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Feasibility of Vermicomposting of Spent Coffee Grounds and Silverskin from Coffee Industries: A Laboratory Study

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Abstract: In the coffee industry, several by-products are generated during the production and consumption of coffee and represent an important waste from an environmental viewpoint. For improving the knowledge about this issue, a laboratory vermicomposting study of coffee silverskin (CS) and spent coffee grounds (SCG) spiked with mature horse manure (HM) in different proportions and using earthworm *Eisenia andrei* was carried out. The 60-day study focused on biological parameters such as total biomass gain, growth rate, cocoon production, and mortality. This study also investigated whether the vermicompost obtained could be useful and lacked toxicity through a seed germination test using hybrid wheat seeds. Results showed a disparity depending on the type of residue and the mixture used. Best options were those treatments with a medium–low amount of residue; 25% for SCG and 25% or 50% for CS. In addition, lack of toxicity was confirmed in all treatments. In conclusion, it is possible to carry out a vermicomposting of SCG and CS with some specific features.

Keywords: coffee industry by-products; coffee silverskin; spent coffee grounds; vermicompost; *E. andrei*

1. Introduction

Coffee is one of the most popular and consumed beverages all over the world. The coffee brew is preferred due to its taste, aroma, and stimulating properties. Moreover, coffee is the second largest traded commodity after petroleum [1]. Coffee processing by-products include those derived from post harvesting processing, coffee roasting, and coffee extraction, namely, immature/defective beans, husks (CH), skin and pulp (CP), parchment, silverskin (CS), and spent coffee grounds (SCG), being some of them toxic and representing serious environmental problems. Among them, CS and SCG are the main coffee industry residues obtained during bean roasting and the process to prepare “instant and espresso coffee”, respectively [1]. CS, also known as “coffee chaff”, is a thin tegument that directly covers the coffee seed. During the roasting process, coffee beans expand, and this thin layer is detached, becoming the main by-product of coffee roasting industries. SCG are the waste product from brewing coffee [2]. Finding alternatives for the use of these both residues is of great importance due to their toxic character, their pollutant potential, the huge amounts available and their potential as valuable resources. There is

a large bibliography on the valorization of SCG from both instant coffee and espresso. For instance, Hachicha et al. [3] reported that the final compost was mature with a germination index (GI) for barley of 120% in less than 20 weeks of composting, and humic acid production was 1.75 times greater in the *Trametes versicolor* inoculated mixture, suggesting that the humification process was enhanced by this white-rot fungus. Domenico et al. [4] showed how composting could be a good sustainable practice for recycling and valorizing SCG as an alternative component of growing media to partially replace commercial peat and fertilizers in the production of potted plants. Other well-known applications of SCG include fuel in the form of pellets [5] or briquettes [6,7], energy production [8], production of biofuels [9] and adsorbents for gaseous emissions [10], as a source of compounds such as antioxidants [11], as a food ingredient in bakery [12], in other alternative value-added products [13], and even as a construction material [14]. Valorization of them would be interesting from environmental and economic standpoints because it would contribute to (1) a reduction of their impact on the environment by decreasing the toxicity, (2) generation of compounds of added value, and (3) creation of employment [1].

Traditionally, landfill shipping is common and even the main option for these wastes [6]. Nowadays, sustainable alternatives to recover organic wastes and by-products are encouraged, according to Directive 2008/98/EC on waste known as Waste Framework Directive and subsequent amendments (e.g., Directive (EU) 2018/851 of the European Parliament and of the Council of 30 May 2018 amending Directive 2008/98/EC on waste). Owing to the environmental issues and pollution problems associated with conventional treatment methods for organic wastes, vermicomposting has been growing as an emerging cost-effective and environmentally sound treatment option for a wide range of industries. Moreover, nowadays biological processes are suggested to process and treat non-toxic wastes with a paradigm to convert them into energy and organic manure [15]. Vermicomposting is a low-cost biotechnological process that biodegrades and stabilizes organic wastes under aerobic and mesophilic conditions, through the action of certain earthworm species capable of feeding on the residue while accelerating their microbial degradation [16]. The importance of earthworms in the breakdown of organic matter and the release of the nutrients that it contains has been known for a long time [17], but it was not until the late 1970s when true interest in the use of earthworms for processing organic wastes began [18].

Thenceforth, the utilization of earthworms in waste management practices has been widespread and well documented in published scientific literature on wastes from several and diverse industries. For instance, the latest studies reported have been rice straw [19] or fly ash [20], to point a couple of examples out.

Only few reports are available to provide knowledge on vermicomposting of coffee wastes. Orozco et al. [21] reported the vermicomposting of CP using the earthworm *Eisenia foetida*. They found that nutrients were in forms that are readily taken up by the plants such as nitrates, exchangeable phosphorus, and soluble potassium, calcium, and magnesium, although they also suggested that potassium was lost due to leaching. Degefe et al. [22] made vermicompost of CH using the same epigeic earthworm as well, and they reported an increase in total Kjeldahl nitrogen (TKN), total potassium (TK), total phosphorus (TP), and calcium whereas the C:N ratio and total organic carbon (TOC) decreased. A vermiconversion using *Eudrilus eugeniae* of CH was performed by Sathianarayanan and Khan [23], who successfully tested for the suppression of plant pathogen *Rhizoctonia solani*. Raphael et al. [24] reported the vermicomposting of CP using the exotic *E. eugeniae* and the native earthworm *Perionyx ceylanesis*. wherein the percentage of nitrogen, phosphorus, potassium, calcium, and magnesium increased while the C:N ratio, pH, and TOC declined. Adi and Noor [25] vermicomposted SCG using *Lumbricus rubellus* in order to better stabilize kitchen wastes composed by animal residues from fish and chicken.

