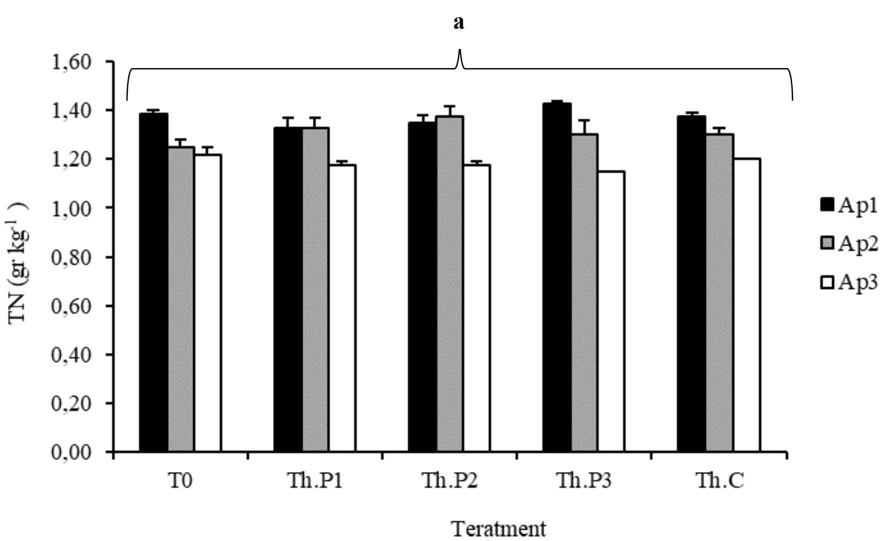
The results confirmed that the fertilization approach, i.e., the application of composts, influenced the microbial biomass carbon (MBC) in the soil (Figure 1). The highest value was found at the end of the study, i.e., in the soil supplemented with the compost derived from coffee husk and brewers' spent grain as compared to the control. The lowest value was measured as the control before compost distribution and cultivation, which was significant in contrast to all other ones. Compared with the other composts, the Pile 2 application therefore had a great positive influence on the microbial biomass carbon, probably due to the composition and higher input of carbon organic into the soil.


a
8,60
8,40
8,20
8,00
7,80
7,60
pH
T0
Th.P1
Th.P2
Th.P3
Th.C
Treatment
Ap1
Ap2
Ap3

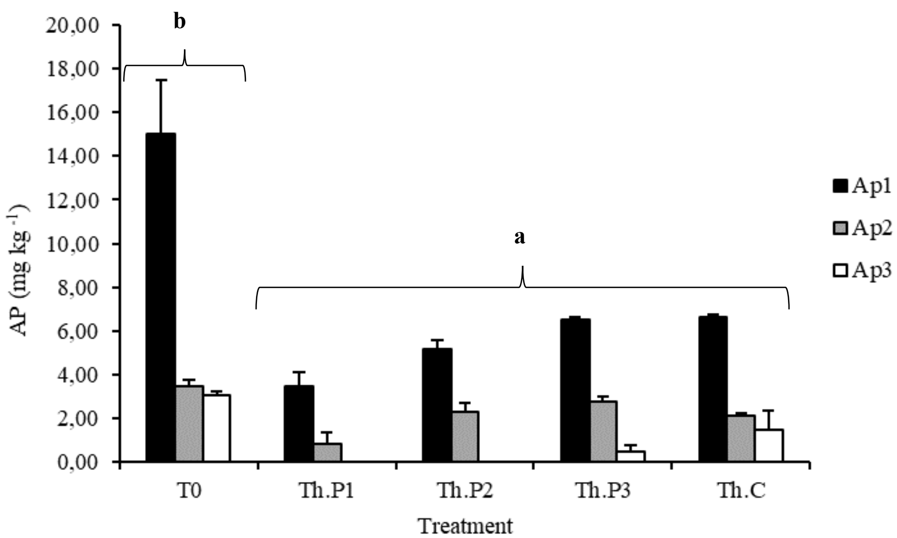

b
a
ab
14,00
12,00
10,00
8,00
6,00
4,00
2,00
0,00
OC (gr kg-1)
T0
Th.P1
Th.P2
Th.P3
Th.C
Treatment
Ap1
Ap2
Ap3

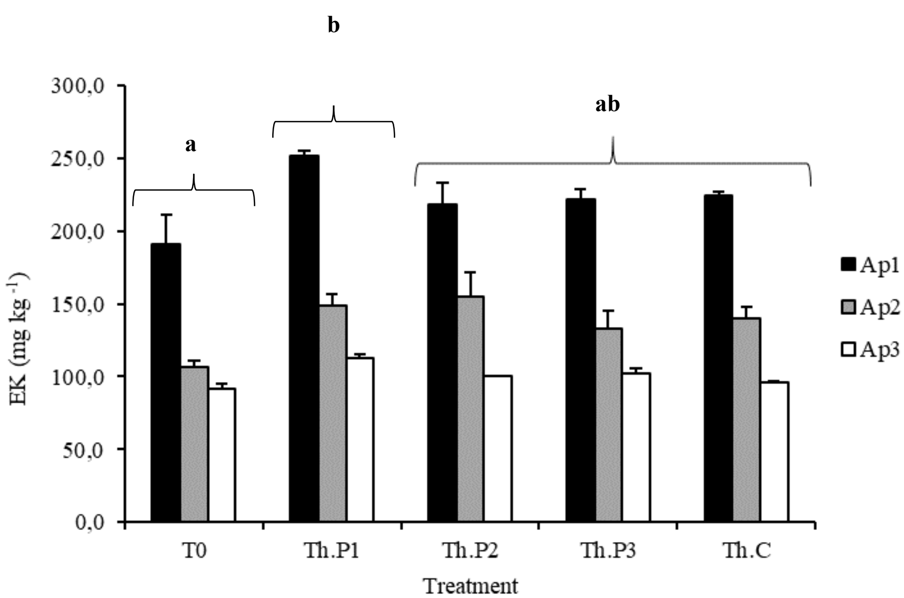


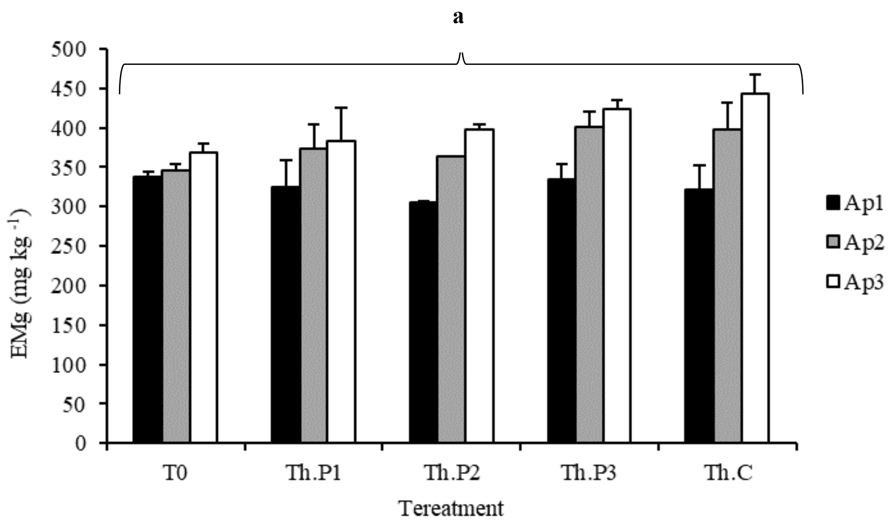
ab
b
a
10,00
8,00
6,00
4,00
2,00
0,00
C/N
T0
Th.P1
Th.P2
Th.P3
Th.C
Treatment
Ap1
Ap2
Ap3

