Bioresource Technology 341 (2021) 125925

Contents lists available at ScienceDirect

Bioresource Technology

journal homepage: www.elsevier.com/locate/biortech

Conversion of spent coffee grounds into vermicompost

Ales Hanc [a,*], Tereza Hrebeckova [a], Alena Grasserova [b,c], Tomas Cajthaml [b,c]

[a] Department of Agro-Environmental Chemistry and Plant Nutrition, Czech University of Life Sciences Prague, Czech Republic
[b] Institute of Microbiology, Academy of Sciences of the Czech Republic, Prague, Czech Republic
[c] Institute for Environmental Studies, Faculty of Science, Charles University in Prague, Czech Republic

HIGHLIGHTS

- The addition of straw to spent coffee grounds supported earthworm development.
- The contents of fungi and enzymes decreased with the age of vermicompost.
- Earthworms were able to substantially reduce the caffeine stimulant content.
- Vermicompost contained more P, K and Mg than the variant without earthworms.
- Biowaste with earthworms was stabilized faster than without earthworms.

ARTICLE INFO

Keywords:
Spent coffee grounds
Vermicomposting
Straw pellets
Caffeine
Eisenia andrei

ABSTRACT

The present study was focused on vermicomposting of spent coffee grounds (SCG) and its mixtures with straw pellets. The process was evaluated in terms of biological and physico-chemical properties. The greatest number and biomass of earthworms was found in the treatment with 25% vol. SCG + 75% vol. straw pellets. In this treatment, the upper youngest layer exhibited 1.6-fold and 4.5-fold greater earthworm number and biomass, respectively, than the bottom oldest layer. Earthworm weight decreased in direct proportion to the layer age. The oldest treatment layer was characterized by lesser contents of fungi and six hydrolytic enzymes, compared to the younger layers. Further, the oldest treatment layer had suitable agrochemical properties. Earthworms were able to substantially reduce the caffeine stimulant content, which is considered the most representative pharmaceutically active compound.

1. Introduction

Worldwide, the coffee processing industry produces almost 33 million tons of solid coffee waste (coffee pulps, mucilages, and hulls) per year. The coffee processing industry utilizes almost 15 L of water per kilogram of freshly harvested green coffee during the various stages. The process discharges effluent which can pollute a receiving water system and soil (Alemayehu et al., 2020). In 2019, annual coffee bean production exceeded 10 million tons (International Coffee Organization, 2020). The residue after preparing the coffee drink are spent coffee grounds (SCG). Since coffee brewing involves the extraction of selected compounds from coffee beans, a large amount of unused waste is generated (about 90 % by weight) (McNutt and He, 2019; Blinová et al., 2017). Especially, in cafes, SCG can account for a substantial proportion of food waste. SCG can be defined as an organic residue with high humidity and small particle size (Esquivel and Jiménez, 2012). SCG contain large amounts of organic matter, including polysaccharides, especially cellulose (with glucose as a main component), and hemicellulose (with mannose, galactose, and arabinose as main components), which together make up half of the SCG dry mass. Lignin (25% wt. on a dry mass basis), protein (almost 20% wt. on a dry mass basis), and oil (with over 15% wt. on a dry mass basis) are also significant in SCG (Ballesteros et al., 2014; Mussatto et al., 2011; Yordanov et al., 2016; Kovalcik et al., 2018). Most SCG end up in landfills or in the sewage system, which is a serious environmental problem related to biodegradable organic matter decomposition and the release of potentially toxic compounds, for example, polyphenols, tannins, and caffeine (Low et al., 2015; Murthy and Madhava Naidu, 2012). SCG are characterized

* Corresponding authors at: Ales Hanc, Department of Agro-Environmental Chemistry and Plant Nutrition, Czech University of Life Sciences Prague, Kamycka 129, 165 00 Prague 6, Czech Republic.
E-mail address: hanc@af.czu.cz (A. Hanc).

https://doi.org/10.1016/j.biortech.2021.125925
Received 31 July 2021; Received in revised form 6 September 2021; Accepted 8 September 2021
Available online 13 September 2021

by a great caffeine content, but the specific value depends on the coffee sort and processing (Peshev et al., 2018). Caffeine is considered as pharmaceutically active compound (PhAC) pollutant. Due to its large amount in the environment, it is suitable as an indicator of anthropogenic inputs of PhACs in unregulated water bodies. Caffeine residues are very stable in the water environment (Li et al., 2020).

In practice, only a small part of SCG production is used. According McNutt and He (2019), possible applications for SCG include:

- biodiesel, bioethanol, biochar, and biogas production (Obruca et al., 2014; Tongcumpou et al., 2019)
- direct fuel source
- extraction of phenolic compounds, antioxidants, and phytosterols (Nzekoue et al., 2020)
- use as subgrade filler material, effective adsorbent for a wide range of contaminants, composites, and bricks incorporating SCG
- direct application into soil – there is frequent anecdotal recommendation for the use of locally produced SCG as fertilizer into the soil. However, this is against the conclusions of scientific experiments. For example, in an experiment with five horticultural plants in different soil types with various SCG amendment rates, detrimental plant growth effects were found, regardless of soil type and fertiliser addition. Growth suppression was not explained by a change in any of the major parameters, such as pH or soil nitrogen availability. The more likely reason was the phytotoxic effects of SCG caused by high levels of caffeine, tannins and polyphenols (Hardgrove and Livesley, 2016)
- composting is a viable way of valorising this residue (Santos et al., 2017)

Composting using earthworms (vermicomposting) is considered the most advanced composting method (Lim et al., 2016). Vermicomposting is a biooxidative and stabilizing process of organic materials conversion, which, unlike classical composting, uses interactions between earthworms and microorganisms, and does not involve the decomposition thermophilic phase (Domínguez and Edwards, 2011). It is evident that earthworms and endosymbiotic microbes during vermicomposting tend to eliminate pathogens by enhancing enzymatic activities in both gut- and cast-associated processes. Pathogen reduction during vermicomposting can be plausibly attributed to direct actions like microbial inhibition due to intestinal enzymatic action, and secretion of coelomic fluids with antibacterial properties, as well as indirect actions like stimulation of endemic microbes leading to competition and antagonism. Further, the pathogen reduction during vermicomposting is largely selective, and earthworms exert a differential effect according to the earthworm species and whether the pathogen considered is Gram-positive or -negative, owing to its cell wall composition (Swati and Hait, 2018). Digging, fragmentation, and aeration are provided to a greater extent by earthworms, making vermicomposting one of the low-cost waste treatment systems (Baghel et al., 2018). The technology is fully environmentally friendly (Abbasi et al., 2015).

Vermicomposts are characterized by very good maturity and stability, containing high-quality humic substances, enzymes, and plant growth hormones (Hanc et al., 2019a; Hřebečková et al., 2019a; Ravindran et al., 2016).

There are many reports on different valorization of SCG. Some of them are on classical composting. Only a few studies have been published on vermicomposting of this specific waste. Adi and Noor (2009) used *Lumbricus rubellus* for 49-day vermicomposting that was conducted after 21 days of pre-composting. Three different combination of treatments were prepared combined cow dung, kitchen waste and coffee grounds. The presence of coffee grounds showed higher percentage of nutrients in vermicompost produced. The data reveal that coffee grounds can be decomposed through vermicomposting and help to enhance the quality of vermicompost produced rather than sole use of kitchen waste in vermicomposting. Sanchez-Hernandez and Domínguez (2017) concluded that vermicomposting reduced substantially the residue mass of SCG in the very short term, yielding a nutrient-rich and enzymatically active vermicompost. González-Moreno et al. (2020) conducted 60-day laboratory vermicomposting experiment with 9 treatments differed with proportions of horse manure, spent coffee grounds (SCG) and coffee silver skin (CS). Best options were treatments with a medium–low amount of residue (25% for SCG and 25% or 50% for CS) due to the specific characteristics of these wastes and possible toxicity. They recommended that SCG is used with other amendments or alkaline residues in order to increase the pH level..

