ORIGINAL ARTICLE



A biological insight of hops wastes vermicomposting by Eisenia Andrei

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Received: 20 March 2023 / Accepted: 23 October 2023 / Published online: 18 December 2023 © The Author(s) 2023

Abstract

The study was conducted to assess the feasibility of using *Eisenia andrei* earthworms for vermicomposting hop remains from a lupulin extraction enterprises for the brewing industry. Vermicomposting process was conducted within 70 days using hop (*Humulus lupulus*) wastes blended with horse manure at five different ratios for triplicate in laboratory conditions. Number of worms, cocoons, and hatchlings were observed and recorded weekly as earthworm biomass, population build-up and reproduction biological parameters. The results showed an indirect relationship between the hop content and the growth and reproductive performance of the worms. Notwithstanding this fact, 100% of survival occurred in all combinations. A 50% blend of hop wastes and horse manure is suggested to ensure the optimizing usefulness of *E. andrei*. In addition, moment of maximum splendour of worm population build-up and reproduction parameters measured was achieved at around 40 or 50 days since the beginning of the test, seeing a clear and widespread decline from that moment.

Keywords Circular economy · Brewing industry · Waste recycling · Humulus lupulus · Eisenia andrei

Abbreviations

- CP Cocoon production.
- GR Growth rate.
- HM Horse manure.
- HW Hop waste.
- NCE Number of clitellated earthworms.
- NC Number of cocoons.
- NE Number of earthworms.
- NH Number of hatchlings.
- SD Standard deviation.

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TB Total biomass. TBG Total biomass gain

Introduction

The global climate emergency situation currently represents a challenge in terms of the maximum use of natural resources. For this reason, at the present time it is working on the implementation of a real circular economy, where waste generation is minimized and, subproducts or remains become new, useful and usable resource or raw materials, avoiding inadequate disposal in landfills [1–3].

In application of the waste hierarchy implemented in the European Union, the following level form of recovery of organic waste other than abandonment, landfill disposal or another inefficient forms of recovery such as incineration, would be recycling [2]. For instance, through composting or similar techniques such as vermicomposting, which allow prioritizing the obtaining of quality products (compost and/ or vermicompost) to replace raw materials (chemical fertilizers), reducing greenhouse gas emissions and other environmental improvements [4].

Vermicomposting is one of the main worldwide composting techniques used to recycle agro-industries organic waste [5]. It can be carried out, either as a single treatment or also combined with composting as a pre-treatment [6], and despite the fact that the study its use began around the 70 s of the last century [7], today it is still a widely used naturebased solution for organic wastes transformation, such as distilled grape marc [8], olive mill wastewater [9], aquatic weed [10], lavender [11] or the invasive tree *Acacia dealbata* [12]. This is due to its great potential, low economic cost and its environmental respectful, in order to obtain a high quality compost applicable in the agricultural sector as fertilizer and improver of soil properties [13–16].

Hop is a perennial climbing plant of the *Cannabaceae* family, whose dried fruits are essential beer ingredients because they provide a pungent aroma and bitterness flavour [17]. According to the Agriculture and Rural Development of the European Commission data [18], there are about 2,600 hop farms spread across 14 EU countries. This means 26,500 hectares, which represents 60% of the total area used for this crop worldwide, and a production of 50,000 tons per year. As a consequence, the generation of waste from farms and industries where hops is used, is more than foreseeable.

New ways of managing hop sediments from brewing and fermentation of beer are constantly being sought and the existing ones improved as part of the circular economy [19]. Nevertheless, studies in vermicomposting have only focused on brewery industry wastes so far [20, 21], neglecting those from agribusiness for brewery production companies. Although it is true that, recently a traditional and an on-farm composting trials have been conducted. For instance, Afonso et al. [22] transformed hop wastes blended with manure and with wheat straw into compost, with varied results depending on the mixtures made. Luskar et al. [23] obtained positive results in on-farm composting from hop wastes mixed with different additives. In view of the bibliographic reported so far, an absence is noted in the scientific community of the transformation of hop waste through vermicomposting.

The aim of the study was to analyze the potential of this residue to be decomposed through vermicomposting employing *Eisenia andrei* [24] worms blended with mature horse manure (HM), focusing on biological parameters, in order to cover this scarcity of knowledge in this field. The choice of this species was because it is one of the most used worldwide [12].

To achieve this objective, a laboratory investigation was carried out. Different hop to manure combinations were considered and analyzed by means of biological parameters, such as population variations, sexual development, and reproduction rate, in order to understand the earthworm dynamics versus hop concentration in a ten-week vermicomposting process. Finally, with the present work we want to contribute new knowledge about the vermicomposting of this waste, hops, little studied until now.

Materials and methods

Earthworms, wastes and sampling design

E. andrei earthworms were purchased from Vermican Soluciones de Compostaje S.L., a local company specialising in composting in the Navarrese town of Cordovilla (Spain). Hop wastes (HW) were acquired from a company in the Navarrese town of Olite (Spain) called Montes de Cristal y Acero S.L., which grows and produces hop plants and obtains lupulin as raw material for brewery companies. Horse manure (HM) was collected in an Equine Center in the Navarrese town of Labiano (Spain). Both wastes were used dry for the preparation of the mixtures and their composition are shown in Table 1.