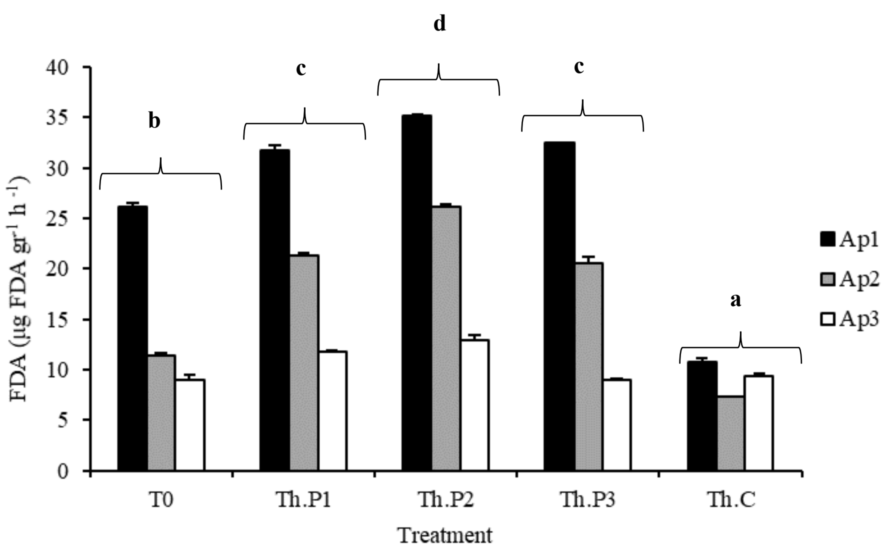


a
1,60
1,40
1,20
1,00
0,80
0,60
0,40
0,20
0,00
TN (gr kg-1)
T0
Th.P1
Th.P2
Th.P3
Th.C
Teratment
Ap1
Ap2
Ap3


b
a
20,00
18,00
16,00
14,00
12,00
10,00
8,00
6,00
4,00
2,00
0,00
AP (mg kg-1)
T0
Th.P1
Th.P2
Th.P3
Th.C
Treatment
Ap1
Ap2
Ap3


b
ab
a
300,0
250,0
200,0
150,0
100,0
50,0
0,0
EK (mg kg-1)
T0
Th.P1
Th.P2
Th.P3
Th.C
Treatment
Ap1
Ap2
Ap3


a
EMg (mg kg -1)
500
450
400
350
300
250
200
150
100
50
0
T0
Th.P1
Th.P2
Th.P3
Th.C
Tereatment
Ap1
Ap2
Ap3


d
c
b
c
a
FDA (µg FDA gr-1 h -1)
40
35
30
25
20
15
10
5
0
T0
Th.P1
Th.P2
Th.P3
Th.C
Treatment
Ap1
Ap2
Ap3

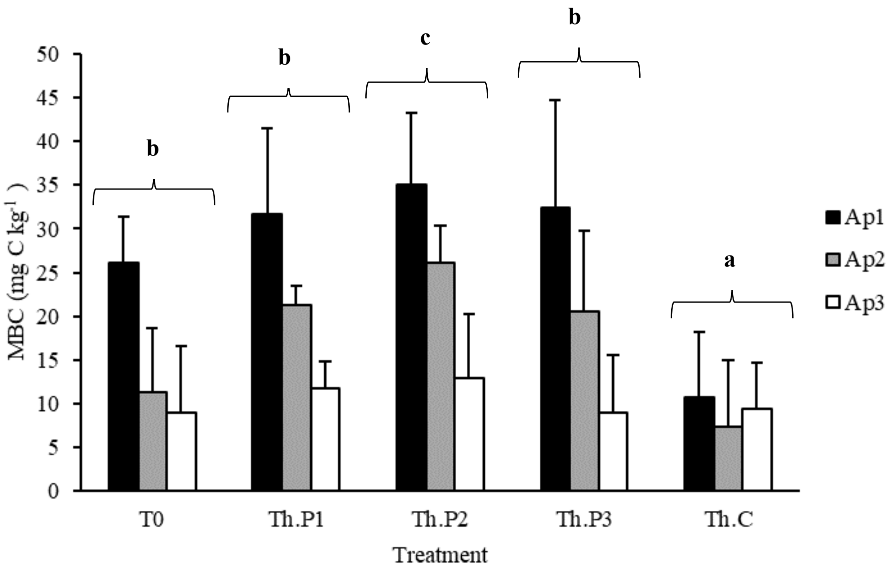


Fig. 1: The analyzed parameters of field soil under treatment of three agri-food derived composts. Abbreviations: OC, Organic carbon; TN, Total nitrogen; CN, C/N; AP, Available P; EK, Exchangeable K; EMg, Exchangeable Mg, pH, MBC, Microbial biomass carbon; FDA, FDA hydrolysis; P1, compost with coffee husk (CH) and brewers' spent grain (BSG) in proportion 2:1; P2, compost with CH and cow manure (CM) in proportion 4:1; P3, compost with a mixture of CH, BSG, and CM in proportion 5:3:2; Ctrl, control. T0, time before compost distribution and cultivation; Th, time harvest after wheat harvesting.

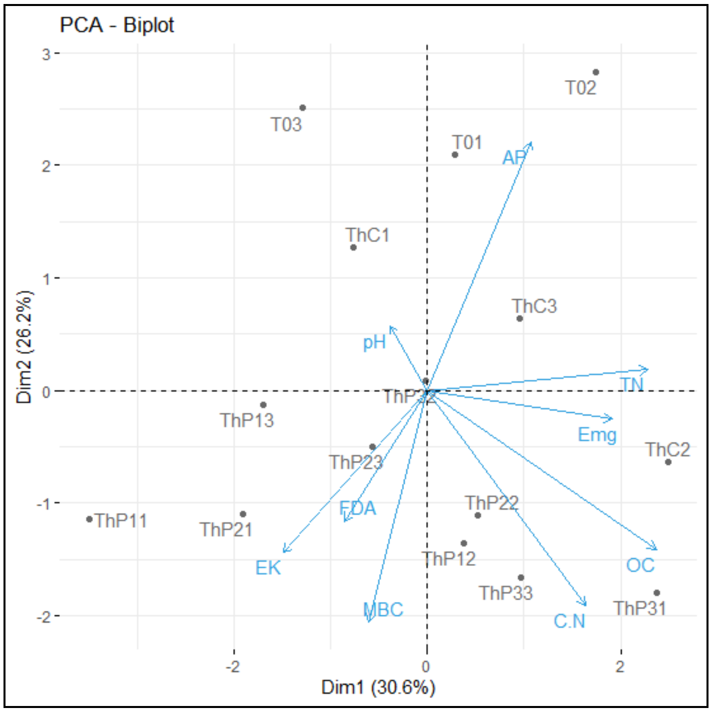
3.2. Principal components analysis of soil traits

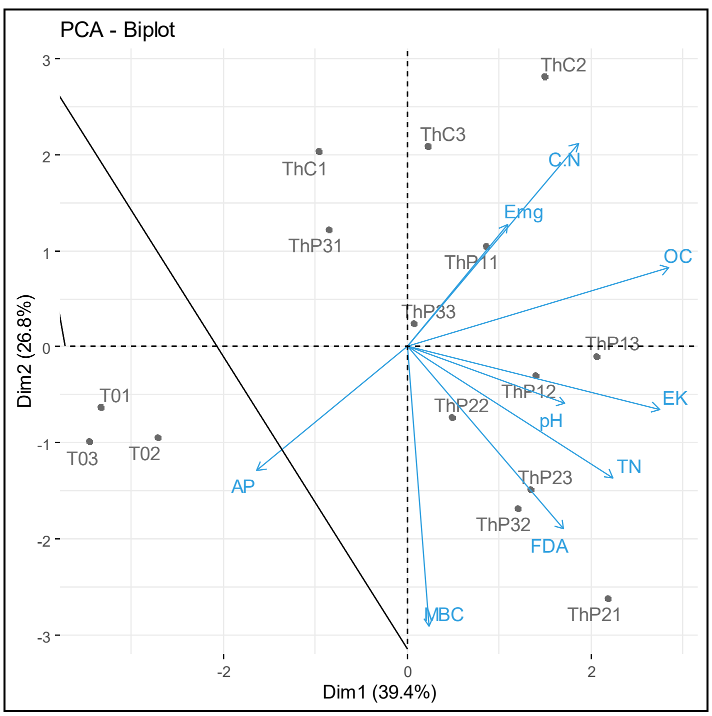
To decline data dimensionality as well as achieve a better picture of the effect of three composts on wheat yield throughout the horizons and soil horizons physic-chemical features, PCA was accomplished (Figure 2; Table 2). In the terms of Ap1 horizon, the first two components, explaining 56.8% of the total variance, were considered as favorable representative of basic information. The first principal component accounted for the main part of the whole variation with 30.6 % and the second was 26.2%. The variables contribution and correlation on the PC1 and PC2 were displayed in Figure 3 and Table 3. The first and second components accounted for the highest percentage of variations in data, including AP, TN, and EMg. In the terms of Ap2 horizon, the first two components explained 66.2% of the total variance. The PC1 accounted for 39.4% of whole variation and the PC2 for 26.8%. The PC1 and PC2 accounted for the highest percentage of variations in data, including CN, OC, and EMg. In the terms of Ap3 horizon, the first two components, explaining 58.5% of the total variance, were registered as favorable preventative of important data. The first principal component accounted for the main part of the whole variation with 33.2% and the second was 25.3%. The first and second components accounted for the highest percentage of variations in data, including CN, OC, and EMg.