In comparison with the above studies, the main novelties of our work include using of straw pellets as amendment material, system with continuous feeding of earthworms where layers of different ages can be evaluated and effect of earthworms on reduction of caffeine.

The aim of the study was to investigate the feasibility of the vermicomposting SCG and its mixtures with straw pellets on the basis of: i) survival and development of earthworms, ii) occurrence of the main groups of microorganisms, iii) enzyme activity, iiii) physico-chemical parameters, and iiiii) caffeine content.

Our research results are useful for food waste producers, small-scale processors and large-scale vermicomposting plants, and vermicompost users, especially growers.

2. Materials and methods

2.1. Raw material and earthworms

A SCG mixture of coffee varieties from the La Divina Providencia from Nicaragua, Karuhiu Uhteri from Kenya, and Nensebo from Ethiopia was used for the vermicomposting experiment. It was obtained from Misto Café in Prague (N 50°5.94017′, E 14°24.26082′). After receiving this material, the SCG were refrigerated in dark sealed bags. Due to the lesser pH and C:N ratio in SCG, moistened straw pellets with 34% dry matter content was used in the experiment. The pellets exhibited an alkaline pH (7.9). The EC was 2 times greater, and the C:N 4 times greater compared with the SCG (Table 1). *Eisenia andrei* earthworms were used in the study. It belongs to the group of epigeic earthworms. It is one of the most commonly used earthworms for vermicomposting in a mild climate.

2.2. Experimental design

The experiment was set up under laboratory conditions in plastic vermicomposters Worm Factory with four perforated trays of individual size 40 × 40 × 18 cm, marked from oldest (I) to youngest layer (IV). They were gradually filled with biowaste every 6 weeks during 6 months. Five treatments were established:

1: SCG 100% vol. with *Eisenia andrei* (layers I to IV)
2: SCG 75% vol. + straw pellets 25% vol. with *Eisenia andrei* (layers I to IV)
3: SCG 50% vol. + straw pellets 50% vol. with *Eisenia andrei* (layers I to IV)
4: SCG 25% vol. + straw pellets 75% vol. with *Eisenia andrei* (layers I to IV)
5: SCG 50% vol. + straw pellets 50% vol. without earthworms (layers I to IV)

For the four first treatments, 10*L* of bedding layer containing grape marc with earthworms *Eisenia andrei* (50 earthworms per liter) was put down. After that, new layer – 15 L of feedstocks was placed into the new tray above. The top of the vermicomposter was covered with a composting fabric and a plastic lid. To prevent the earthworms from escaping or crawling among vermicomposters, the experiment was conducted in constant light. The room air temperature was maintained at 22 °C. Every 12 h the room air was replaced with outdoor air.

Three 1 kg samples were taken from each layer and weighed. All potential earthworms were separated manually, counted, and weighed from each sample taken. About 50 g of the remaining sample was stored in a refrigerator (temperature 4 °C). Another 500 g portion was placed in a drying room and dried for about 14 days at a constant temperature of

Table 1
Physico-chemical parameters of SCG and wet straw pellets used in the study.

	Dry matter[%]	pH/H_2O	EC[μS/cm]	ORP[mV]	C/N	$P_{tot.}$[mg/kg]	$K_{tot.}$[mg/kg]	$Mg_{tot.}$[mg/kg]	Caffeine[mg/g]
SCG	34.5 ± 1.0	6.0 ± 0.1	723 ± 47	45.5 ± 6.6	17.7 ± 1.3	1301.7 ± 131.4	9904.3 ± 917.4	1586.0 ± 238.6	3.27 ± 0.14
Straw pellets	34.2 ± 2.4	7.9 ± 0.1	1345 ± 59	−30.5 ± 3.1	65.7 ± 7.0	1100 ± 100	9300 ± 600	900 ± 100	n.d.

Values are means ± SD; n.d. – no data.
pH, EC, and ORP were determined in wet matter; other parameters in dry matter
SCG = spent coffee grounds

35 °C, and then ground. The remaining sample was frozen at −25 °C and lyophilized.

2.3. Physico-chemical and biological analyses

2.3.1. Analyses from refrigerated samples

To determine the pH, electrical conductivity (EC) and oxidation–reduction potential (ORP), a 10 g sample was weighed into a sealable flask, and then 50 ml of demineralized water was added. The mixture was then shaken for 10 min. Values of pH and ORP were measured with a calibrated meter (WTW 340i). Then, the sample was filtered and the EC was measured with the WTW cond 730 conductometer.

2.3.2. Analyses from dried samples

For total carbon (C_{tot}) and nitrogen (N_{tot}) determination, the CHNS Vario MACRO cube analyzer (Elementar Analysensysteme GmbH, Germany), was used. The total contents of macronutrients (P, K, and Mg) were determined by decomposition obtained by pressurized wet-ashing (HNO_3 + H_2O_2) of dried samples in a closed system of Ethos 1 (MLS GmbH, Germany). The contents of ammonium and nitrate nitrogen (N-NH_4^+, N-NO_3^-), dissolved organic carbon (DOC) and the available nutrients (P, K, and Mg) were determined in CAT solution, which is a mixture of 0.01 mol/L $CaCl_2$ and 0.002 mol/L diethylene triamine pentaacetic acid (DTPA) (1:10 w/v), according to the international BSI EN 13651, 2001. The N-NH_4^+, N-NO_3^-, and DOC contents in the extracts were measured using the SKALAR SANPLUS SYSTEM® (the Netherlands). The total and available element concentrations were determined using ICP-OES (VARIAN VistaPro, Australia).

2.3.3. Analyses from lyophilized samples

Phospholipid fatty acid (PLFA) analysis were determined in the samples (in triplicates) according to Stella et al., 2015. The samples were extracted using phosphate buffer, chloroform, and methanol (0.8:1:2; v/v/v) according to Bligh and Dyer, 1959. Gas chromatography-mass spectrometry (GC–MS; 450-GC, 240-MS Varian, Walnut Creek, CA, USA) was employed for determination of methylated esters of fatty acids. The authentic chemical standards were obtained from Sigma-Aldrich, Prague, Czech Republic and Matreya LLC, USA. Actinobacterial biomass was estimated as the sum of 10Me-16:0, 10Me-18:0, 10Me-17:0, biomass of gram positive bacteria (G +): a17:0, i17:0, i16:0, i15:0, a15:0, and i14:0; biomass of gram negative bacteria (G −): 16:1ω5,16:1ω7, cy17:0, 18:1ω7, and cy19:0. The total bacterial biomass was determined on the basis of 15:0, 17:0, 16:1ω7, and 16:1ω9, together with the other above mentioned bacterial PLFA. Fungi were estimated according to 18:2ω6,9.

The enzymatic activities of hydrolytic enzymes were measured in 96-well microplates. The mixture of lyophilized sample (0.2 g) and acetate buffer (20 ml; pH 5.0; c = 50 mmol/L) was homogenized using the Ultra-Turrax (IKA Labortechnik, Germany) according to Štursová and Baldrian (2011). The substrates for these hydrolytic enzymes were as follows: for β–D–glucosidase it was MUFG (c = 2.75 mmol/L), which is a mixture of 4-methylumbellyferyl-β-D-glucopyranoside and dimethyl sulfoxide, for acid phosphatase it was MUFP(c = 2.75 mmol/L), which is a mixture of 4-methylumbellyferyl-phosphate and dimethyl sulfoxide, for arylsulphatase it was MUFS (c = 2.50 mmol/L), which is a mixture of 4-methylumbellyferyl sulphate potassium salt and dimethyl sulfoxide, for lipase it was MUFY (c = 2.50 mmol/L), which is a mixture of 4-methylumbellyferyl-caprylate and dimethyl sulfoxide, for chitinase it was MUFN (c = 1.00 mmol/L), which is a mixture of 4-methylumbellyferyl-N-acetylglucosaminide and dimethyl sulfoxide, for cellobiohydrolase it was MUFC (c = 2.50 mmol/L), which is a mixture of 4-methylumbellyferyl-N-cellobiopyranoside and dimethyl sulfoxide, for alanine aminopeptidase it was AMCA (c = 2.50 mmol/L), which is a mixture of L-alanine-7-amido-4-methylcoumarin and dimethyl sulfoxide, for leucine aminopeptidase it was AMCL (c = 2.50 mmol/L), which is a mixture of L-leucine-7-amido-4-methylcoumarin and dimethyl sulfoxide. Enzymes were measured as a fluorescence change using the Tecan Infinite® M200 (Austria) after 5 min and 125 min of incubation (40 °C) according to Baldrian (2009).