Other papers interested in coffee industry by-products only add references about composting or quote any of the examples from the previous paragraph [26,27]. Finally, the review on utilization and composition of CS made by Narita and Inouye [28] shows a lack of reports about vermicomposting.

Using earthworms to treat organic residues is not always possible or has limitations. For instance, earthworms are very sensitive to ammonia and salt: $<1 \text{ mg g}^{-1}$ of ammonia and $<0.5\%$ salts [18]. Furthermore, scientific literature shows some cases where vermicomposting was not possible or the substrates were inadequate for earthworms survival [29].

The aim of this work was to analyze the potential of CS and SCG to be directly decomposed through vermicomposting using *E. andrei* [30] worms. A laboratory investigation was carried out employing an epigeic earthworm in different combinations mixed with horse manure (HM), focusing on biological parameters such as earthworm growth and cocoon production patterns. Moreover, a germination assay was posteriorly carried out to determine the maturity of vermicompost produced.

2. Materials and Methods

2.1. Materials

Mature HM was collected in an Equine Center in Navarrese town of Labiano. SCG and CS were collected from an instant coffee factory and a coffee roasting factory, respectively, on the outskirts of Pamplona, and both products were very homogeneous. An initial analysis compound was obtained based on the guidelines set by the Annex VI 'Analytical methods' of the Royal Decree 506/2013 of 28 June on fertilizer products. This job was made by Agrolab Analítica S.L., a company located in Navarrese town of Mutilva, which is accredited by the Spanish Ministry of Agriculture as a competent laboratory for official control for initial and contradictory analyses of fertilizer products. The physicochemical characteristics of the materials are shown in Table 1.

Table 1. Physicochemical characteristics of the raw materials.

PARAMETERS	HM	CS	SCG
pH	8.3	8.5	3.9
EC (dS/m)	1.4	2	0.5
OMC (g 100 g ⁻¹)	73.9	91.3	99.1
TOC (g 100 g ⁻¹)	42.9	53	57.5
TKN (g 100 g ⁻¹)	3.2	5.1	2.1
Ammonia (g 100 g ⁻¹)	0.2	0.5	<0.1
Organic N (g 100 g ⁻¹)	3	4.6	2
C:N ratio	14	11	28
TP (g 100 g ⁻¹)	2.1	0.4	0
TK (g 100 g ⁻¹)	2.2	3.3	<0.1
Ca (g 100 g ⁻¹)	6.6	2.2	0.2
Mg (g 100 g ⁻¹)	1.2	0.8	<0.1
Na (g 100 g ⁻¹)	0.2	<0.1	<0.1
S (g 100 g ⁻¹)	1.7	1.2	0.3
Fe (g 100 g ⁻¹)	0.21	0.15	0.58

EC = electrical conductivity; OMC = organic matter content; TOC = total organic carbon; TKN = total Kjeldahl nitrogen; TP = total phosphorus; TK = total potassium.

Plastics boxes were acquired from Martín Contenedores para Logística S.L. in the Navarrese town of Aoiz, and the rest of materials were from other suppliers in the surroundings of Pamplona. *E. andrei* worms and coconuts were purchased from Vermican Soluciones de Compostaje S.L., a local company specialized in composting.

2.2. Experimental Design and Vermicomposting

The combinations carried out in this study are shown in Table 2. Plastic boxes of size $60 \times 40 \times 40$ cm with small holes at the bottom for drainage were used. Each box was completed with a detachable plastic cover on the top and other smaller plastic trays of size $60 \times 40 \times 15$ cm at the bottom in order to collect leaches. Inside, a wet coconut bed was used to maintain moisture and to

restrain the worms from escaping through the small holes at the bottom of the box. A total of eight combinations of SCG or CS with HM at ratios of 25:75%, 50:50% 75:25%, and 100:0% were prepared. HM 100% was used as reference. Six kilograms of each waste combination and 200 worms were held in each box. For each combination three boxes were prepared. The duration of the experiment was 60 d in total and was kept in laboratory. The moisture content was maintained in the range 50–80% by spraying the surface with water one or twice per week, whereas environmental temperature was constantly maintained between 20–24 °C.

Table 2. For the vermicomposting trial, a total of nine combinations of horse manure (HM), spent coffee grounds (SCG), and coffee silverskin (CS) mixtures were prepared.