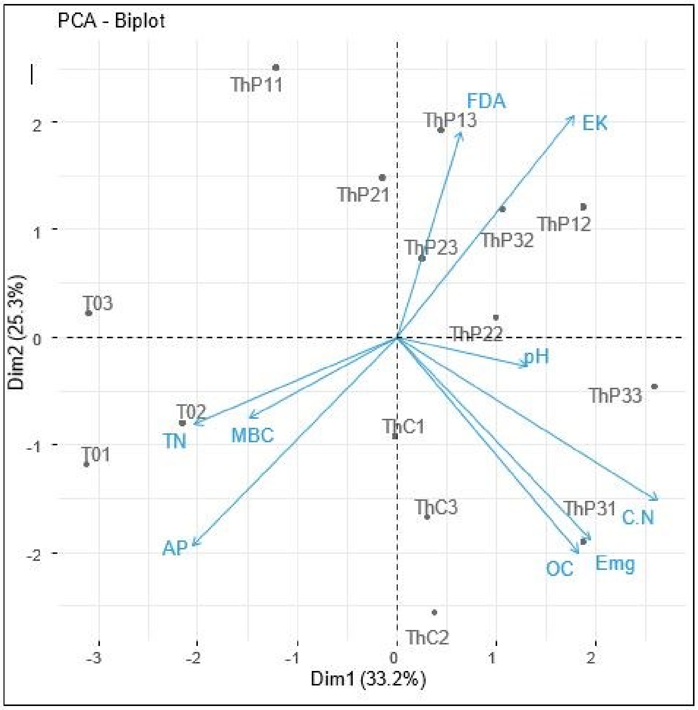
Table 2. Principal component analysis for physic-chemical features over different horizons

Variable	Ap1		Ap2		Ap3	
	PC1	PC2	PC1	PC2	PC1	PC2
pH	-0.1388ns	0.2052ns	0.5561*	-0.1909ns	0.4138*	-0.083ns
OC (gr kg^{-1})	0.8424**	-0.5016*	0.9213**	0.2652ns	0.5772*	-0.6309**
C/N	0.5822*	-0.680**	0.6012**	0.6852**	0.8282**	-0.4804*
TN (gr kg^{-1})	0.8106**	0.0664ns	0.7240**	-0.4389*	-0.6459**	-0.255ns
AP (mg kg^{-1})	0.3810*	0.7854**	-0.5277*	-0.4143*	-0.651**	-0.614**
EK (mg kg^{-1})	-0.53088*	-0.5104*	0.8881**	-0.208ns	0.5616*	0.6494**
EMg (mg kg^{-1})	0.6804**	-0.0903ns	0.3529*	0.4112*	0.6163**	-0.5957*
FDA (μg FDA gr^{-1} h^{-1})	-0.3055*	-0.4128*	0.5508*	-0.6129**	0.203851ns	0.6054**
MBC (mg C kg^{-1})	-0.2168	-0.73225	0.078049	-0.93967	-0.4687	-0.236ns

ns: not significant, *significant (α =5%), ** significant (α =1%)


PCA - Biplot
Dim1 (30.6%)
Dim2 (26.2%)
T02
T03
T01
AP
ThC1
ThC3
pH
TN
ThP32
ThP13
Emg
ThC2
ThP23
FDA
ThP22
ThP11
ThP21
EK
ThP12
OC
ThP33
ThP31
MBC
C.N
Ap1


PCA - Biplot
Dim1 (39.4%)
Dim2 (26.8%)
ThC2
ThC3
ThC1
C.N
Emg
ThP31
ThP11
OC
ThP33
ThP13
T01
ThP12
EK
ThP22
pH
T03
T02
TN
ThP23
AP
ThP32
FDA
MBC
ThP21
Ap2



Ap3

Fig. 2: Principal component analysis (PCA) for physic-chemical features over different horizons and times. Abbreviations: OC, Organic carbon; TN, Total nitrogen; CN, C/N; AP, Available P; EK, Exchangeable K; EMg, Exchangeable Mg; pH, Ph; MBC, Microbial biomass carbon; FDA, FDA hydrolysis. T0, time before compost distribution and cultivation; Th, time harvest after wheat harvesting.

3.3. Correlation of soil traits

From our results, there is a strong positive correlation between organic C and total N in the soil. The exchangeable K exhibited a strong negative correlation with pH and exchangeable Mg. In the other part of the story, there is no strong correlation between microbial biomass carbon and C/N ratio in the soil (Figure 3).

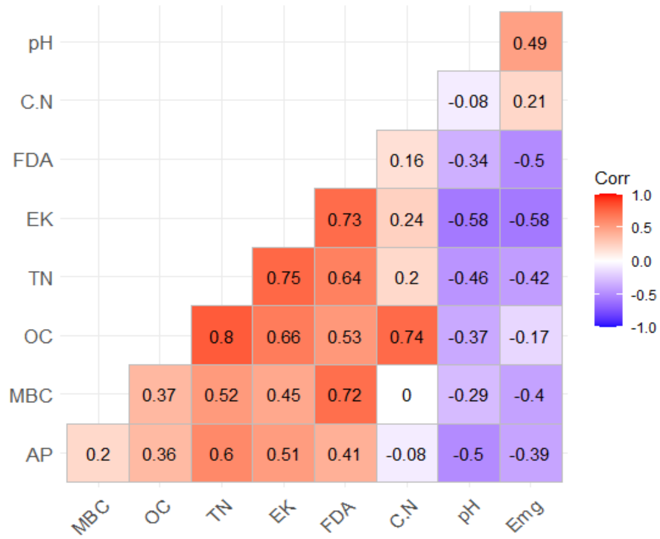


Fig. 3: The correlation among analyzed parameters of field soil. Abbreviations: OC, Organic carbon; TN, Total nitrogen; CN, C/N; AP, Available P; EK, Exchangeable K; EMg, Exchangeable Mg; pH, Ph; MBC, Microbial biomass carbon; FDA, FDA hydrolysis.

3.4. Wheat yield

From our observation, the composts with coffee husk (CH) and brewers' spent grain (BSG) in proportion 2:1 (Pile 1), CH and cow manure (CM) in proportion 4:1 (Pile 2), and a mixture of CH, BSG, and CM in proportion 5:3:2 (Pile 3) improved the wheat grain yield in the field. The highest yield of wheat was observed in the compost with coffee husk and brewers' spent grain (Pile 1), ~5.2 ton ha^{-1} on average. Moreover, the lowest yield of crop biomass was registered in the control without the compost application, ~3.6 ton ha^{-1} on average (Figure 4). Totally, wheat grain yield ranged from ~3.3 ton ha^{-1} (control) to ~5.2 ton ha^{-1} (Pile 1 treatment).

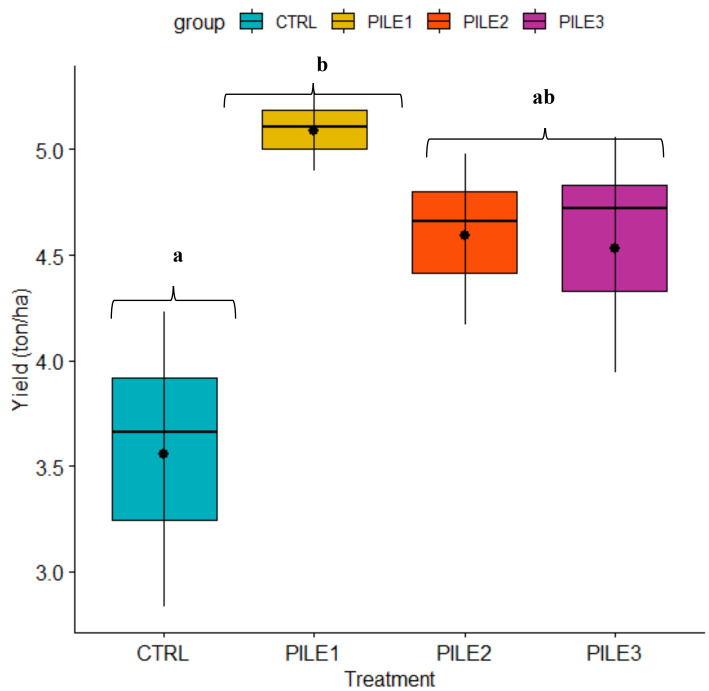


Fig. 4: The wheat yield in the field under treatment of three agri-food derived composts. Abbreviations: Pile 1, compost with coffee husk (CH) and brewers' spent grain (BSG) in proportion 2:1; Pile 2, compost with CH and cow manure (CM) in proportion 4:1; Pile 3, compost with a mixture of CH, BSG, and CM in proportion 5:3:2; Ctrl, control.