Caffeine was analyzed in the lyophilized samples using liquid chromatography-tandem mass spectrometry (LC-MS/MS). At first, the lyophilised samples were extracted using Accelerated Solvent Extractor ASE 200 (Dionex; Palaiseau, France) with heated methanol as an extraction solvent (temperature 80 °C, pressure 10.3 MPa, 3 extraction cycles and 5-min static periods in between the cycles). Methanol extracts were appropriately diluted with 50% methanol and analysed with liquid LC-MS/MS (Cimetiere et al., 2013). The system consisted of Agilent 1260 Infinity II liquid chromatograph coupled with Agilent 6470 LC/TQ mass spectrometer equipped with Agilent Jet Stream electrospray ion source (Agilent Technologies, Santa Clara, CA, USA). The caffeine analyses was performed using chromatographic column Poroshell 120 2.7 μm, 3 mm × 100 mm (Agilent Technologies, Santa Clara, CA, USA). Injection volume was 2 μl, mobile phase consisted of 0.5 mM NH_4F in Milli-Q water (0.01% formic acid; Honeywell) (A) and methanol (Honeywell) (B); the flow rate was 0.4 ml min^{-1}. Column temperature throughout the analysis was maintained at 40 °C. Gradient elution was as follows (min/%B): 0/5; 1/20; 8 – 9/100; 9.1 – 12/5. Caffeine was monitored in positive ion mode, specific ion transitions (m/z) were: 195.1 → 83; 195.1 → 110 and 195.1 → 138 (fragmentor voltage: 105 V, collision energy: 20 eV). Following electrospray conditions were applied: drying gas temperature: 200 °C, drying gas flow 8 L/min, nebulizer pressure: 45 psi, sheath gas temperature: 400 °C, sheath gas flow: 12 L/min, capillary voltage: 2500 V, nozzle voltage: 0 V.

2.4. Statistical analysis

All the results are the means of three replicates. The tests of normality and homogeneity were performed. Since some data did not have normal and homogeneous distribution, a strict nonparametric Kruskal-Wallis ANOVA test ($P \leq 0.05$) was used with the help of STATISTICA 12 software (StatSoft, Tulsa, USA). Spearman's correlations were explored at the 0.05 probability levels.

3. Results and discussion

3.1. Earthworms

The youngest upper layers contained the greatest biomass and also a number of earthworms (Table 2). The increasing addition of straw pellets reduced the proportion of number and biomass of earthworms in the upper layer (number of earthworms: 88%, 64%, and 37%; earthworm

Table 2
Number and biomass of earthworms in individual treatments and layers.

Layer (age)	100% SCG(Treatment 1)		75% SCG + 25% straw pellets (Treatment 2)		50% SCG + 50% straw pellets (Treatment 3)		25% SCG + 75% straw pellets (Treatment 4)	
	Number [pcs/kg]	E. biomass [g/kg]	Number [pcs/kg]	E. biomass [g/kg]	Number [pcs/kg]	E. biomass [g/kg]	Number [pcs/kg]	E. biomass [g/kg]
IV(45 days)	42 ± 5	12.0 ± 1.7	99 ± 24	38.7 ± 6.5	70 ± 13	27.5 ± 3.4	629 ± 83	71.5 ± 7.1
III(90 days)	41 ± 18	13.0 ± 5.6	14 ± 3	4.2 ± 1.2	31 ± 8	13.6 ± 3.9	354 ± 66	28.4 ± 5.5
II(135 days)	6 ± 4	1.2 ± 0.8	0	0	8 ± 1	2.8 ± 1.4	320 ± 6	15.8 ± 0.6
I(180 days)	0	0	0	0	1 ± 1	0.2 ± 0.2	400 ± 51	16.0 ± 0.2

Values are means ± SD.
SCG = spent coffee grounds
E. biomass = Earthworm biomass
days in brackets = age of layers at the time of sampling

biomass: 90%, 62%, and 54% in treatments 2, 3, and 4, respectively), and thus increased their presence in the bottom and the middle layers. The biomass for layer I (age 180 days) was less for treatment 3 (0.2 g) than for treatment 4 (16 g). Their long presence in treatment 4 was due to the longer biodegradability because of the greater C/N ratio. Significant proportion of straw and, conversely, a lesser proportion of SCG created suitable conditions for earthworm survival (greater aeration and less bulk density of the mixture), and encouraged earthworms to multiply. Earthworm weight was calculated from Table 2 as ratio of earthworm biomass and number of earthworms, and decreased in direct proportion to the layer age. Compared to the other treatments, the earthworm weight was much lesser for treatment 4 (0,07 g). The fewest number and least biomass of earthworms were found in the first vermicomposter with 100% coffee grounds. They accounted for 4% of the total number and 10% of total earthworm biomass of all vermicomposters. The lesser earthworm biomass in SCG itself could be caused by the lesser carbon compound content as a source of energy for earthworms, and/or the relatively great density and thus lesser air content in SCG and pressure of the above layers. The toxicity of the SCG itself may be important here (Cervera-Mata et al., 2020). In our experiment, the greatest earthworm number, which exceeded all other treatments by about 15 times, was found in the fourth treatment with 75% straw pellets. Therefore, the description of further biological and chemical parameters will be directed to this treatment and treatment 3, where earthworms occurred in all layers.

3.2. Microorganisms

The oldest layers of treatment 3 and 4 contained by 23% and 11%, respectively, more total microbial biomass expressed as phospholipid