Treatments	Name Given	HM	SCG	CS
T1	Control	100	0	0
T2	HM75/25SCG	75	25	0
T3	HM50/50SCG	50	50	0
T4	HM25/75SCG	25	75	0
T5	100% SCG	0	100	0
T6	HM75/25CS	75	0	25
T7	HM50/50CS	50	0	50
T8	HM25/75CS	25	0	75
T9	100% CS	0	0	100

2.3. Biological Analysis

The initial situation was compared to the final one after the planned time had elapsed, employing a widespread methodology in vermicomposting studies [31]. Earthworms were separated from the parental or waste mixture by hand sorting method, likewise for cocoons. Worms were also washed in tap water to remove adhered material from their body and weighed. The growth rate of the worms (GR), total biomass gain (TBG), and cocoon production ratio (PR) were calculated as shown in Equations (1)–(3), respectively. The percentage of mortality was also calculated.

$$\text{GR (mg/worm/day)} = \frac{\text{Final weight} - \text{Initial weight}}{\text{Total vermicomposting time} \times \text{Average number of earthworms during the vermicomposting period}} \quad (1)$$

$$\text{TBG} = \frac{\text{Final weight} - \text{Initial weight}}{\text{Initial weight}} \quad (2)$$

$$\text{PR (cocoons/worm/week)} = \frac{\text{Number of cocoons}}{\text{Total vermicomposting time} \times \text{Average number of earthworms during the vermicomposting period}} \quad (3)$$

2.4. Seed Germination

This test was considered for the assessment of the maturity of the vermicompost. Employing an adaptation of the methods of seed germination essay published in earlier studies, 5 g samples from each combination at the age of 60 d were mixed with 50 g of deionized water. Fresh extract of each sample and deionized water in 1:1 proportion were poured into a Petri dish lined with filter paper. Deionized water was considered as control. Twenty-five bread wheat (*Triticum aestivum* hybrid cultivar Hybiza) seeds were placed on the filter papers and were kept for 5 d under dark conditions at 25 °C. Each combination was performed in triplicate.

Total germination, also known as germination percentage (GP), percentage of relative seed germination (RSG), relative root growth (RRG), relative shoot growth (RShG), and germination index (GI) were calculated by Equations (4)–(8), based on the available reference [32].

$$GP = \frac{\text{Number of germinated seeds}}{\text{Total number of seeds tested}} \times 100 \quad (4)$$

$$RSG = \frac{\text{Number of germinated seeds in vermicompost extract}}{\text{Number of germinated seeds in control}} \times 100 \quad (5)$$

$$RRG = \frac{\text{Mean root length in vermicompost extract}}{\text{Mean root length in control}} \times 100 \quad (6)$$

$$RShG = \frac{\text{Mean shoot length in vermicompost extract}}{\text{Mean shoot length in control}} \times 100 \quad (7)$$

$$GI = \frac{RSG \times RRG}{100} \quad (8)$$

A primary root ≥ 5 mm was used as the operational definition of positive seed germination, in accordance with USEPA [33].

2.5. Statistical Analysis

Statistical analysis was carried out using R computer software package. One-way analysis of variance (ANOVA) was performed to analyze the significant differences between combinations during vermicomposting at 5% level of significance. Tukey's test was used to determine any significant difference between each combination in the variables of interest involved. An identical germination essays analysis was carried out too. The normality and homoscedasticity of variance for all the variables involved were corroborated in all the cases.

3. Results and Discussion

3.1. Biological Results

The growth and reproduction of the earthworm species used for vermicomposting of organic waste materials are considered as a good sign of the effective vermicomposting process [34]. In the current study, the growth and reproduction of the earthworm *E. andrei* was assessed in terms of TBG, GR, PR, and mortality at the end of the vermicomposting period in different treatments.

As shown Figure 1, TBG of the earthworm *E. andrei* was higher in T2 (2.01 ± 1.28) followed by T1 (1.03 ± 0.47) and T7 (0.99 ± 0.47), with no statistical differences between them. In T6 (0.60 ± 0.34), T3 (0.59 ± 1.06), and T8 (0.52 ± 0.08), no statistical differences were also found between its values. The lowest values were reached for the combinations T4 (-0.44 ± 0.57), T5 ($22,120.90 \pm 0.10$), and T9 (-1.00 ± 0.00), which also were not statistically different from each other. There were statistically significant differences between T2 with respect to T4, T5, and T9, as well as between T1 and T7 with respect to T5 and T9. The trend of TBG was $T2 > T1 > T7 > T6 > T3 > T8 > T4 > T5 > T9$. These results show that mixtures composed of 100% by-product give poor results, similar to recent studies, such as Usmani et al. [35], who reported that poor biomass growth of earthworms was observed in the treatment comprised solely of coal fly ash, without manure, and Gong et al. [36] where better results were obtained when garden wastes were mixed with cattle manure and/or spent mushroom substrate than in solitary. Similar conclusions were obtained during vermicomposting of water lettuce by Suthar et al. [37].