3.5. Lipid fractions of Wheat

Lipids accounted for approximately ~1,1% of the dry weight of wheat treated with three composts. Of the lipids extracted from wheat in the recent work, C18:2 n-6, C18:1Δ9c, C16:0, and alpha-18:3 were the most abundant types, accounting for an average of 95% of the fatty acids analyzed. These were followed by Vacc, C18:0, and C20:1Δ11, which accounted for averages of 1,3%, 1,1%, and 0,97% of the total fatty acids, respectively. No main effects of composts were observed for fatty acids. However, Pile 1 increased the C18:1Δ9c and C16:0 level more than other composts (Table 3).

Table 3. Lipid fraction of wheat plants treated by three agri-food derived composts

Lipid Fraction	Pile1 (Mean± Stdev.)	Pile2 (Mean± Stdev.)	Pile3 (Mean± Stdev.)	Ctrl (Mean± Stdev.)
C12:0	0,10±0,08	0,02±0,00	0,03±0,02	0,02±0,00
C14:0	0,14±0,01	0,13±0,01	0,13±0,01	0,14±0,01
C15:0	0,10±0,01	0,11±0,02	0,12±0,02	0,10±0,01
C16:0	18,73±1,96	15,98±0,39	17,08±0,50	17,94±1,25
C16:1	0,23±0,01	0,19±0,01	0,19±0,01	0,20±0,01
C17:0	0,11±0,01	0,09±0,02	0,11±0,02	0,09±0,01
C17:1Δ10	0,04±0,02	0,02±0,02	0,04±0,01	0,02±0,01
C18:0	1,30±0,74	0,49±0,05	0,43±0,03	0,38±0,03
C18:1Δ9c	22,50±1,69	20,46±0,17	20,62±0,08	20,95±0,27
Vacc	1,14±0,26	0,88±0,03	0,88±0,04	0,87±0,06
C18:2 n-6	51,63±0,08	55,75±0,15	54,86±0,35	53,90±0,81
alpha-18:3	4,23±0,21	4,77±0,06	4,51±0,16	4,37±0,15
C20:1Δ11	0,97±0,05	1,14±0,08	1,02±0,05	1,07±0,09
Fat%	1,10±0,17	1,11±0,03	1,13±0,01	1,19±0,09

4. DISCUSSION

4.1. Soil physicochemical features

Giving compost influence on soil nutrients, the application of composts such as coffee husk and brewers' spent grain (Pile 1), coffee husk and cow manure (Pile 2), and a mixture of coffee husk, brewers' spent grain, and cow manure (Pile 3), especially the Pile 1, can induce enhancing effect on most soil physic-chemical features studied. Our findings supported the observations of other researchers who exhibited that the use of these natural materials raised the soil fertility. For instance [6] observed that by applying the coffee husk compost, 2-3 kg plant^{-1} year^{-1} in the course of three years increased 20% to 30% over mineral fertilizers. They displayed that coffee growth, nutrient levels, and soil fertility were improved relative to plots that no using the compost. Awopetu et al., (2015) [7] also demonstrated that compost derived from brewer's spent grains can increase maize yield. Bouajila and Sanaa (2011)[17] also documented that the use of cow manure compost led to an increment of organic carbon, with the compost treatment being the most efficient. Totally, it can be concluded that the organic compost application can improve soil fertility and thereby crop yield.

The composts clearly increased nutrients and organic C content. Since the composts contain a large amount of valuable nutrients such as C, S, Mg, Ca, K, P, and N and also a number of crucial trace elements [18], thereby they can be specified as multi-nutrient organic fertilizers [19]. Notably, the nutrient level and other key chemical features, like C/N ratio, are depend on the compost processing conditions and utilized organic feedstock. By a suitable mix of compost materials, nutrient-rich composts can be created which serving as a substitute for commercially available mineral fertilizers in farming [1]. Of course, we must point out that, total nutrient content of composts is not plant-available fully at once. This can be attributed to the presence and intensity of binding forms within the organic matrix, leading to an incomplete nutrient immobilization [20]. Besides, the fertilization influence will last longer owing to a gradual and slow release of nutrients to the soil [21]. Thus, with the application of the composts, especially Pile 1, there is much better conservation from leaching over mineral fertilizers. Particularly, the P and N fertilization influence of composts is restricted, as observed in our work, owing to low microbial immobilization and mineralization rate [20][22].

The cation exchange capacity is concerned as one of the useful parameters for assessing the fertility of the soil, specifically for mineral keeping and thereby it hampers cations from leaching into the water underground. In recent work, the exchangeable K level raised significantly in the soil supplemented with the Pile 1 compost. In line with our observation, Agegnehu et al.

(2014)[18] observed that the compost amendment led to an increment of cation exchange capacity owing to the input of organic matter being rich in functional groups into the soil.

Soil pH is an effective factor for crop productivity since many soil organisms and crops contain a tendency for slight acidic or alkaline situations and therefore it affects their growth. Besides, pH influences the accessibility of nutrients in the soil. In the current experiment, the application of composts has an insignificant, restricting efficacy owing to its richness in alkaline cations like exchangeable Mg which were liberated from organic matter owing to mineralization [18].

Compared with the other composts, the use of Pile 2 experienced a higher positive effect on the microbial biomass carbon (MBC) value, perhaps owing to its composition and input of organic carbon into the soil. In other words, the microbial composition and MBC, from the narrower taxonomic level to the broad functional level (e.g., fungi and bacteria) usually respond to organic carbon resources [23][24][25]. Thus, the composts apply a stimulation effect on both the soil-born microbiota and the microbial biomass carbon. This finding is quite interesting and can be explained on the basis of observations reported by Ref. [8] who showed that the MBC level in the soil was influenced not only by the compost application but likely by the activity of crop roots and their exudates into the soil.

The enzymes available in soil were utilized as sensitive parameters of soil productivity, quality, and fertility. Thus, their activities reveal microbial activity and thereby accessibility of nutrients for plant uptake [26]. FDA is carried out by many enzymes like lipases, proteases, and esterase [27]. Besides, FDA can be utilized as a parameter of microbial activity relative to the soil environment state [28]. When the FDA values were evaluated, we can conclude our findings display a positive efficacy of organic fertilizers, composts especially Pile 2, on the total microbial activity. As observed by Ref. [29], the use of composts increased microbial activity as compared to the control soil. The authors reported that the microbial activity was ~2.5- fold more in the compost-supplemented soil when compared with the control soil.

Totally, our observations confirmed that the use of coffee husk and brewers' spent grain compost (P1), can improve the nutrient status in the soil. Besides, due to its multiple positive impacts on the biological and physic-chemical features of soil, this compost could contribute to the improvement of wheat production. It also seems that the compost possesses an equalizing influence of the accessibility of crop to nutrients, heat and air balance of soil, and seasonal/annual fluctuations concerning water, thus the ultimate wheat yield. Moreover, because of the slow release of minerals and their accessibility in compost-mediated fertilization, this compost can obtain the favorable output. Therefore, for achieving the long-term goals in sustainable agriculture, composting can be a rational item for establishing effective management of plant-nutrient in the world [19].

4.2. Wheat yield

From our observations, wheat grain yield increased from ~1.3-fold (Pile 1 and Pile 2 treatment) up to ~1.6-fold (Pile 1 treatment) by the application of compost. The improvement influence of Pile 1 compost may be induced by higher levels of plant minerals such as N, P, K, and Mg and root augmentation resulted from the compost using [5]. Besides, this compost may increase the available form of nutrients for the crop in soil and then improves nutrient uptake by root growth that leads to crop weight and height rise [30]. Similar findings have been reported by Ref. [31] who demonstrated that productivity of crops treated with 100 ton ha^{-1} compost enhanced significantly by ~80% in comparison with the control. The enhanced influence of compost on yield and agronomical traits was also documented by Ref. [22] who exhibited an increment in the grain yield of wheat cultivars relative to the control via ~13 kg of compost. Moreover, Nadjet *et* *al.* (2014) [30] observed that compost can lead to the best-marked development of wheat crops and better performance in the number/weight of spikes and the number/weight of seeds when compared to the control. As for wheat, it has been reported that maize yield was enhanced significantly by ~105 and ~125 % due to use of compost at the rate of 5 and 7 ton fed^{-1}, respectively, relative to the control [32]. Gamal (2009) [33] also demonstrated that the use of compost (5 ton ha^{-1}) raised the productivity of sorghum by 45% in comparison with no compost application. Notably, the application of compost does not only improve the grain yield of crops in terms of field yield, but it can be also improved that the quality of their products is affected in a positive manner [20]. Totally, it can be interesting to add an adequate amount of Pile 1 compost for a better increment of wheat productivity.