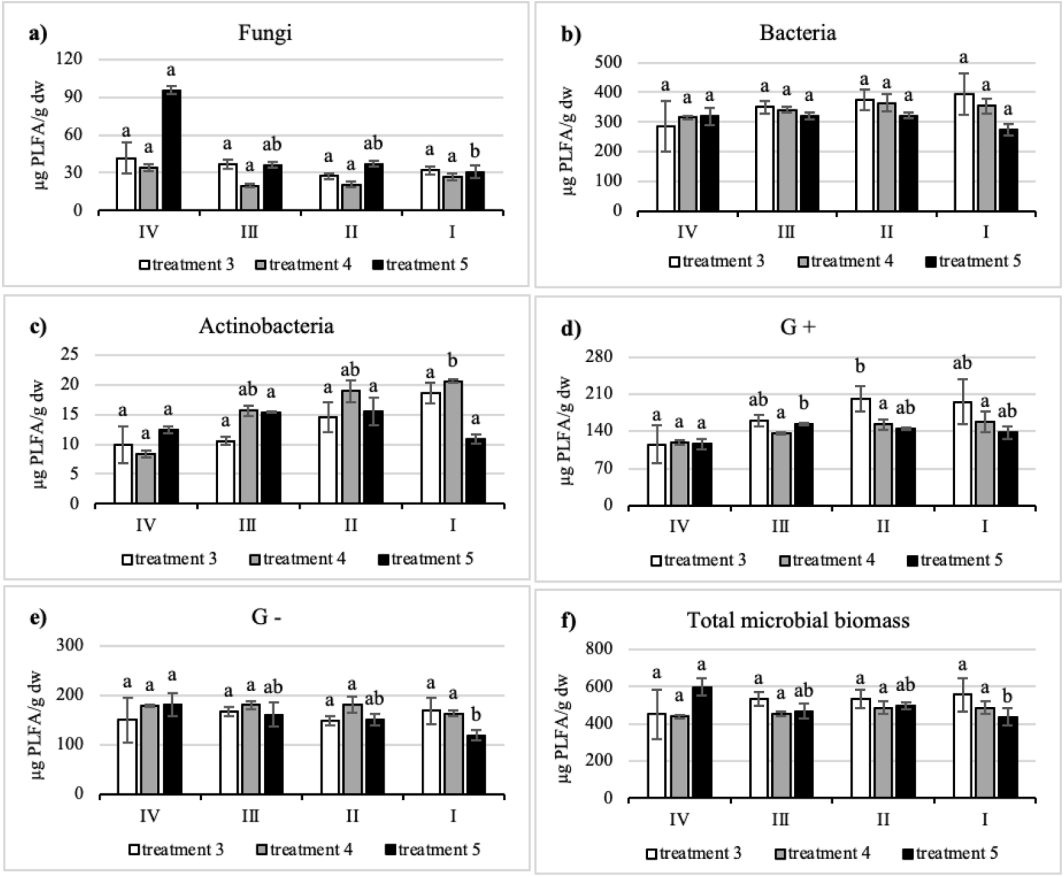

Fig. 1. Changes in the microbial biomass expressed as content of phospholipid fatty acids (PLFA) (a-f) in layers IV, III, II, and I of the treatments 3, 4, and 5. Values are the means ± SD. Letters indicate significant differences (Kruskal-Wallis test, $P \leq 0.05$) among layers within a treatment. dw – dry weight.

fatty acids (PLFA) than the youngest layer, in which the microorganisms were probably much represented in the earthworm digestive tracts (Fig. 1). This is confirmed by a strong indirect correlation between the total microbial biomass and earthworm number ($R = -0.94$, $p < 0.05$ for treatment 3; $R = -0.71$, $p < 0.05$ for treatment 4), and the total microbial biomass and earthworm biomass ($R = -0.92$, $p < 0.05$ for treatment 3; $R = -0.88$, $p < 0.05$ for treatment 4). Conversely, in treatment 5 without earthworms, which is identical to treatment 3 (with respect to composition), biomass decreased directly proportionally with the layer age. Fungi are a valuable food source for earthworms. Epigeic earthworms have enzymes in their digestive tract that allow them to digest fungi (Zhang et al., 2000). Likewise, treatments 3 and 4 contained 32% and 50%, respectively, less fungi than treatment 5 without earthworms. Conversely, the presence of bacteria was slightly greater in treatments 3 and 4, specifically by 14% and 11%, respectively, in comparison with treatment 5 without earthworms. A strong indirect correlation was found between bacteria and earthworm number ($R = -0.99$, $p < 0.05$ for treatment 3; $R = -0.91$, $p < 0.05$ for treatment 4), and between bacteria and earthworm biomass ($R = -0.99$, $p < 0.05$ for treatment 3; $R = -0.96$, $p < 0.05$ for treatment 4).

3.3. Enzyme activities

The activity of all enzymes varied in the individual treatments and layers depending on the specific enzyme (Fig. 2). The greatest enzymatic activity values were found in the non-earthworm treatment 5. At the beginning of the process in the youngest treatment layer, the activity of the 8 enzymes was 1.75 and 2.96 times greater compared to treatments 3 and 4, respectively. In the oldest layer, it was only 1.52 and 2.21-fold. This was probably due to the greater number and biomass of earthworms, where decomposition took place mainly in the earthworm bodies. Fifty-seven bacterial 16S rDNA clones, including enzyme-producing microorganisms, were identified in the intestines of *Eisenia fetida* by using polymerase chain reaction-denaturing gradient gel electrophoresis (PCR-DGGE) analysis (Hong et al., 2011). In our experiment, as the earthworm representation in layers III and II decreases, the enzymatic activity increases. In the oldest layer, in which the presence of earthworms slightly increased, enzymatic activity was reduced again. Activity of lipaze was the highest among 8 determined hydrolytic enzymes due to the high content of lipids in coffee, and in connection with caffeine, which promotes the breakdown of lipids.

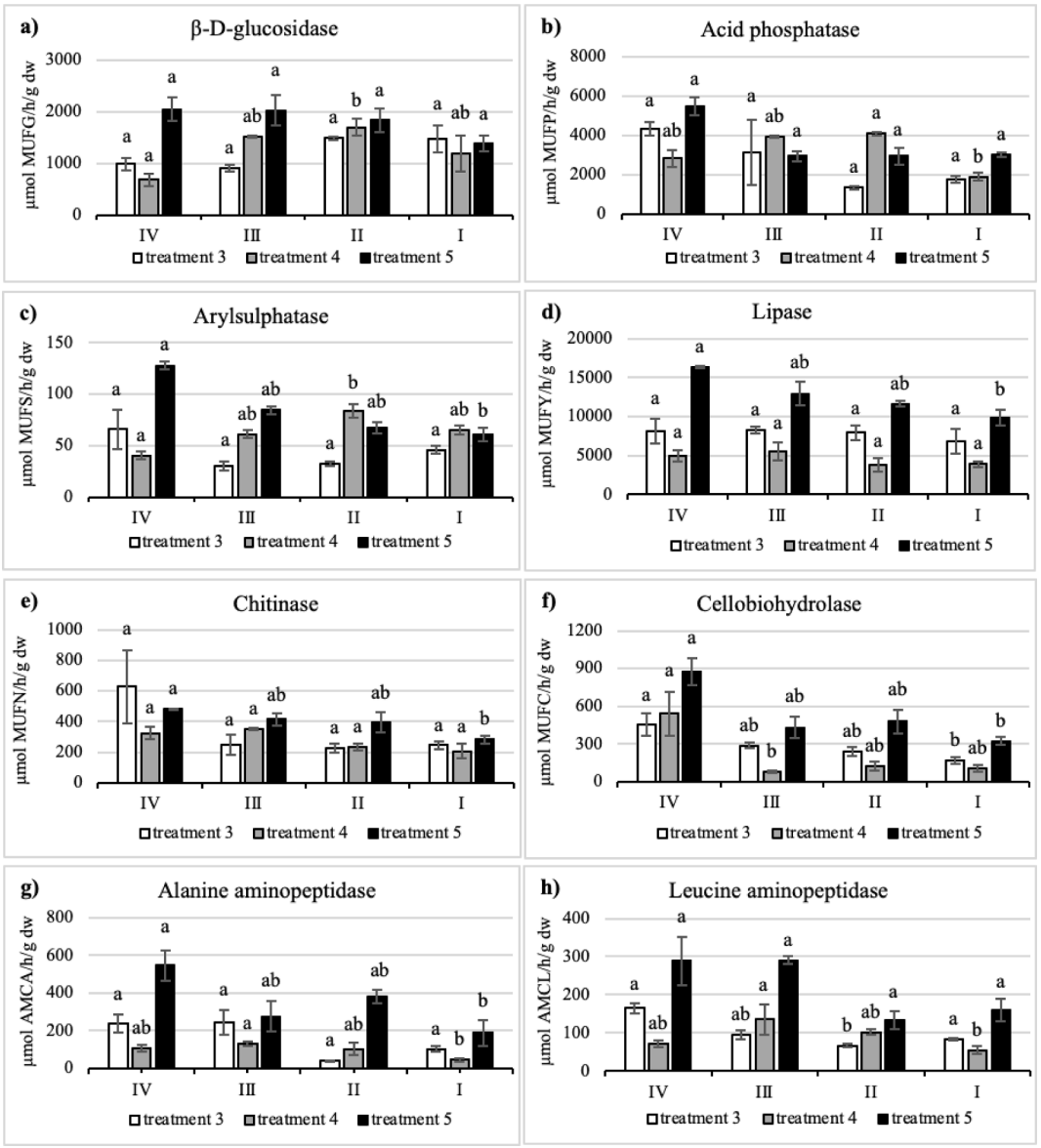

Fig. 2. Changes in enzymatic activity of hydrolytic enzymes (a-h) in layers IV, III, II, and I of the treatments 3, 4, and 5. Values are the means ± SD. Letters indicate significant differences (Kruskal-Wallis test, $P \leq 0.05$) among layers within a treatment. dw – dry weight.