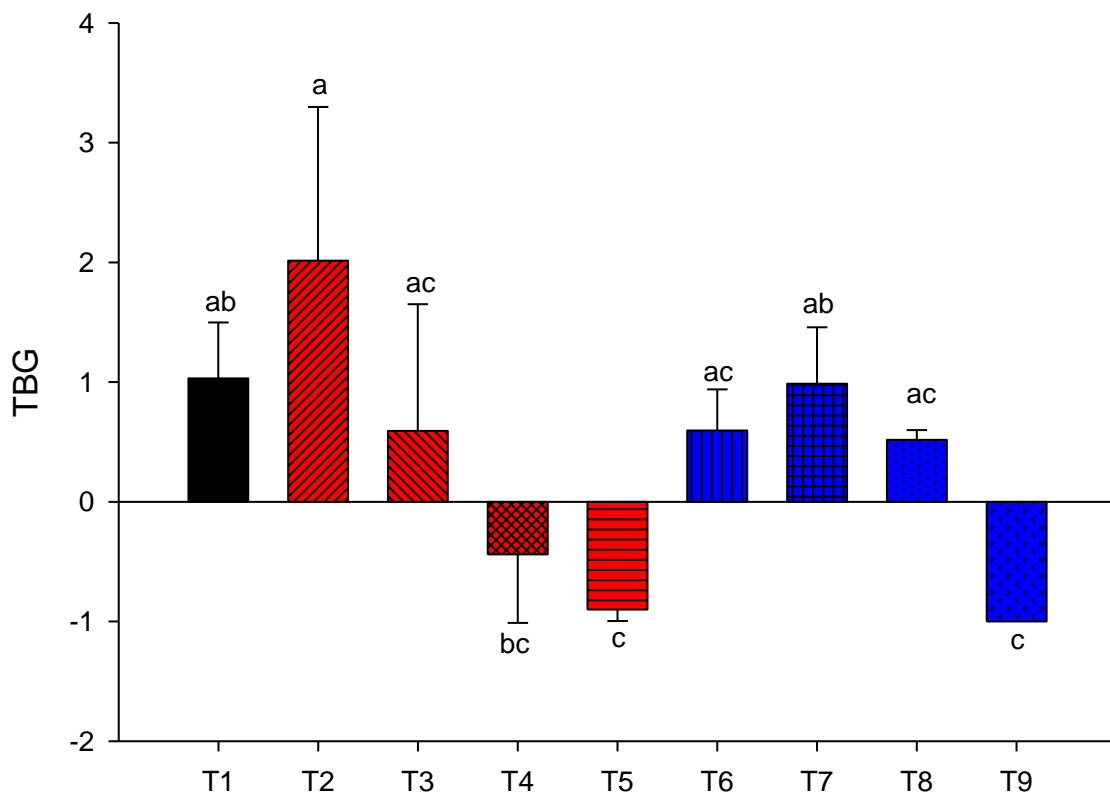


Figure 1. Total biomass gain (TBG) in different feed mixtures of spent coffee grounds (SCG) and coffee silverskin (CS) with horse manure (HM) followed by different letters are significantly different (one-way ANOVA; Tukey's test, $p \leq 0.05$).

GR (mg weight gained/worm/day) has historically been considered a good comparative index to compare the growth of earthworms. Domínguez and Edwards [38] showed that the GR can vary considerably depending on the stocking rate or the moisture content, obtaining values ranging from 5.11 to 17.54 mg/worm/day, employing *E. andrei* in pig manure. According to the results observed for the GR (Figure 3) of *E. andrei*, T2 (4.39 ± 2.36 mg/worm/day) and T7 (3.37 ± 1.43 mg/worm/day) yielded the highest values, followed by T8 (2.83 ± 0.48 mg/worm/day), T1 (2.39 ± 1.17 mg/worm/day), T6 (1.72 ± 1.44 mg/worm/day), and T3 (1.01 ± 2.00 mg/worm/day) (Figure 2). These values did not differ statistically from each other. Lower values were found in T4 (-1.87 ± 2.20 mg/worm/day), T5 (-3.85 ± 1.52 mg/worm/day), and T9 (-9.54 ± 1.42 mg/worm/day). Adi and Noor [25] reported losses and gains in weight and in terms of the number of earthworms using SCG mixed with kitchen waste and cow dung. Noticing the negative values of the parameter, the lowest one yielded by T9 was statistically different from the rest of the cases. T5 was statistically different from all the combinations that yielded positive values, whereas T4 was only statistically different with respect to the highest ones, i.e., T2 and T7. Thus, the trend of GR was $T2 > T7 > T8 > T1 > T6 > T3 > T4 > T5 > T9$.