4.3. Lipid fractions of wheat

There is no significant difference among the composts for the total content of fatty acids. Anyway, the C18:2 n-6, C18:1Δ9c, C16:0, and alpha-18:3 was found as the most abundant types, accounting for an average of 95% of the fatty acids evaluated. Slight differences in the content and composition of fatty acids among the compost-treated wheat crops can be derived from the genotypic structure and expected to affect the processing features of the wheat products [34]. Some researchers have also demonstrated a difference between the lipid profiles of wheat cultivars [35]. Albeit our work is the most detailed research so far published on the efficiency of natural composts on the wheat lipid profile, it must be stated that only a single harvest year was evaluated. Consequently, year-to-year differences among samples due to environmental changes should be considered, including those associated with agronomy practices and genotypic structure. Therefore, taking advantage of diversity in processing quality resulting from the content and composition of fatty acids by wheat breeders has a challenge.

5. CONCLUSION

On the basis of the above observations, it can be inferred that the application of composts, especially the compost derived from coffee husk and brewers' spent grain, had a great influence on soil physicochemical features and wheat yield. Consequently, it can be suggested to integrate or, in particular situations, substitute the mineral fertilization to achieve high soil fertility and wheat productivity. Finally, our work reveals a procedure of using agricultural wastes to generate compost and likely will contribute to reducing environmental pollutions in eco-sustainable agriculture.

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CHAPTER5

Effect of Coffee-husk based compost on yield and lipid fraction of *Cannabis sativa L.*

ABSTRACT

Coffee husk supplemented with brewers' spent grain (BSG) and cow manure (CM) in three different proportions and mixture were studied. The prepared compost distributed two times in *Cannabis Sativa L.* field. Finally, the seeds collected, and lipid fractions extracted through GC-MS. Results showed, cannabis yield increased from ~367 kg per ha (control) to ~567 kg per ha (Compost 1). No main effects of composts were observed for fatty acids. Soil features were improved by the application of composts, especially the compost derived from coffee husk and brewers' spent grain. Moreover, this compost had positive influence on the microbial enzyme activate and biomass, probably due to the composition and higher input of carbon organic into the soil. As a result, it can be suggested to use this compost to reduce environmental pollution, improve soil fertility, and achieve high production of cannabis crop.

Key Words: Compost; *Cannabis sativa L.*; Lipid Fraction; Soil Features.

1. INTRODUCTION

Recently, the interest on *Cannabis sativa L.* has drastically increased. However, the main attention is generally addressed to psychoactive and non-psychoactive compounds, such as D9-tetrahydrocannabinol (D9-THC) and cannabidiol (CBD) [1]. In the past, the genus cannabis was allocated into three main species: drug-type (*C. indica*), with high levels of D9-THC, a fiber-type (*C. sativa*) with low levels of D9-THC and an intermediate one *C. ruderalis Janish* [2]. Recently, it was decided to classify all different species as *C. sativa* also called "hemp" when referred to industrial use (fiber-type), or "therapeutic" also called "marijuana" (drug-type) for the variety with high content of D9-THC (>0.6%; w/w). To date, the main use of hemp is largely related to food; in fact, hemp seeds are generally used for producing oil and flour and, depending on the countries local regulations, they could or not be employed on the basis of their pharmacological properties [3]. However, hemp contains more than 500 different cannabinoids, of which about ten have been classified according to their chemical structure, such as D9-tetrahydrocannabivarin (THCV), cannabidiol (CBD), cannabigerol (CBG), D8-tetrahydrocannabinol (D8-THC), D9-tetrahydrocannabinol (D9-THC), cannabichromene (CBC), cannabinol (CBN), cannabidiolic acid (CBDA), D9-tetrahydrocannabinolic acid (THCA) and cannabigerolic acid (CBGA) [4].

Cannabis can be a suitable crop for alternative agricultural practices such as organic agriculture, due to natural resistance to many pathogens and herbivores. The popular belief is that cannabis is a relative low input crop and adaptable to marginal soils [5]. Essential plant nutrients such as N, P and K from synthetic fertilizers are commonly used by conventional farmers, however there are other potential fertilizer products originating from organic and biological sources. Vermicompost extract stimulated seed germination, hypocotyl and radicle growth as well as increased chlorophyll content in cannabis [5]. Conant *et al.,* (2017) [6] tested a bioinoculant on cannabis and in addition to greater bud yield bioinoculant application also led to significant increases in plant height and basal steam area. Plant biomass increase has been documented in response to the application of humic acid [7] and biofertilizers [8][9]. Werse (2016) [10] found that 43% of cannabis growers from Germany, Austria and Switzerland confirmed to have ecological ideology activist reasons to grow cannabis.

Composting can be an environmentally safe, cost-effective, and efficient biological procedure for recycling residual biomass in agriculture [11]. Several advantages have been suggested for the application of compost as fertilizer, like enhancing microbial activity and organic content [12].

High content of nutrients such as K, P, and N can be obtained from compost usage [13]. Moreover, from [14], it has been recorded that natural compost implementation has a promoting influence on wheat yield, agronomical traits, and grain nutrient content than chemical fertilization. Therefore, it can be suggested to substitute chemical fertilizers with organic compost to achieve the high production of this crop.

As a natural product, coffee husk possesses tannins and caffeine, which can make it slow degradation toxic and, in the environment, leading to the disposal challenge. Nevertheless, this compost is rich in lignocellulose materials, which make it a favorable substrate for microbial activities [15]. Multiple, alternative applications of coffee husk have been suggested because it improves the biological and physic-chemical properties of soil, enhances soil organic matter, and decreases the depletion of natural sources [15]. Brewer's spent grains, a by-product of the brewing industry that makes up %80 of brewing waste, can be utilized as soil fertilizer alongside material recently known as wastes [16]. As reported previously, these materials in suitable mixtures by microorganisms can be added to soil. Moreover, the compost derived from brewer's spent grains can compete admiringly with chemical fertilizers in terms of crop yield in the field [16]. Cow manure, comprising a large amount of K, P, and N endangers human health and has become the main contaminant in environment due to inadequate application [17]. Aerobic composting can stabilize organic nutrients in manure and keep down their harmful influences through fungi and bacteria [18][19]. As above-mentioned, composting presents a promising tool for converting livestock manure into a nutrient source for horticultural and agricultural applications [17].

With such situations in mind, this work was aimed at the comparison of three types of composts [coffee husk (CH) and brewers' spent grain (BSG) in proportion 2:1 (Compost 1), CH and cow manure (CM) in proportion 4:1 (Compost 2), and a mixture of CH, BSG, and CM in proportion 5:3:2 (Compost 3)] on different soil properties and hemp yield to produce agri-food-derived compost, which improving the soil features and hemp performance in the field.

2. METHOD AND MATERIALS

2.1. Composting and experimental design

The current study was focused on the comparison of composts with coffee husk (CH) and brewers' spent grain (BSG) in proportion 2:1 (Compost 1), CH and cow manure (CM) in proportion 4:1 (Compost 2), and a mixture of CH, BSG, and CM in proportion 5:3:2 (Compost 3). The dimension of every compost pile was 1 m^3, and the weight of its materials was around 300 kg. All piles were covered with a plastic film in order to prevent excessive moisture loss and preserved in the greenhouse. The aerated static pile was followed as the composting method [20]. To aerate the pile, layers of loosely piled bulking agents like wood chips were added so that air could pass from the bottom to the top of the pile. Compost made in three months and sampling was done four times (T) in the first day and subsequently every month. Compost samples preserved in -20^{0}C fridge and named as T1, T2, T3 and T4.

The field preparation was performed in the April 2019 in Agugliano, Marche region (Coordinates: 43°33′N 13°23′E; Elevation: 203 m), Italy, and around 15 kg of compost were distributed two times (before cultivation and in four leave level) in any plot with 18 m^2, and hemp seeds cultivated in the arranged field by Completely Randomized Design (CRD) method in three replications. The total dimension of the field was 216 m^2, with margin and corridor was 513 m^2. Totally, 12 blocks with 18 m^2 dimensions were considered. Soil samples were collected at T0, T1 and Th (Time harvest) in 50 cm depth in three horizons (Ap1, Ap2, and Ap3) and then air-dried at room temperature, grinded and sieved at 2 mm, and stored at 4 °C for further soil analysis.