Caffeine inhibits phosphodiesterase (an enzyme that catalyzes the hydrolysis of cyclic adenosinemonophosphate (cAMP)), leading to an increase in cAMP concentration. Thus, caffeine indirectly affects the regulation of cAMP-dependent protein kinases responsible for the regulation of glycogen, sugars and lipid metabolism. Activation of hormone-sensitive lipases leads to increased lipolysis, which causes increased plasma levels of free fatty acids and glycerol. There is also an increased release of catecholamines (Wahlang et al., 2018).

3.4. Physico-chemical parameters

The dry matter decreased from the youngest to the oldest layer (Table 3). The differences between layers IV and I were statistically significant. Among monitored treatments, the greatest increase was recorded in treatment 4 (by 47% between layers I and IV), where greater earthworm activity and proportion of straw pellets were important. Increasing moisture with layer age showed that the humified material (vermicompost) has a greater water absorption capacity than feedstock (Munnoli and Bhosle, 2011). The pH ranged from 7.1 to 8.1. In treatment 3 with earthworms, there was a significant pH increase over time, unlike the composition of an identical earthworm-free treatment 5, in which the pH was almost unchanged. There was a statistically significant difference between treatments 3 and 5 in the oldest layer I. There was no pH change in treatment 4, which was due to the greater proportion of straw pellets with their greater pH value (7.9). This probably did not give the possibility to increase the pH further. The EC decreased significantly (by 23%) in treatment 3 between layers IV and I. The layer I EC value was much less for treatment 3 (626 μS/cm) than for the earthworm-free treatment 5 (785 μS/cm). This can be explained by the fact that some of the salts were found in earthworm bodies removed from the sample prior to vermicompost analysis. In another 10-week SCG vermicomposting study, EC was maintained within the range of 0.64–0.70 ds m^{-1}, and treatments receiving cardboard had a lesser EC compared with the non-cardboard treatments (Liu and Price, 2011). Similar values and findings were obtained in our experiment. Redox potential gradually decreased with vermicompost age, which indicates gradual reduction of aerobic conditions. One of the important factors that reduced ORP in older layers was the pressure of the layers located above them. In control treatment 5, the ORP was by 35% and by 74% less compared to variants 3 and 4, respectively. This indicates that treatment 5 tended more to anaerobic conditions. On the contrary, earthworms have ability to intensively aerate material (Hanc and Dreslova, 2016) and thus ORP can increased. Earthworm number and earthworm biomass showed the same correlation with ORP in treatment 3 ($R = 0.91$, $p < 0.05$). In treatment 4, the ORP correlated more with earthworm biomass ($R = 0.70$, $p < 0.05$) than with earthworm number ($R = 0.42$, $p < 0.05$). The greater ORP in treatment 4 compared to treatments 3 and 5 was also due to the greater proportion of straw pellets having lesser bulk density and greater aeration ability. The C:N ratio decreased directly proportionally with increasing layer age. Treatment 4 exhibited the greatest C/N ratio, but there was the greatest decrease between layer IV and I (by 27%). For treatments 3 and 5, this decrease was only 8% and 20% respectively. The lesser C/N ratio in treatment 3 compared with treatment 5 (especially in the youngest layer) showed that earthworms accelerate biowaste decomposition. Wastes in which N-NH_4^+ values exceed 200 mg/kg of dry matter are unsuitable for vermicomposting, especially due to the volatilization of NH_3, which is lethal to earthworms (Míchal et al., 2019). N-NH_4^+ ranged between 40 and 50 mg/kg, and decreased during vermicomposting (Table 3). Lesser values were found in treatment 4 with 25% SCG compared with treatments 3 and 5, which suggests that the N-NH_4^+ level decreased with straw. Similarly, the N-NO_3^- values observed in treatment 4 were 6.5-fold less than in treatments 3 and 5, and accounted for only 0.03% of the total N content. With increasing composting time, the N-NO_3^- content increases (Hanc et al., 2017). This was confirmed in treatment 5 without earthworms, where after 180 days the N-NO_3^- value was 16

Table 3
Physico-chemical parameters in layers IV, III, II, and I of treatments 3, 4, and 5.

	50% SCG + 50% straw pelletswith earthworms (Treatment 3)	25% SCG + 75% straw pelletswith earthworms (Treatment 4)	50% SCG + 50% straw pelletswithout earthworms(Treatment 5)
Dry matter [%]			
IV (45 days)	19.9 ± 0.8 aA	22.6 ± 0.7 aA	24.6 ± 1.9 aA
III (90 days)	19.1 ± 0.8 abA	19.0 ± 0.7 abA	19.4 ± 0.6 abA
II (135 days)	17.4 ± 0.6 abAB	16.4 ± 0.3 abA	20.1 ± 1.1 abB
I (180 days)	16.5 ± 0.2 bAB	15.4 ± 0.5 bA	18.2 ± 0.2 bB
pH/H_2O			
IV (45 days)	7.2 ± 0 abA	7.8 ± 0.1 aB	7.3 ± 0 aAB
III (90 days)	7.1 ± 0 aA	7.9 ± 0 aB	7.2 ± 0.1 aAB
II (135 days)	7.6 ± 0.1 abAB	7.8 ± 0.1 aA	7.1 ± 0.1 aB
I (180 days)	8.1 ± 0.1 bA	7.8 ± 0 aAB	7.2 ± 0.1 aB
EC [μS/cm]			
IV (45 days)	808.7 ± 55.9 aA	664.7 ± 70.5 aA	767.0 ± 58.1 aA
III (90 days)	711.3 ± 19.7 abA	691.3 ± 37.0 aA	758.7 ± 166.7 aA
II (135 days)	766.7 ± 44.3 abA	742.0 ± 28.9 aA	805.7 ± 57.8 aA
I (180 days)	626.3 ± 33.9 bA	709.3 ± 46.7 aA	785.0 ± 127.5 aA
ORP [mV]			
IV (45 days)	36.8 ± 2.3 aAB	52.8 ± 7.1 aA	18.6 ± 2.3 aB
III (90 days)	25.0 ± 1.7 abA	51.5 ± 4.1 aB	13.4 ± 1.0 abA
II (135 days)	22.6 ± 2.2 abA	49.4 ± 7.6 aB	12.7 ± 2.6 abA
I (180 days)	8.3 ± 1.8 bA	44.1 ± 6.0 aB	8.2 ± 1.0 bA
C/N			
IV (45 days)	11.2 ± 0.3 abA	18.7 ± 0.7 aAB	13.6 ± 0.6 aB
III (90 days)	11.8 ± 0.1 aA	14.5 ± 0.1 abB	12.0 ± 0.2 abA
II (135 days)	11.1 ± 0.1 abAB	14.0 ± 0.4 abA	10.9 ± 0.4 abB
I (180 days)	10.3 ± 0.2 bA	13.6 ± 0.4 bB	10.3 ± 0.2 bA
N-NH_4^+ [mg N/kg]			
IV (45 days)	49.5 ± 0.2 aA	46.2 ± 2.9 aA	49.4 ± 0.7 aA
III (90 days)	47.2 ± 1.2 abA	46.7 ± 1.1 aA	45.3 ± 0.5 bA
II (135 days)	47.2 ± 0.4 abA	45.3 ± 1.6 aA	47.4 ± 0.7 abA
I (180 days)	45.4 ± 0.7 bAB	42.4 ± 1.2 aA	46.9 ± 1.2 abB
N-NO_3^- [mg N/kg]			
IV (45 days)	65.2 ± 3.1 aA	6.7 ± 2.4 abA	4.9 ± 0.3 aA
III (90 days)	65.2 ± 1.4 aAB	13.8 ± 0.7 aA	85.7 ± 0.7 bB
II (135 days)	48.1 ± 7.1 aAB	5.3 ± 0.8 bA	58.4 ± 0.5 abB
I (180 days)	33.6 ± 1.5 aAB	8.0 ± 0.8 abA	80.3 ± 1.1 abB