The design of the experiment was an adaptation of a model previously used by other authors [14, 25, 26]. In this particular case, 750 cm³-capacity plastic cups were used, filled each one with 200 g of different combinations of HW and HM. HW and HM were mixed and subjected to vermicomposting in the concentrations of 100:0 (Treatment 1 (T1)), 75:25 (T2), 50:50 (T3), 25:75 (T4) and 0:100 (T5) on a dry weight basis as is shown in Table 2. Those five treatments were carried out for triplicate during 10 weeks in total. In each combination eight young non-clitellated earthworms were added. All test samples were kept under the same moisture $(70 \pm 10\%)$ and environmental temperature $(25 \pm 3^{\circ}C)$ conditions during the investigation, using a darkened heating chamber inside a laboratory of the Public University of Navarre as a safeguard place for all the aforementioned combinations.

 Table 1 Initial physico-chemical properties and nutrients of hops wastes (HW) and horse manure (HM)

Physico-chemical param- eter	Unit	Hops wastes	Horse manure	
Moisture	%	67.2 ± 0.1	82.1±0.1	
pH	-	8.2 ± 0.1	7.8 ± 0.1	
Electric conductivity	$dS m^{-1}$	1.7 ± 0.1	2.9 ± 0.1	
Total organic matter	$g \ 100 \ g^{-1}$	46.2 ± 0.1	62.5 ± 0.1	
Organic carbon	$g \ 100 \ g^{-1}$	26.8 ± 0.1	36.3 ± 0.1	
Nitrogen	$g \ 100 \ g^{-1}$	0.7 ± 0.1	2.3 ± 0.1	
Phosphorus	$g \ 100 \ g^{-1}$	1.4 ± 0.1	3.7 ± 0.1	
Potassium	$g \ 100 \ g^{-1}$	14.1 ± 0.1	7.2 ± 0.1	
C:N ratio	-	13 ± 1	17±1	

Nutrient data are expressed on a dry weight (dw) basis

Values are means ± standard error

Standards used are Regulation (EC) No 2003/2003 of the European Parliament and of the Council of 13 October 2003 relating to fertilizers and the Spanish Royal Decree 506/ 2013, of June 28, on fertilizer products

Table 2The different combinations of hops wastes (HW) and horsemanure (HM) considered for the experimental vermicompostinginvestigation

Treatment	Name given	Horse manure (%)	Hop wastes (%)	
T1	100HW	0	100	
T2	HW75/HM25	25	75	
T3	HW50/HM50	50	50	
T4	HW25/HM25	75	25	
T5	Control	100	0	

Biological analysis

Survival, biomass formation and reproduction of earthworms are the best signs to analyse the vermicomposting process [27]. Changes in live weight of earthworms and newly produced cocoons were measured once a week in the plastic containers, following the method used previously by other authors, such as Esmaeili et al. [28] Singh and Suthar [29] or Yadav and Garg [30]. Earthworms and cocoons were separated from the parental waste mixture by hand sorting method. Worms were washed in tap water to remove adhered material from their body and weighted. Adult earthworms were then returned to their original test container whereas cocoons and hatchlings were taken out and put back in separate stock cultures. A similar procedure was used by Bhat et al. [27] or Parthasarathi et al. [31] in earlier studies, to cite a couple of examples.

The growth rate of the worms (GR) and cocoon production (CP) were calculated as shown in Eqs. (1) and (2), respectively.

Total biomass (TB), number of earthworms (NE), number of clitellated earthworms (NCE), number of cocoons (NC), number of hatchlings (NH) and rate of mortality were also taken into consideration and logged.

Statistical analysis

A statistical analysis was carried out using R computer software package. A 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 differences between combinations in the variables of interest. The normality and homoscedasticity of variance for all the variables involved were corroborated in all the cases.

Results and discussion

Both population growth, as well as reproductive issues, are two clear indicators to monitor the process [32]. In the current study, those aforementioned were measured for the epigeic earthworm *E. andrei* and assessed weekly throughout the vermicomposting period in different treatments and compared weekly till the age of 70 days. Data are presented in Table 3 and Table 4, with a format comparable to other previous studies (e.g. [33–36]).

$$GR\left(\frac{\frac{mg}{worm}}{week}\right) = \frac{Final \ weight - Initial \ weight}{Vermicomposting \ time*Average \ n^{\circ} \ earthworms}$$
(1)

$$CP(cocoons/worm/week) = \frac{Number of cocoons}{Vermicomposting time*Average n^{\circ} earthworms}$$
(2)

Table 3 Data of *E. andrei* built-up during the vermicomposting process of hops wastes (HW) and horse manure (HM) in the five different treatments