2.2. Experiments methods

Germination index (GI %) of compost was evaluated by the germination test of Chinese cabbage [21]. Microbial biomass carbon (MBC) in soil was estimated through the fumigation extraction procedure [22]. Hydrolysis of fluorescein diacetate (FDA) in soil was determined as reported by [23].

2.3. Statistical analysis

To investigate the relationship between depths and soil variables, the repeated measure analysis was used along with Tukey's range post hock test ($P < 0.05$) in the SPSS V. 25 software. The same analysis was done on compost parameters too. To compare hemp product, the one-way ANOVA followed by Tukey's range test ($P < 0.05$) were used in the R software. Before the ANOVA, the dataset was evaluated for variance homogeneity and distribution normality by Levene's and Shapiro-Wilk tests, respectively, both at a 5% significance level.

3. RESULTS AND DISCUSSION

3.1. Compost analysis

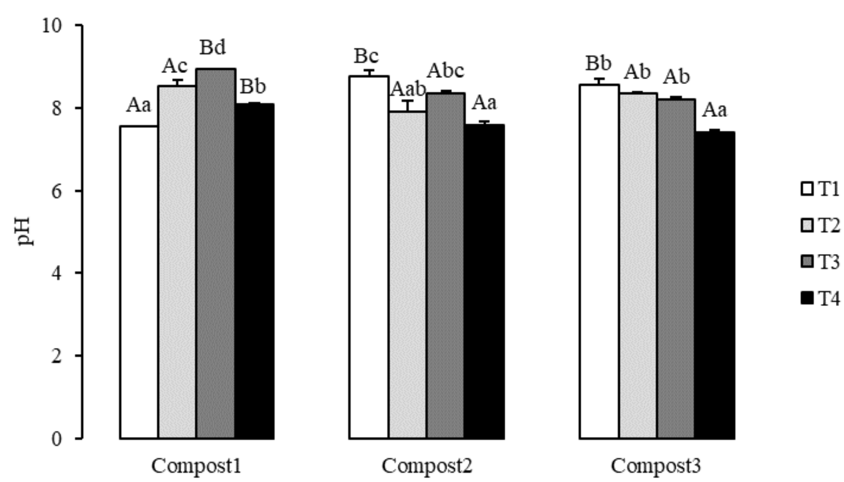
3.1.1. General characters of compost (C/N, pH)

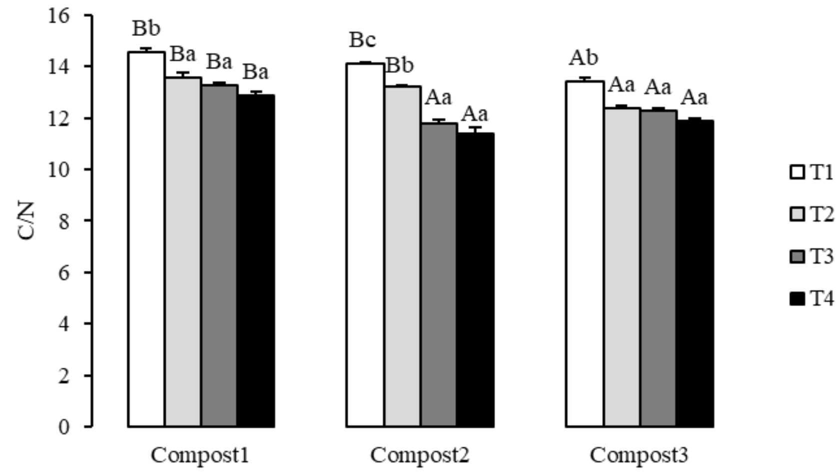
As observed in the recent work, pH clearly differed in three composts (Fig. 1). The pH value for each compost pile on the 0^{th} day was between 7.5 and 8.5. The pH remarkably increased up to 60^{th} day for C1, however, those of C2 and C3 remained in neutral-phase values or sequentially decreased. The C1 treatment displayed a max pH registered ~9 on the 60th day of study. The initial pH (~7.5) was increased in the C1 treatment up to the 60th day with a subsequent decrement at the end of compost production (90th day). The increased pH from the 0th to 60th day revealed protein breakdown in the organic wastes with the release of NH_3 [24]. Besides, this alkaline pH maybe because of the faster biosynthesis of phenolic metabolites within the degradation time [25]. However, pH was decreased in the 90th day, i.e., at the end step of decomposition. All the accounts, this is suggestive of a fast degradation of organic materials (C1, C2, C3) with regard to the course of composting. As stated by [26], for efficient composting, natural wastes with low pH values in the 90th day of composting (such as C2 and C3 in this work) must be blended with other materials achieving suitable situations so that the initial pH of the substrate for compost production can be considered in the ~7.5 value.

In compost production, the C/N ratio is important so that if this ratio being high at the onset of the process, the decomposition of organic materials will be slower. The soil generally comprises a C/N ratio of 10:1. Data for the effect of temperature variations on the C/N ratio within the composting process has been shown in Fig. 1. The findings demonstrated that the C/N ratio, especially C2 and C3, was better than all the other treatments. Throughout three months, the C/N ratio decreased from ~14.5 (0th day) to ~12.5 for the treatment C1 followed by C3 and C2. Our observations are indicative of an acceptable maturity (C/N ratio of 10 up to 15) [27]. The lower reduction of the C/N ratio from 30th up to 60th day in the C1 revealed more evolution of CO2 owing to C transformation [26]. Besides, the drop in moisture level may reduce the metabolic reaction of microorganisms, leading to an increased C/N ratio. Interestingly, we observed that although the highest decrement in the ratio of C to N was recorded on the last day of the experiment. It is recommended that for efficient composting with a lower period of time, the primary acclimatizes must focus on the initial steps of composting activity [28] and the finding is in line with that of [29] and [30].

3.1.2. Germination Index (GI%)

Quality control throughout the composting must ensure sufficient physical and chemical features, as well as a sufficient degree of maturity and stability [26]. An appropriate assessment of compost maturity is imperative for the successful use of agri-food wastes in agriculture. The available analyses differ in approach, duration, costs, simplicity, and precision. However, bioassays are concerned the most direct procedure for maturity because it displays the influence of compost maturity on plant growth [26]. Overall, the index maturity decreased from the first to the 30th day, and then from the 30th day to the 90th day with an increase in all three composts (Fig. 1). The highest GI % was observed in C1 (77.34%) followed by C3 and C2 (70.48 and 66.20%). This is in agreement with findings observed by [31], indicating that the decomposition had slow down, and thereby the composts obtained maturity.





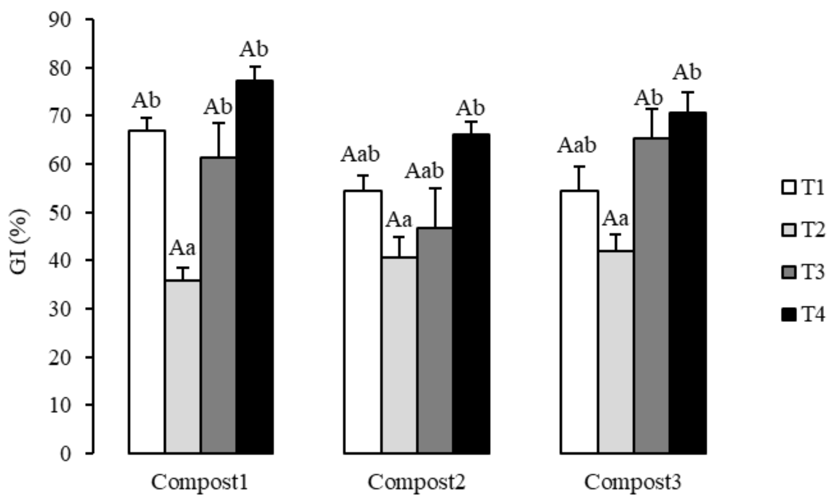


Fig. 1: The analyzed parameters of three agri-food derived composts. Abbreviations: Compost1, coffee husk (CH) and brewers' spent grain (BSG) in proportion 2:1; Compost 2, CH and cow manure (CM) in proportion 4:1; and Compost 3, a mixture of CH, BSG, and CM in proportion 5:3:2.

3.2. Soil physic-chemical features

3.2.1. Soil description

Based on the description of soil in the hemp field in the different horizons and times, at T0, there was a diffuse existence of the mesofauna at the soil surface. The soil was specified by three Ap horizons, displaying hard rupture-resistance related to brittleness and firmness. In T0 the Ap horizons were 0-51 cm thick. The structure was moderately developed, with friable platy aggregate in the Ap horizon. The color of soil ranged from reddish yellow in the upper horizon to light brown in the lower one. Very few fine roots were found to the Ap horizons. Depth and thickness of Ap1 and Ap2 horizons had a tendency to reduce in the soils supplemented with the composts after the crop harvesting. Overall, our findings revealed that the application of composts, especially the compost derived from coffee husk and brewers' spent grain, had remarkable effects on the soil profile (Table 1).