(continued on next page)

Table 3 (*continued*)

	50% SCG + 50% straw pelletswith earthworms (Treatment 3)	25% SCG + 75% straw pelletswith earthworms (Treatment 4)	50% SCG + 50% straw pelletswithout earthworms(Treatment 5)
$N\text{-}NH_4^+/N\text{-}NO_3^-$			
IV (45 days)	0.76 ± 0.04 abA	7.38 ± 2.08 abAB	10.15 ± 0.55 aB
III (90 days)	0.72 ± 0.01 aAB	3.39 ± 0.25 aA	0.53 ± 0.00 bB
II (135 days)	1.00 ± 0.14 abAB	8.71 ± 1.55 bA	0.81 ± 0.01 abB
I (180 days)	1.35 ± 0.08 bAB	5.33 ± 0.62 abA	0.58 ± 0.01 abB
DOC [mg C/kg]			
IV (45 days)	18808 ± 2440 aA	10084 ± 823 aA	16101 ± 2041 aA
III (90 days)	11875 ± 2468 abA	10560 ± 1706 aA	12910 ± 848 aA
II (135 days)	15025 ± 908 abA	11954 ± 1077 aA	12993 ± 1111 aA
I (180 days)	10245 ± 602 bA	10726 ± 651 aA	14898 ± 926 aA

Values are the means ± SD. Different lowercase letters in a column indicate significant differences among layers, capital letters indicate significant differences among treatments (Kruskal-Wallis test, $P \leq 0.05$).
SCG = spent coffee grounds; days in brackets = age of layers at the time of sampling

times greater. The nitrification index ($N\text{-}NH_4^+/N\text{-}NO_3^-$) is considered a parameter of compost and vermicompost maturity (Karak et al., 2017). There was a significant decrease in the $N\text{-}NH_4^+/N\text{-}NO_3^-$ ratio in treatment 5 between layers IV and III (45 and 90 days). In treatment 3 containing earthworms the $N\text{-}NH_4^+/N\text{-}NO_3^-$ ratio was on average 3 times less than in the identical treatment 5 without earthworms. Zhang and Sun (2017) found an indirect relationship between the increasing proportion of SCG and the nitrification index during green waste composting. Treatment 4 exhibited a much greater $N\text{-}NH_4^+/N\text{-}NO_3^-$ ratio than treatment 3 (Table 4). This result was probably due to the greater proportion of straw pellets, which increased the C/N ratio and thus provided more suitable conditions for earthworm activity, as seen previously (Biruntha et al., 2020). This is confirmed by the fairly even earthworm distribution in the whole profile of treatment 4. Earthworms are able to stabilize biowaste faster, as evidenced by the fact that after only 45 days the $N\text{-}NH_4^+/N\text{-}NO_3^-$ ratio was much less for treatment 3 (0.76), as opposed to treatment 5 (10.15). The proportion of DOC in the total carbon content was less in treatment 4 (2.6%) than in treatment 3 (3.2%). There was a decrease in DOC in treatments 3 and 5 between the youngest and the oldest layer by 45% and 7%, respectively. Although treatment 4 exhibited the least DOC, the values practically did not change, because the vermicomposting process was still occurring in all layers.

Table 4
Caffeine content in layers IV, III, II, and I of all studied treatments (ng/g of dry matter).

	IV (45 days)	III (90 days)	II (135 days)	I (180 days)
100% SCGwith earthworms (Treatment 1)	2447 ± 96.6 Aab	142.4 ± 21.6 bAB	2301 ± 89.1 abA	1748 ± 138.0 abAB
75% SCG + 25% straw pelletswith earthworms (Treatment 2)	95.4 ± 6.7 aAB	2791 ± 780.8 abAB	139.5 ± 51.3 abAB	7330 ± 218.9 bB
50% SCG + 50% straw pelletswith earthworms (Treatment 3)	42.0 ± 6.3 abAB	42.1 ± 4.6 abAB	73.9 ± 19.4 aAB	33.4 ± 2.3 bAB
25% SCG + 75% straw pelletswith earthworms (Treatment 4)	26.8 ± 1.9 aA	20.7 ± 0.6 abA	17.7 ± 0.8 bB	20.6 ± 0.5 abB
50% SCG + 50% straw pelletswithout earthworms (Treatment 5)	2174 ± 620.3 aB	4809 ± 48.3 abB	283.2 ± 43.6 abAB	136.8 ± 18.9 bAB

Values are the means ± SD. Different lowercase letters indicate significant differences among layers, capital letters indicate significant differences among treatments (Kruskal-Wallis test, $P \leq 0.05$).
SCG = spent coffee grounds
days in brackets = age of layers at the time of sampling

Total and available contents of basic macroelements are shown in Fig. 3. The contents were influenced by earthworm movement from the bedding layer and between the layers, the element contents in living earthworms, the decomposition of dead earthworm bodies, and conversely, the birth of new earthworms. The P_{tot} content ranged from 0.16 to 0.29%. There was an increase in P_{tot} content towards the bottom older layers, which was probably caused by organic matter mineralization. The greatest increase was recorded in treatment 4 (1.49-fold). The K_{tot} content was the greatest within the monitored elements (1.5 to 2.2%). The greatest increase of 1.25-fold was again seen in treatment 4. The average Mg_{tot} content was 0.24%, with values varying irregularly between layers. With the exception of Mg, the content of available macroelements increased over time. The percentage of the available contents of macronutrients (P, K, and Mg) on average in all of the layers and chosen treatments accounted for 47%, 45%, and 14%, respectively, on the total content. For vermicomposting of distillery residues, the proportion P, K, and Mg constituted 11%, 64%, and 10%, and in the case of kitchen waste 16%, 39%, and 2%, respectively (Hanc et al., 2019b; Hřebečková et al., 2019b). The greatest correlation between the total and available content in the layers was shown in treatment 5 (for P: $R = 0.94$, $p < 0.05$; for K: $R = 0.68$, $p < 0.05$; for Mg: $R = 0.95$, $p < 0.05$). In other treatments, the correlation was strongly influenced by earthworm movement and activity.

3.5. Caffeine

Table 4 shows the caffeine content in all experimental treatments. *Eisenia andrei* were able to decrease the caffeine content, as evidenced by the lesser caffeine content in treatment 3 (average of the layers 48 ng/g) compared to the control treatment 5 of the same composition without earthworms (average of the layers 1851 ng/g) which is 38 times less. The greater content in the control treatment 5 layer III could be caused by the natural movement of the extract from the upper layer. In treatments with a predominance of SCG, lesser caffeine values were found in the younger layers. In treatments 3 to 5, on the contrary, lesser caffeine values were found in the older layers.

Although caffeine has a negative effect on the environment, it is a recognized stimulator of the central nervous system and beneficial health effects have also been described. By using low-pressure or supercritical CO_2 extraction within the biorefinery of SCG extract it is possible to obtain between 0.734 and 41.3 μg/mg of caffeine which corresponded to 18–48% of extracted compounds from coffee beans, and 8–31% from roasted coffee (Santos et al., 2021).