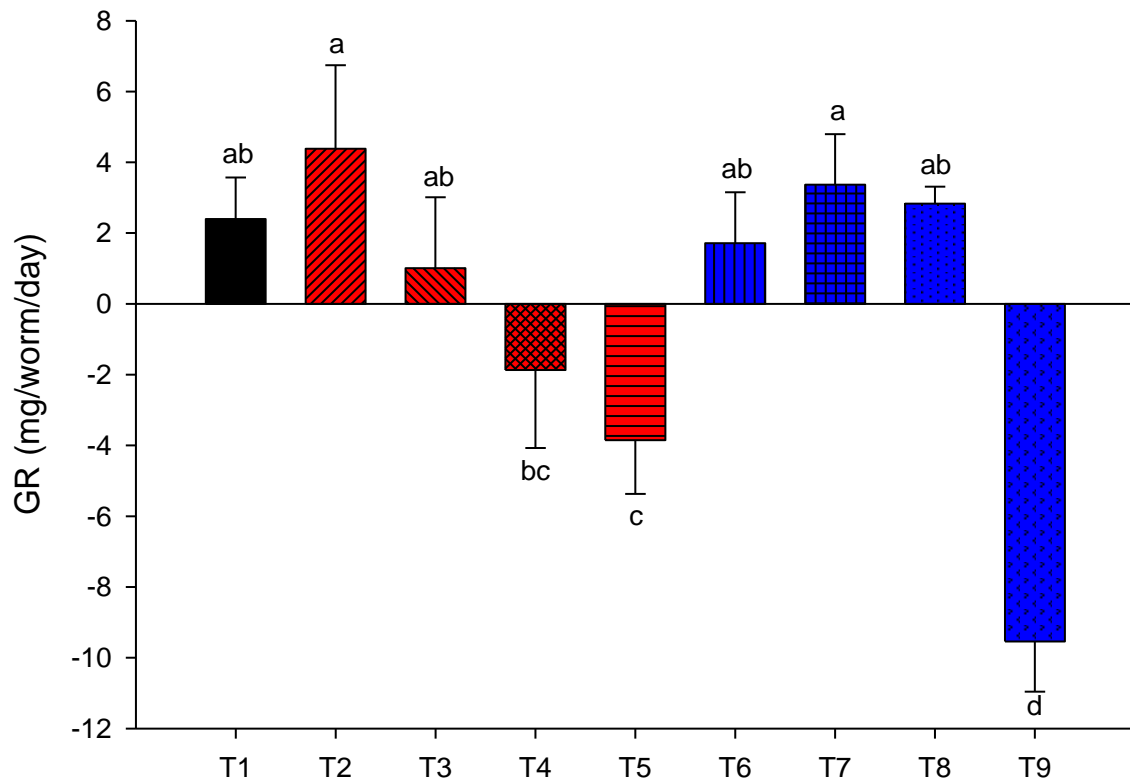


Figure 2. Growth rate (GR) in different feed mixtures of spent coffee grounds (SCG) and coffee silverskin (CS) with horse manure (HM) followed by different letters are significantly different (one-way ANOVA; Tukey's test, $p \leq 0.05$).

These results are in accordance with Elvira et al. [39] who reported weight losses for *E. andrei* in paper pulp mill sludge (-0.11 mg/worm/day) and in pure cultures, a gradual and continuous biomass increment of 4.67 mg/worm/day. Cluzeau et al. [40] also obtained growth rates of 4.5 mg/worm/day in batch culture for immature *E. andrei* fed on HM and peat, and Elvira et al. [41] recorded values from 1.25 to 1.75 and 2.23 mg/worm/day in mixed cultures (*E. andrei* + *D. rubida* and *E. andrei* + *L. rubellus* respectively) in cow manure. Better GRs were recorded by other previous studies employing *E. andrei*. For instance, Elvira et al. [39] reported higher weight gains in cow dung (12.25 mg/worm/day) or rabbit manure (12.78 mg/worm/day). Haimi [42] informed an *E. andrei* GR of 10.76 mg/worm/day in batch culture, whereas a 18.6 – 10.96 ratio was obtained by Dominguez et al. [43] vermicomposting sewage sludge and mixtures with the different bulking materials. Elvira et al. [44] showed a ratio between 9.62 and 8.04 , being cattle manure as control with 8.4 mg/worm/day.

Other earlier studies also reported values which were considerably lower than previous experiments. Suthar [45] reported GR between 1.45 and 6.33 mg/worm/day employing *E. fetida* in different treatments of industrial sludge mixed with cow dung, biogas plant slurry, and wheat straw. Previously, they had described a GR between 6.16 and 10.62 mg/worm/day employing *E. eugenia* vermicomposting post-harvest residues of some local crops with cow dung [46].

Li et al. [31] recently reported a negative growth rate in the treatment for banana peels, cabbage, potato, and its mixture with excess activated sludge, while a positive growth rate was observed in other treatments. They stated that the excessive growth rate of earthworms might lead to higher consumption of nitrogen and faster denitrification process in earthworms' digestive tract.

According to the PR results, showed in Figure 3, the best combinations were those with residue proportions of 25% or 50% . Similar conclusions were observed at 60 d by Bhat et al. [47]. The values in T2 (0.35 ± 0.15 cocoons/worm/week) and T6 (0.35 ± 0.06 cocoons/worm/week) were higher than in the rest of the cases, followed by T7 (0.28 ± 0.18 cocoons/worm/week). The values of T8 (0.09 ± 0.05 cocoons/worm/week), T1 (0.05 ± 0.05 cocoons/worm/week), and T3 (0.05 ± 0.05 cocoons/worm/week)

were higher than those of T4, T5, and T9 (0.00 ± 0.00 cocoons/worm/week), although no statistical differences were found between them. The values in T2 and T6 were statistically higher than in the rest of the cases, except from the one in T7. In turn, T7 was statistically different from T4, T5, and T9. The trend was $T2 = T6 > T7 > T1 = T3 > T4 = T5 = T9$.