Table1: Description of soil in the wheat field in the different horizons and times

NAME	HORIZON[a]	Depth (cm)	Thickness (cm)	Bnd[b]	Matrix colour[c]	Structure[d]	Root[e]	Other observation
T0	Ap1	0-5	3-5	C, W	2.5Y5/4	3, f, m, abk, sbk, co	1, vf, f	-
	Ap2	5-34	-	C, W	2.5Y5/3	3, f, m, abk, sbk, co	1, vf, f	Vegetable residue
	Ap3	34-51	-	C, W	2.5Y5/4	3, vf, m, abk, sbk, co	-	shell
T1.C1	Ap1	0-10	10-12	C, W	2.5Y3/3	3, m, sbk, gr	1, vf, f	-
	Ap2	10-27	16-17	C, W	2.5Y4/3	3, m, abk, sbk	1, vf, f	-
	Ap3	27-44	-	C, W	2.5Y4/2	3, m, abk, sbk		-
T1.C2	Ap1	0-14	10-17	C, W	2.5Y4/3	3, f, m, sbk	1, vf, f	Dead wood
	Ap2	14-28	7-13	C, W	2.5Y4/4	3, m, abk, sbk	1, vf, f	sckeleton
	Ap3	28-43	-	C, W	2.5Y4/3	3, m, sbk		-
T1.C3	Ap1	0-11	11-16	C, W	2.5Y5/4	3, f, m, abk, sbk, Co	1, vf, f	Dead cane
	Ap2	11-28	12-16	C, W	2.5Y4/4	2, f, m, abk, sbk, Co	1, vf, f	-
	Ap3	28-45	-	C, W	2.5Y4/3	3, m, abk, sbk, Co		shell
T1.Ct	Ap1	0-4	4-8	C, W	2.5Y5/3	3, f, vf, abk, sbk, Co	1, vf, f	-
	Ap2	4-34	10-12	C, W	2.5Y4/3	2, f, vf, abk, sbk, Co	1, vf, f	Vegetable residue
	Ap3	34-46	-	C, W	2.5Y4/3	3, f, vf, abk, sbk		Shell, Vegetable
Th. C1	Ap1	0-6	4-8	C, W	2.5Y4/4	3 f, m, sbk, gr	-	-
	Ap2	6-38	20-23	C, W	2.5Y4/2	2 f. vf, m, sbk	2 vf	-
	Ap3	38-40	10-15	C, W	2.5Y4/3	2 f, vf, m, sbk	2 vf	-
Th. C2	Ap1	0-9	8-11	A, W	2.5Y4/3	3 f, m, sbk, gr	-	-
	Ap2	9-11	15-17	C, W	2.5Y4/2	2 f. vf, m, sbk	1 f	-
	Ap3	11-33	10-12	C, W	2.5Y4/2	2 f, m, abk, sbk	2 vf	-
Th. C3	Ap1	0-6	3-6	A, W	2.5Y5/3	3 f, m, sbk, gr	-	compost
	Ap2	6-25	17-20	C, W	2.5Y4/4	3 f, vf, m, abk, sbk	3 vf	compost
	Ap3	25-42	8-12	C, W	2.5Y4/4	2 f, vf, m, abk, sbk	3 vf	-
Th. Ct	Ap1	0-10	9-12	A, W	2.5Y5/3	3 f, gr, m, sbk	-	-
	Ap2	10-29	21-22	C, W	2.5Y4/3	2 f. vf, m, sbk	2,3 vf	-
	Ap3	29-42	10-12	C, W	2.5Y4/4	2 f, vf, m, sbk	2	-

[a] Horizons' designation according to Ref. [32]

[b] A=abrupt, C=clear, W=wavy

[c] Moist and crushed, according to the Munsell Soil Color Charts

[d] 0= Structureless, 1=weak, 2=moderate, 3=strong; vf= very fine, f=fine, m=medium, co=coarse, gr=granular, abk=angular blocky, sbk=sub-angular blocky, pl=platy, fr= friable

[e] 0=Absent, 1=very few, moderately few, 2=common, 3=many; vf=very fine, f=fine, m=medium, co=coarse.

3.2.2. Fluorescein diacetate hydrolysis (FDA)

The measured values of fluorescein diacetate hydrolysis can be compared from two points: the first one (a) is the situation at the onset of the study and the second one (b) the effect of composts throughout the research. FDA amounts were varying (min = 9 µg FDA g^{-1} h^{-1}; max = 26.18 µg FDA g^{-1} h^{-1}) at the onset of the work in the different horizons. After crop harvesting, the comparison of compost-treated soils displayed that a significant difference can be observed in the soils with the application of all composts. These observations showed that compost 2 had a positive influence on FDA value (Table 2). Since the highest fluorescein diacetate value was found in the soils with coffee husk and cow manure, it can be concluded that the composition of compost influences the outputs of the experiment.

3.2.3. Microbial biomass carbon (MBC)

The results confirmed that the fertilization approach, i.e., the application of composts, influenced the microbial biomass carbon (MBC) in the soil (Table 2). The highest value was found at the end of the study, i.e., in the soil supplemented with the compost derived from coffee husk and cow manure as compared to the control. The lowest value was measured as the control before compost distribution and cultivation, which was significant in contrast to all other ones. Compared with the other composts, the compost 2 application therefore had a great positive influence on the microbial biomass carbon, probably due to the composition and higher input of carbon organic into the soil. Giving compost influence on soil nutrients, the application of composts such as coffee husk and brewers' spent grain (Compost 1), coffee husk and cow manure (Compost 2), and a mixture of coffee husk, brewers' spent grain, and cow manure

(Compost 3), can induce enhancing effect on most soil physic-chemical features studied. Our findings supported the observations of other researchers who exhibited that the use of these natural materials raised the soil fertility. For instance, Dzung *et al.,* (2013) [15] observed that by applying the coffee husk compost, 2-3 kg plant^{-1} year^{-1} in the course of three years increased 20% to 30% over mineral fertilizers. They displayed that coffee growth, nutrient levels, and soil fertility were improved relative to plots that no using the compost. Awopetu *et al.,* (2015) [16] also demonstrated that compost derived from brewer's spent grains can increase maize yield. Bouajila and Sanaa (2011) [33] also documented that the use of cow manure compost led to an increment of organic carbon, with the compost treatment being the most efficient. Totally, it can be concluded that the organic compost application can improve soil fertility and thereby crop yield.

Compared with the other composts, the use of compost 2 experienced a higher positive effect on the microbial biomass carbon (MBC) value, perhaps owing to its composition and input of organic carbon into the soil. In other words, the microbial composition and MBC, from the narrower taxonomic level to the broad functional level (e.g., fungi and bacteria) usually respond to organic carbon resources [34][35]. Thus, the composts apply a stimulation effect on both the soil-born microbiota and the microbial biomass carbon. This finding is quite interesting and can be explained on the basis of observations reported by Wang *et al.* (2018) [17] who showed that the MBC level in the soil was influenced not only by the compost application but likely by the activity of crop roots and their exudates into the soil.

The enzymes available in soil were utilized as sensitive parameters of soil productivity, quality, and fertility. Thus, their activities reveal microbial activity and thereby accessibility of nutrients for plant uptake [36]. FDA is carried out by many enzymes like lipases, proteases, and esterase

[37]. Besides, FDA can be utilized as a parameter of microbial activity relative to the soil environment state [38]. When the FDA values were evaluated, we can conclude our findings display a positive efficacy of organic fertilizers, composts especially Compost 2, on the total microbial activity. As observed by Iovieno *et al.* (2009) [39], the use of composts increased microbial activity as compared to the control soil. The authors reported that the microbial activity was ~2.5- fold more in the compost-supplemented soil when compared with the control soil, since organic matter found in compost can provide nutrients for soil microorganisms.

Due to multiple positive impacts on the biological and physic-chemical features of soil, compost could contribute to the improvement of hemp production. It also seems that the compost possesses an equalizing influence of the accessibility of crop to nutrients, heat and air balance of soil, and seasonal/annual fluctuations concerning water, thus the ultimate hemp yield. Moreover, because of the slow release of minerals and their accessibility in compost-mediated fertilization, this compost can obtain the favorable output. Therefore, for achieving the long-term goals in sustainable agriculture, composting can be a rational item for establishing effective management of plant-nutrient in the world [40].