4. Conclusions

Due to the content of toxic substances, SCG itself are not suitable for vermicomposting. The addition of straw pellets to the SCG (up to 75% vol.) improved aeration and reduced bulk density, resulting in the development of earthworms. Strong indirect correlation between the total microbial biomass and earthworm number was found caused by the presence of microorganisms in digestive tract of earthworms. Vermicomposting of SCG was characterized by very strong activity of lipaze due to the high content of lipids and caffeine in coffee. Vermicomposting increased the content of P, K and Mg and decreased the content of caffeine.

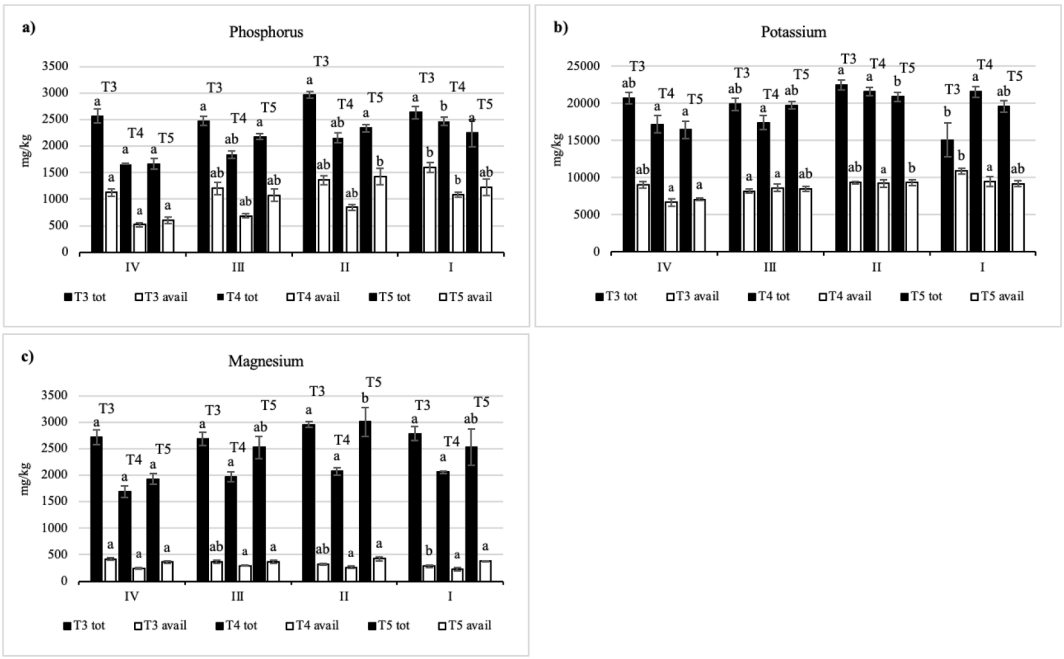

Fig. 3. Total and available content of phosphorus, potassium, and magnesium (a-c) in layers IV, III, II, and I of the treatments 3, 4, and 5. Values are the means ± SD. Letters indicate significant differences (Kruskal-Wallis test, $P \leq 0.05$) among layers within a treatment. T3 – treatment 3 (50% SCG + 50% straw pellets with earthworms), T4 – treatment 4 (25% SCG + 75% straw pellets with earthworms), T5 – treatment 5 (50% SCG + 50% straw pellets without earthworms), tot – total content, avail – available content.

CRediT authorship contribution statement

Ales Hanc: Conceptualization, Methodology, Investigation, Visualization, Funding acquisition, Project administration. **Tereza Hrebeckova:** Resources, Formal analysis, Data curation. **Alena Grasserova:** Formal analysis, Methodology. **Tomas Cajthaml:** Conceptualization, Data curation.

Declaration of Competing Interest

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

Acknowledgements

This work was supported by the Ministry of Agriculture of the Czech Republic under NAZV project No. QK1910095. The authors would like to thank Sona Houzvova from KOKOZA, o.p.s. for consultations on SCG issue and Christina Baker Starrman for revision of the English text.

References

Abbasi, S.A., Nayeem-Shah, M., Abbasi, T., 2015. Vermicomposting of phytomass: limitations of the past approaches and the emerging directions. J. Clean. Prod. 93, 103–114.
Adi, A.J., Noor, Z.M., 2009. Waste recycling: Utilization of coffee grounds and kitchen waste in vermicomposting. Bioresour. Technol. 100 (2), 1027–1030.
Alemayehu, Y.A., Asfaw, S.L., Tirfie, T.A., 2020. Management options for coffee processing wastewater. A review. J. Mater. Cycles Waste 22 (2), 454–469.
Baghel, B., Sahu, R., Pandey, D., 2018. Vermicomposting an economical enterprise for nutrient and waste management for rural agriculture. Int. J. Curr. Microbiol. Appl. Sci. 7 (2), 3754–3758.
Baldrian, P., 2009. Microbial enzyme-catalyzed processes in soils and their analysis. Plant Soil Environ. 55 (No. 9), 370–378.
Ballesteros, L.F., Teixeira, J.A., Mussatto, S.I., 2014. Chemical, functional, and structural properties of spent coffee grounds and coffee silverskin. Food Bioprocess. Technol. 7 (12), 3493–3503.
Biruntha, M., Karmegam, N., Archana, J., Karunai Selvi, B., John Paul, J.A., Balamuralikrishnan, B., Chang, S.W., Ravindran, B., 2020. Vermiconversion of biowastes with low-to-high C/N ratio into value added vermicompost. Bioresour. Technol. 297, 122398. https://doi.org/10.1016/j.biortech.2019.122398.
Bligh, E.G., Dyer, W.J., 1959. A rapid method of total lipid extraction and purification. Can. J. Biochem. Phys. 37 (8), 911–917.
Blinová, L., Bartošová, A., Sirotiak, M., 2017. Biodiesel production from spent coffee grounds. Research Papers Faculty of Materials Science and Technology Slovak University of Technology, 25, 113–121.
BSI EN 13651, 2001. Soil Improvers and Growing Media - Extraction of Calcium Chloride/DTPA (CAT) Soluble Nutrients.
Cervera-Mata, A., Navarro-Alarcón, M., Rufián-Henares, J.Á., Pastoriza, S., Montilla-Gómez, J., Delgado, G., 2020. Phytotoxicity and chelating capacity of spent coffee grounds: Two contrasting faces in its use as soil organic amendment. Sci. Total Environ. 717, 137247. https://doi.org/10.1016/j.scitotenv.2020.137247.
Cimetiere, N., Soutrel, I., Lemasle, M., Laplanche, A., Crocq, A., 2013. Standard addition method for the determination of pharmaceutical residues in drinking water by SPE–LC–MS/MS. Environ. Technol. 34 (22), 3031–3041.
Domínguez, J., Edwards, C.A., 2011. Relationships between composting and vermicomposting. in: Edwards, C.A., Arancon, N.Q., Sherman, R. (Eds.), Vermiculture Technology. CRC Press, Taylor & Francis Group, Boca Raton, pp. 11–25.
Esquivel, P., Jiménez, V.M., 2012. Functional properties of coffee and coffee by-products. Food Res. Int. 46 (2), 488–495.
González-Moreno, M.A., García Gracianteparaluceta, B., Marcelino Sádaba, S., Zaratiegui Urdin, J., Robles Domínguez, E., Pérez Ezcurdia, M.A., Seco Meneses, A., 2020. Feasibility of vermicomposting of spent coffee grounds and silverskin from coffee industries: A laboratory study. Agronomy 10 (8), 1125. https://doi.org/10.3390/agronomy10081125.
Hanc, A., Dreslova, M., 2016. Effect of composting and vermicomposting on properties of particle size fractions. Bioresour. Technol. 217, 186–189.
Hanc, A., Enev, V., Hrebeckova, T., Klucakova, M., Pekar, M., 2019a. Characterization of humic acids in a continuous-feeding vermicomposting system with horse manure. Waste Manage. 99, 1–11.
Hanc, A., Hrebeckova, T., Kuzel, S., 2019b. Vermicomposting of distillery residues in a vertical-flow windrow system. Waste Biomass Valori. 10 (12), 3647–3657.
Hanc, A., Ochecova, P., Vasak, F., 2017. Changes of parameters during composting of bio-waste collected over four seasons. Environ. Technol. 38 (13-14), 1751–1764.
Hardgrove, S.J., Livesley, S.J., 2016. Applying spent coffee grounds directly to urban agriculture soils greatly reduces plant growth. Urban for. urban gree. 18, 1–8.
Hong, S.W., Lee, J.S., Chung, K.S., 2011. Effect of enzyme producing microorganisms on the biomass of epigeic earthworms *(eisenia fetida)* in vermicompost. Bioresour. Technol. 102 (10), 6344–6347.
Hřebečková, T., Hanč, A., Roy, A., 2019a. Changes of chemical and biological parameters during vermicomposting of kitchen biowaste with an emphasis on pathogens. Waste Forum 2019, 187–196.
Hřebečková, T., Wiesnerová, L., Hanč, A., 2019b. Changes of enzymatic activity during a large-scale vermicomposting process with continuous feeding. J. Clean. Prod. 239, 118127. https://doi.org/10.1016/j.jclepro.2019.118127.