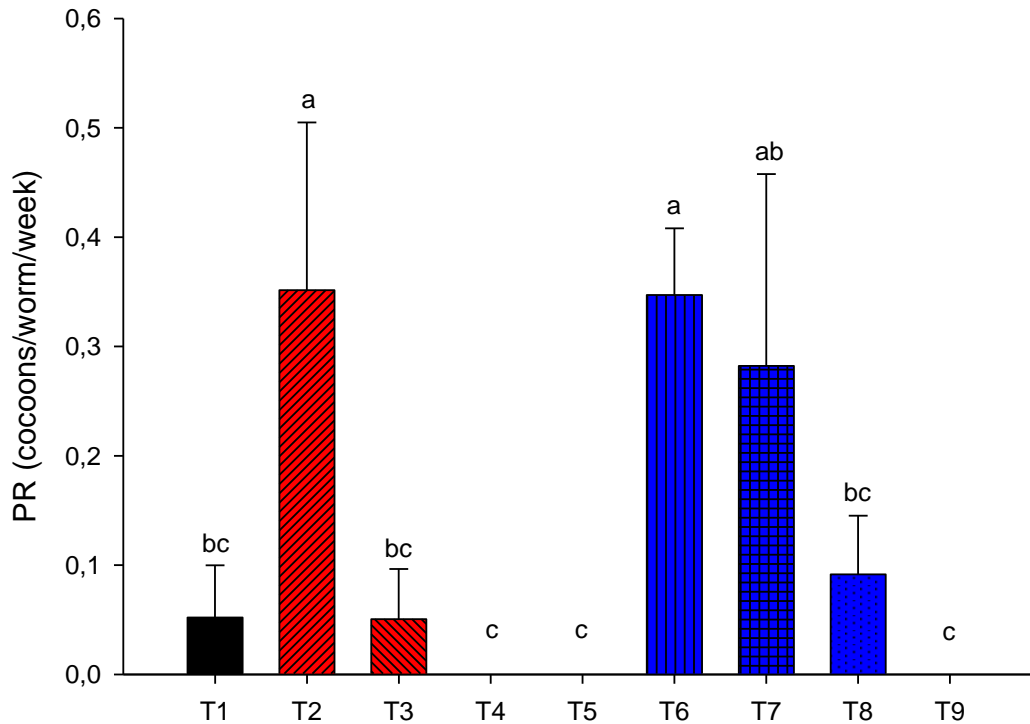


Figure 3. Cocoon production ratio (PR) in different feed mixtures of spent coffee grounds (SCG) and coffee silverskin (CS) with horse manure (HM) followed by different letters are significantly different (one-way ANOVA; Tukey's test, $p \leq 0.05$).

Domínguez et al. [43] showed huge differences in total *E. andrei* cocoon production in the sewage sludge and in the mixtures with the different bulking agents, with ratios between 0.05 and 3.16 cocoons/earthworm/week, and Frederickson et al. [48] obtained ranges between 0.46 and 1.56 cocoons/worm/week vermicomposting green wastes. Highest values reported in earlier studies were 1.82 [40], 2.14 [39], and 3.08 cocoons/worm/week [42]. Edwards et al. [49] reported that the important difference rates of cocoon production in different organic wastes are related to the quality of the waste material used as feed. The results clearly illustrate that both wastes are not as excellent as other manures or organic wastes, but they also do not badly behave in general terms, with the main exception of 100% waste proportion treatments.

The earthworm mortality rate is the bioindicator of palatability and suitability of feeds for earthworms in vermicomposting of industrial or toxic wastes [50]. In this study, the mortality of the earthworms (Figure 4) reached the highest values in T9 ($100.00 \pm 0.00\%$) and T5 ($87.50 \pm 12.50\%$), followed by T4 ($50.00 \pm 43.30\%$), with no statistical differences between them.

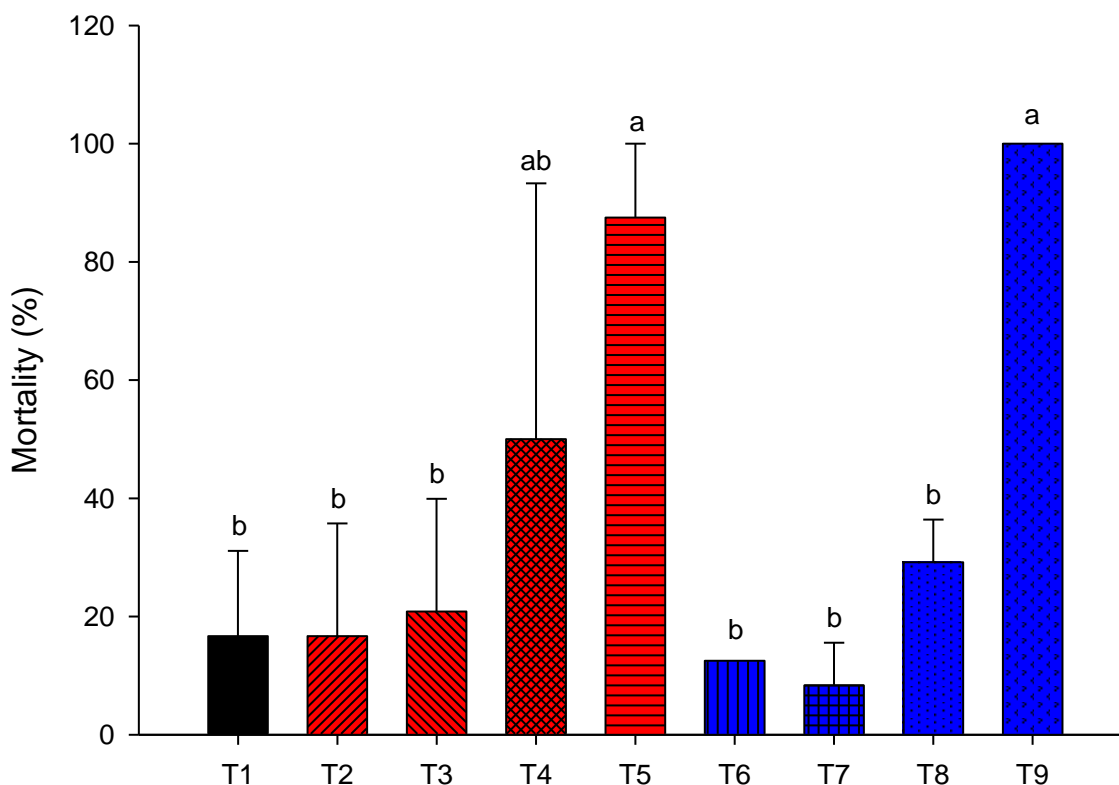


Figure 4. Percentage of mortality in different feed mixtures of spent coffee grounds (SCG) and coffee silverskin (CS) with horse manure (HM) followed by different letters are significantly different (one-way ANOVA; Tukey's test, $p \leq 0.05$).