Table2: Microbial biomass carbon and Fluorescein diacetate hydrolysis of hemp soil

	MBC (mg C kg^{-1})			FDA (µg FDA gr^{-1} h^{-1})		
	Ap1	Ap2	Ap3	Ap1	Ap2	Ap3
T0	382.27±18.71 ABa	409.04±21.77 CDa	366.31±15.18 Ca	26.18±0.5 Bc	11.36±0.6 Bb	9±0.75 ABCa
T1.C1	376.39±64.91 ABb	381.65±25.73 Cb	249.53±22.74 Aa	31.25±1.7 Cc	17.39±1.91 CDb	8.1±2.19 ABCa
T1.C2	382.17±3.94 ABab	422.36±43.79 CDb	318.34±20.67 BCa	34.94±1.72 Cc	22.01±1.11 Eb	9±2.51 ABCa
T1.C3	432.98±19.79 ABb	395.57±9.55 CDb	249.53±22.74 Aa	33.19±2.96 Cc	16.5±2.09 Cb	6.85±3.67 ABa

T1.Ct	346.32±20.92 Ac	307.05±7.89 ABb	245.59±11.48 Aa	13.13±2.6 Ab	11.39±1.3 Bb	6.33±1.13 Aa
Th.C1	431.27±43.16 ABb	378.74±5.02 BCab	321.70±6.25 BCa	31.7±0.9 Cc	21.32±0.37 Eb	11.73±0.36 BCa
Th.C2	454.40±40.44 Bb	461.50±17.26 Db	365.31±29.79 Ca	35.1±0.37 Cc	26.11±0.59 Fb	12.96±0.73 Ca
Th.C3	442.15±53.20 ABb	408.34±39.70 CDb	259.58±35.22 ABa	32.43±0.1 Cc	20.59±0.94 DEb	9.01±0.19 ABCa
Th.Ct	344.34±27.52 Aa	298.31±30.25 Aa	349.14±30.60 Ca	10.72±0.69 Ac	7.32±0.08 Aa	9.36±0.54 ABCb

3.3. Yield

3.3.1. Hemp yield

From our observation, the composts with coffee husk (CH) and brewers' spent grain (BSG) in proportion 2:1 (Compost 1), CH and cow manure (CM) in proportion 4:1 (Compost 2), and a mixture of CH, BSG, and CM in proportion 5:3:2 (Compost 3) improved the hemp grain yield in the field. The highest yield of hemp was observed in the compost with coffee husk and brewers' spent grain (Compost 1), ~567.2 kg per ha on average. Moreover, the lowest yield of crop biomass was registered in the control without the compost application, ~367.2 kg per ha on average (Fig. 2). Totally, hemp seeds yield ranged from ~367.2 kg ha^{-1} (control) to ~567.2 kg ha^{-1} (Compost 1 treatment). From our observations, hemp seeds yield increased from ~1.2 -fold (Compost 2 and 3 treatments) up to ~1.5-fold (Compost 1 treatment) by the application of compost. The improvement influence of Pile 1 compost may be induced by higher levels of plant minerals such as N, P, K, and Mg and root augmentation resulted from the compost using [13]. Besides, this compost may increase the available form of nutrients for the crop in soil and then improves nutrient uptake by root growth that leads to crop weight and height rise [41]. Similar findings have been reported by Sefidkoohi *et al.* (2012) [42] who demonstrated that productivity of crops treated with 100 ton ha^{-1} compost enhanced significantly by ~80% in comparison with the control.

Application of biofertilizer and humic acid alone, or in combination, generally increased cannabis plant height, chlorophyll content and photosynthetic efficiency, especially immediately after a period of water stress [5]. It has been reported that maize yield was enhanced significantly by ~105 and ~125 % due to use of compost at the rate of 5 and 7 ton fed-1, respectively, relative to the control [43]. Gamal (2009) [44] also demonstrated that the use of compost (5 ton ha^{-1}) raised the productivity of sorghum by 45% in comparison with no compost application. Notably, the application of compost does not only improve the grain yield of crops in terms of field yield but it can be also improved that the quality of their products is affected in a positive manner [45]. Totally, it can be interesting to add an adequate amount of compost 1 compost for a better increment of hemp productivity.

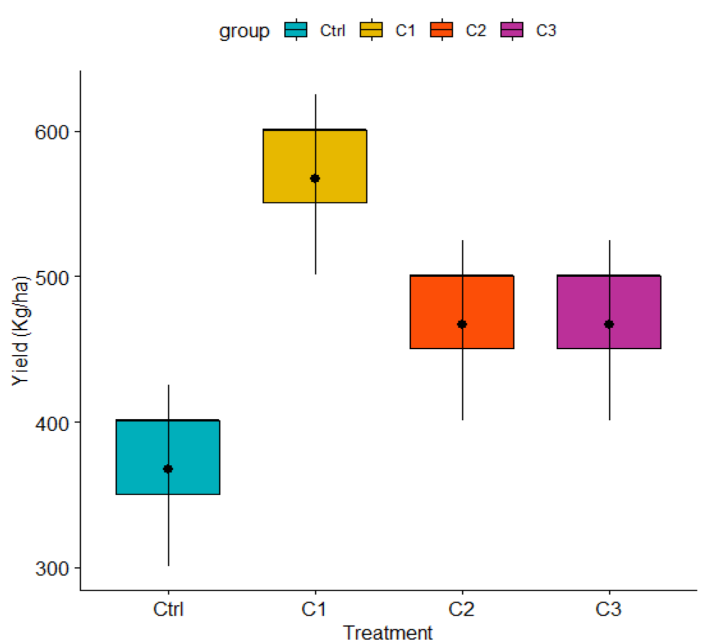


Fig. 2: The hemp yield in the field under treatment of three agri-food derived composts. Abbreviations: Compost 1, compost with coffee husk (CH) and brewers' spent grain (BSG) in proportion 2:1; Compost 2, compost with CH and cow manure (CM) in proportion 4:1; Compost 3, compost with a mixture of CH, BSG, and CM in proportion 5:3:2; Ctrl, control.

3.3.2. Lipid fraction of Hemp

Lipids accounted for approximately ~26-30% of the dry weight of hemp treated with three composts and control. Of the lipids extracted from hemp in the recent work, C18:2 n-6, alpha-18:3 and C18:1?9c were the most abundant types, accounting for an average of 90% of the fatty acids analyzed. These were followed by C16:0, and C18:0, which accounted for averages of 7.5%, and 2.5% of the total fatty acids, respectively. No main effects of composts were observed for fatty acids (Table 3). There is no significant difference among the composts for the total content of fatty acids. Anyway, slight differences in the content and composition of fatty acids among the compost-treated hemp crops can be derived from the genotypic structure and expected to affect the processing features of the hemp products [46].

Albeit this work is the most detailed research so far published on the efficiency of natural composts on the hemp lipid profile, it must be stated that only a single harvest year was evaluated. Consequently, year-to-year differences among samples due to environmental changes should be considered, including those associated with agronomy practices and genotypic structure.

Table3: Lipid fraction of hemp plants treated by three agri-food derived composts

	C1	**C2**	**C3**	**Control**
LIPID FRACTION	(Mean± Stdev.)	(Mean± Stdev.)	(Mean± Stdev.)	(Mean± Stdev.)
C12:0	0,01±0,00	0,03±0,02	0,01±0,00	0,02±0,02
C14:0	0,09±0,04	0,12±0,06	0,06±0,00	0,06±0,01
C15:0	0,03±0,01	0,03±0,01	0,02±0,00	0,03±0,01

C16:0	7,77±0,25	8,23±1,11	7,83±0,00	7,87±0,60
C16:1	0,13±0,01	0,16±0,02	0,15±0,00	0,13±0,00
C17:0	0,06±0,01	0,08±0,03	0,06±0,01	0,06±0,01
C17:1?10	0,02±0,01	0,24±0,23	0,02±0,01	0,02±0,01
C18:0	2,48±0,17	2,14±0,08	2,32±0,00	2,42±0,20
C18:1?9c	12,90±0,03	12,83±0,15	13,22±0,05	12,96±0,01
Vacc	0,84±0,06	0,86±0,03	0,93±0,00	0,86±0,05
C18:2 n-6	56,31±0,29	56,14±0,73	56,30±0,10	55,90±0,00
alpha-18:3	19,00±0,12	18,76±0,80	18,72±0,15	19,36±0,83
C20:1?11	0,39±0,01	0,42±0,03	0,38±0,01	0,37±0,03
Fat%	27,06±0,86	27,53±0,25	26,88±0,60	30,75±0,18

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