International Coffee Organization, 2020. http://www.ico.org/documents/cy2019-20/cmr-1219-e.pdf. Accessed 28 January 2020.
Karak, T., Kutu, F.R., Paul, R.K., Bora, K., Das, D.K., Khare, P., Das, K., Dutta, A.K., Boruah, R.K., 2017. Co-composting of cow dung, municipal solid waste, roadside pond sediment and tannery sludge: role of human hair. Int. J. Environ. Sci. Technol. 14 (3), 577–594.
Kovalcik, A., Obruca, S., Marova, I., 2018. Valorization of spent coffee grounds: A review. Food Bioprod. Process 110, 104–119.
Li, S., Wen, J., He, B., Wang, J., Hu, X., Liu, J., 2020. Occurrence of caffeine in the freshwater environment: Implications for ecopharmacovigilance. Environ. Pollut. 263, 114371. https://doi.org/10.1016/j.envpol.2020.114371.
Lim, S.L., Lee, L.H., Wu, T.Y., 2016. Sustainability of using composting and vermicomposting technologies for organic solid waste biotransformation: recent overview, greenhouse gases emissions and economic analysis. J. Clean. Prod. 111, 262–278.
Liu, K., Price, G.W., 2011. Evaluation of free composting systems for the management of spent coffee grounds. Bioresour. Technol. 102, 7966–7974.
Low, J.H., Rahman, W.A.W.A., Jamaluddin, J., 2015. The influence of extraction parameters on spent coffee grounds as a renewable tannin resource. J. Clean. Prod. 101, 222–228.
McNutt, J., He, Q.(., 2019. Spent coffee grounds: A review on current utilization. J. Ind. Eng. Chem. 71, 78–88.
Míchal, P., Hanč, A., Švehla, P., 2019. Inhibiting effect of ammonia during vermicomposting of sewage sludge and the possibilities of its elimination. Waste Forum 2019, 144–152.
Munnoli, P.M., Bhosle, S., 2011. Water-holding capacity of earthworms' vermicompost made of sugar industry waste (press mud) in mono- and polyculture vermireactors. Environmentalist 31 (4), 394–400.
Murthy, P.S., Madhava Naidu, M., 2012. Sustainable management of coffee industry by-products and value addition: A review. Resour. Conserv. Recy. 66, 45–58.
Mussatto, S.I., Machado, E.M.S., Martins, S., Teixeira, J.A., 2011. Production, composition, and application of coffee and its industrial residues. Food Bioprocess. Technol. 4 (5), 661–672.
Nzekoue, F.K., Khamitova, G., Angeloni, S., Sempere, A.N., Tao, J., Maggi, F., Xiao, J., Sagratini, G., Vittori, S., Caprioli, G., 2020. Spent coffee grounds: A potential commercial source of phytosterols. Food Chem. 325, 126836. https://doi.org/10.1016/j.foodchem.2020.126836.
Obruca, S., Benesova, P., Petrik, S., Oborna, J., Prikryl, R., Marova, I., 2014. Production of polyhydroxyalkanoates using hydrolysate of spent coffee grounds. Process Biochem. 49 (9), 1409–1414.
Peshev, D., Mitev, D., Peeva, L., Peev, G., 2018. Valorization of spent coffee grounds – A new approach. Sep. Purif. Technol. 192, 271–277.
Ravindran, B., Wong, J.W.C., Selvam, A., Sekaran, G., 2016. Influence of microbial diversity and plant growth hormones in compost and vermicompost from fermented tannery waste. Bioresour. Technol. 217, 200–204.
Sanchez-Hernandez, J.C., Domínguez, J., 2017. Vermicompost derived from spent coffee grounds: assessing the potential for enzymatic bioremediation. In: Galanakis, C. (Ed.), Handbook of Coffee Processing By–product. Academic Press, Elsevier, London, pp. 369–398.
Santos, C., Fonseca, J., Aires, A., Coutinho, J., Trindade, H., 2017. Effect of different rates of spent coffee grounds (SCG) on composting process, gaseous emissions and quality of end-product. Waste Manage. 59, 37–47.
Santos, É.M.D., Macedo, L.M.d., Tundisi, L.L., Ataide, J.A., Camargo, G.A., Alves, R.C., Oliveira, M.B.P.P., Mazzola, P.G., 2021. Coffee by-products in topical formulations: A review. Trends Food Sci. Technol. 111, 280–291.
Stella, T., Covino, S., Burianová, E., Filipová, A., Křesinová, Z., Voříšková, J., Větrovský, T., Baldrian, P., Cajthaml, T., 2015. Chemical and microbiological characterization of an aged PCB-contaminated soil. Sci. Total Environ. 533, 177–186.
Swati, A., Hait, S., 2018. A comprehensive review of the fate of pathogens during vermicomposting of organic wastes. J. Environ. Qual. 47 (1), 16–29.
Štursová, M., Baldrian, P., 2011. Effects of soil properties and management on the aktivity of soil organic matter transforming enzymes and the quantification of soil-bound and free activity. Plant Soil 338, 99–110.
Tongcumpou, C., Usapein, P., Tuntiwiwattanapun, N., 2019. Complete utilization of wet spent coffee grounds waste as a novel feedstock for antioxidant, biodiesel, and biochar production. Ind. Crop Prod. 138, 111484. https://doi.org/10.1016/j.indcrop.2019.111484.
Wahlang, B., McClain, C., Barve, S., Gobejishvili, L., 2018. Role of cAMP and phosphodiesterase signaling in liver health and disease. Cell. Signal. 49, 105–115.
Yordanov, D., Mustafa, Z., Milina, R., Tsonev, Z., 2016. Multi-criteria optimisation process of the oil extraction from spent coffee ground by various solvents. Oxid. Commun. 39, 1478–1487.
Zhang, B., Li, G., Shen, T., Wang, J., Sun, Z., 2000. Changes of microbial biomass C, N, and P and enzyme activities in soil incubated with the earthworms *Metaphire guillelmi* or *Eisenia fetida*. Soil Biol. Biochem. 32, 2055–2062.
Zhang, L.u., Sun, X., 2017. Using cow dung and spent coffee grounds to enhance the two-stage co-composting of green waste. Bioresour. Technol. 245, 152–161.