Butt [51] found that the earthworm survival in the vermicomposting subsystem mainly depended on the chemical structure of the feedstock. Some toxic substance produced during the degradation process led to mortality in earthworms in waste mixtures [52]. Suthar and Singh [50] concluded that more availability of toxic chemicals, due to higher proportions of residue, possibly caused mortality at a higher rate in composting earthworms. Moreover, it has often been stated that SCG has biologically toxic properties because of substances such as caffeine, tannins, and polyphenols [1]. In fact, Roberts and Wyman Dorough [53] reported caffeine as a very toxic ($10\text{--}100 \mu\text{g}/\text{cm}^2$) substance for earthworms in their studies with *E. foetida*.

The lowest mortality values corresponded to T7 (8.33 ± 7.22) and T6 (12.50 ± 0.00 %), followed by T1 (16.67 ± 14.43 %), T2 (16.67 ± 19.09 %), T3 (20.83 ± 19.09 %), and T8 (29.17 ± 7.22 %), with no statistical differences between all of them. T5 and T9 showed statistically significant differences with respect to the control (T1) and combinations T2, T3, T6, T7, and T8. The extremely low pH in the case of SCG and the high ammonia content in the case of CS were two determining factors in this study. This is in concordance with Suthar et al. [37], who asseverated that a low feeding rate and non-acclimatized environment for newly inoculated worms could have affected the survival of earthworms in waste mixtures. In summary, the mortality trend was $T9 > T5 > T4 > T8 > T3 > T2 = T1 > T6 > T7$ for this study. Overall, the earthworm mortality was higher in treatments that contained more coffee waste proportions. This tendency, where the increase in residue, and therefore decrease in manure, led to a greater increase in mortality, was observed historically in other reports employing different earthworms [36,37,54,55].

Total mortality in T9, whose composition was totally CS, is remarkable but not unique. For instance, Elvira et al. [39] previously showed the death of all *E. foetida* individuals due to a fermentation process in domestic waste treatment. A previous pre-composting would be suggested, but because it was carried out before, the final material obtained in only few days had radically changed and was considered

more composted than pre-treated. These authors discourage its use because it is understood there is no point in vermicomposting already biostabilized material, in concordance with the aim of this study.

3.2. Germination Results

The seed germination test is normally employed to assess the degree of maturity and phytotoxicity of composts for agricultural use [34]. Table 3 depicts the effect of the different combinations on germination parameters of bread wheat seeds. Specifically, the mean values and standard deviations of the GP, RSG, RRG, RShG, and GI are shown. According to Zucconi et al. [32], a GI of $\geq 80\%$ indicates the disappearance of phytotoxins in composts. The ranks of the parameters GP, RSG, RRG, RShG, and GI were $(42.7 \pm 6.1\text{--}66.7 \pm 11.5 \%)$, $(84.9 \pm 18.2\text{--}135.0 \pm 41.3 \%)$, $(133.6 \pm 79.8\text{--}275.2 \pm 253.3 \%)$, $(171.8 \pm 121.8\text{--}345.1 \pm 294.3 \%)$, and $(124.3 \pm 83.1\text{--}384.3 \pm 339.7 \%)$, respectively.

Table 3. Germination essay results: Data of vermicomposts extracts on germination percentage (GP), relative seed germination (RSG), relative root (RRG) and shoot growth (RShG), and germination index (GI) of hybrid Hybiza bread wheat and without significant differences (one-way ANOVA; Tukey's test, $p \leq 0.05$).

Mean Values (and Standard Deviations) of Bread Wheat Seeds Essay Germination Parameters					
Treatment	GP (%)	RSG (%)	RRG (%)	RShG (%)	GI (%)
T1	49.3 \pm 8.3 ^a	96.9 \pm 13.3 ^a	161.5 \pm 85.8 ^a	231.4 \pm 86.6 ^a	158.1 \pm 90.0 ^a
T2	45.3 \pm 25.4 ^a	90.5 \pm 58.4 ^a	158.8 \pm 159.1 ^a	238.9 \pm 244.5 ^a	204.5 \pm 262.0 ^a
T3	50.7 \pm 18.9 ^a	104.3 \pm 48.8 ^a	133.6 \pm 79.8 ^a	171.8 \pm 121.8 ^a	158.7 \pm 116.4 ^a
T4	54.7 \pm 18.0 ^a	107.0 \pm 32.5 ^a	275.2 \pm 253.3 ^a	307.7 \pm 255.4 ^a	346.3 \pm 388.0 ^a
T5	44.0 \pm 6.9 ^a	91.0 \pm 34.4 ^a	185.6 \pm 198.2 ^a	179.6 \pm 190.9 ^a	205.5 \pm 253.6 ^a
T6	49.3 \pm 12.2 ^a	102.6 \pm 43.1 ^a	153.3 \pm 135.9 ^a	205.8 \pm 175.4 ^a	181.4 \pm 195.2 ^a
T7	66.7 \pm 11.5 ^a	135.0 \pm 41.3 ^a	256.4 \pm 216.0 ^a	345.1 \pm 294.3 ^a	384.3 \pm 339.7 ^a
T8	42.7 \pm 6.1 ^a	84.9 \pm 18.2 ^a	140.2 \pm 84.7 ^a	244.5 \pm 128.0 ^a	124.3 \pm 83.1 ^a

Same letter indicates not significant differences. Values are the mean \pm SD ($n = 3$).

The results showed that there were no statistically significant differences (p -value > 0.05) between the different combinations for the aforementioned germination parameters. This can be explained by the large standard deviations of the data. Likewise, the trends associated with those parameters were $T7 > T4 > T3 > T1 = T6 > T2 > T5 > T8$, $T7 > T4 > T3 > T6 > T1 > T5 > T2 > T8$, $T4 > T7 > T5 > T1 > T2 > T6 > T8 > T3$, $T7 > T4 > T8 > T2 > T1 > T6 > T5 > T3$ and $T7 > T4 > T5 > T2 > T6 > T3 > T1 > T8$ respectively.

T4 yielded the highest values for all the germination parameters among the SCG combinations, whereas in CS mixtures it was T7. In fact, T7 was the combination where higher values were found, compared to the rest.

Karak et al. [56], who employed seed of wheat (*T. aestivum* cv. PBW 3) besides Indian mustard (*Brassica campestris* cv. Pusa Jaikisan), in a traditional municipal solid waste compost, reported 89–96 and 86.9–98.25% of GI respectively. In vermicomposting, Karmegam et al. [34] presented a study where GP and GI ranged from 86 to 98% and 279–360 for maize (*Zea mays* var. Co 6) and 84–98% and 285–372 for cowpea (*Vigna unguiculate* var. VBN) respectively. Testing a jute mill waste vermicompost, Das et al. [57] reported lower values of RSG, RRG, RShG, and GI in a germination essay with *Vigna radiata* and *V. mungo*, not reaching the value of 160%. Unuofin and Mnkeni [58] had GI values that ranged from 98 to 162% for tomato (*Lycopersicon esculentum*), 48–81 for carrot (*Daucus carota*), and 51–128 for radish (*Raphanus sativus*). Che et al. [59] observed that the GI values of vermicomposting showed an increasing trend, and on day 20 the GI value had reached 86%, whereas in Zhang and Sun [60], 48 h seed germination ranged from 72 to 162% employing pakchoi (*Brassica rapa* L., Chinensis group) seeds. Better results were shown in our study. Nevertheless, Hussain et al. [61] published values which fall

within a range of approximately from 600 to 1100% for RRG, 100–500 for RSG, 700–1600 for RShG, and 1000–5500 for GI.

3.3. Troublesome Inconvenience Analysis

During the vermicomposting process there was notable trouble of great relevance. In T9, total earthworm death occurred during the first days. An increase in temperature above 40–45 °C occurred, similar to the beginning of the thermophilic phase of the traditional composting process [62]. Particular characteristics of CS at that initial stage, including high humidity, a low C:N ratio by high ammonia content, and the small size and shape of particles, were obviously crucial in that outbreak. Earthworms are very sensitive to ammonia and do not survive long in organic wastes containing much ammonia. In addition, almost all do not withstand in environments above 30 °C or pH below 5.0 [18]. It has been reported that the increase in nitrogen content in feedstocks leads to the death and decay of the earthworms [19]. That is why, in addition to the adverse environmental conditions caused by the initial state of CS, the high content of ammonia was the main reason for deaths and suggests that a pre-treatment would be necessary. As a consequence of the aforementioned, the absence of data for this combination T9 is clearly explained.

Not as drastic as T9, but also remarkable, was the decrease in the total number of worms at the end of the study at mortality and attempts to escape during the first weeks in combination with a higher proportion of SCG, due to the low pH. As a curiosity, discoloration of the worms was also detected in T4 and T5, observing quite striking pinkish and whitish tones during the first weeks, but without apparent consequences in comparison other combinations.

4. Conclusions

The study reveals that coffee industrial by-products can be decomposed, obtaining a new value-added material in the form of compost. The results suggest that, in order to achieve high utilization of coffee by-products through vermicomposting, the best options are the treatments with a medium–low amount of residue due to the specific characteristics of these wastes and possible toxicity due to some of their own natural substances that comprise them. Furthermore, a pre-treatment is necessary for CS when used alone in vermicomposting, and it is also recommended that SCG is used with other amendments or alkaline residues in order to increase the pH level. Broadly, more trials need to be conducted in the future.

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Abbreviations

CH	coffee husk
CP	coffee pulp
CS	coffee silverskin
EC	electrical conductivity
GI	germination index
GP	germination percentage
GR	growth rate
HM	horse manure
NC	number of cocoons
OMC	organic matter content
PR	cocoon production ratio
RRG	relative root growth
RSG	relative seed germination
RShG	relative shoot growth
SCG	spent coffee grounds
TBG	total biomass gain
TK	total potassium
TKN	total Kjeldahl nitrogen
TOC	total organic carbon
TP	total phosphorus

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