Treatment	Peak grow rate achieved (mg/worm/ week)	Mean grow rate (mg/worm/ week)	Initial biomass (g)	Final biomass (g)	Maximum bio- mass achieved in (week)	Survival (%)	Total number of hatchling (n°)	Juvenile bio- mass achieved (g)
T1	54.6 ± 42.5^{b}	$7.9 \pm 1.7^{\circ}$	1.06 ± 0.15^{a}	$1.69 \pm 0.06^{\circ}$	8th	100%	0.0 ± 0.0^{c}	0.00 ± 0.00^{d}
T2	$107.5 \pm 18.9^{\rm ab}$	11.2 ± 2.9^{bc}	$1.15\pm0.20^{\rm a}$	2.04 ± 0.04^{bc}	3rd	100%	$17.0 \pm 14.0^{\rm bc}$	0.83 ± 0.47 ^{cd}
Т3	115.0 ± 15.4^{ab}	18.3 ± 2.1^{ab}	1.06 ± 0.08^{a}	2.52 ± 0.11^{ab}	5th	100%	25.7 ± 0.6^{ab}	1.62 ± 0.29^{bc}
T4	$126.3 \pm 16.4^{\rm a}$	15.3 ± 4.6^{abc}	$1.04\pm0.07^{\rm a}$	$2.26\pm0.43^{\rm b}$	4th	100%	42.0 ± 5.6^{a}	2.07 ± 0.34^{ab}
T5	$121.3 \pm 18.4^{\rm a}$	22.4 ± 4.0^a	$1.23\pm0.23^{\rm a}$	$3.02\pm0.14^{\rm a}$	4th	100%	44.0 ± 13.2^{a}	2.78 ± 0.47^a

Values are mean \pm SD (n=3). Different letters (a, b, c and d) within the same columns indicate significant differences among times points (Tukey's test)

Treatment*	Clitellum devel- opment started in (week)	Peak number of clitellated earth- worms achieved	Clitellum regres- sion started in (week)	Total number of cocoons after 10 weeks	Cocoons appear- ance started in (week)	Cocoons per worm (cocoons/worm)	Cocoons pro- duction (cocoons/worm/ week)
T1	3rd	3.3 ± 0.6^{d}	8th	1.0 ± 1.7^{d}	7th	0.13 ± 0.22^{d}	0.01 ± 0.02^{d}
T2	2nd	$5.3 \pm 1.2^{\circ}$	8th	20.3 ± 5.5 ^{cd}	3rd	2.54 ± 0.69 ^{cd}	0.25 ± 0.07 ^{cd}
Т3	2nd	$6.0 \pm 0.0^{\mathrm{bc}}$	8th	36.7 ± 12.5^{bc}	3rd	$4.46 \pm 1.56^{\rm bc}$	$0.45 \pm 0.16^{\rm bc}$
T4	2nd	7.7 ± 0.6^{ab}	7th	53.3 ± 16.6^{ab}	3rd	6.67 ± 2.07^{ab}	0.67 ± 0.21^{ab}
T5	2nd	8.0 ± 0.0^{a}	7th	$73.3 \pm 10.7^{\rm a}$	3rd	$9.17 \pm 1.33^{\rm a}$	0.92 ± 0.13^{a}

Table 4 Data of reproduction by E. andrei during the vermicomposting process of hops wastes (HW) and horse manure (HM) in the different treatments

Values are mean \pm SD (n=3). Different letters (a, b, c and d) within the same columns indicate significant differences among times points (Tukey's test)

Earthworms development

The proper progress of vermicomposting will depend on the substrate used, which evidences into good fattening parameters and sexual maturity in general terms [32, 37, 38]. In this study, population growth, based on TB and GR in the different combinations of HW and HM, was significantly different (p-value < 0.05) throughout the vermicomposting period. All the results are shown in Figs. 1A and B, and Table 3. A positive growth of earthworm during the first weeks can be observed in general, with a subsequent progressive decrease later in time, these changes being



Vermicomposting period (Weeks)

Fig. 1 In a the evolution of the total earthworm biomass (g) during the vermicomposting process of hops wastes (HW) and horse manure (HM) mixed in five different combinations: 100HW (T1), HW75/ HM25 (T2), HW50/HM50 (T3), HW25/HM25 and control (T5). And in b, the evolution of the Growth Rate (GR) in mg/worm/week during the vermicomposting process of hops wastes (HW) and horse manure (HM) mixed in five different combinations: 100HW (T1), HW75/ HM25 (T2), HW50/HM50 (T3), HW25/HM25 and control (T5). Mean \pm SD values followed by different letters in the same week are different in terms of statistical significance (p-value < 0.05)(A)(B)

more evident with a greater amount of HM. The number of earthworms was also measured, but no change was undergone, keeping the number of eight at all times. The upward trend during the first weeks and the subsequent progressive decline were more noticeable in T5 and T4 (Fig. 1a). The highest TB was achieved in the 4th week for T5 (4.26 ± 0.31), followed by T4 (3.13 ± 0.42). A week earlier, T2 (2.27 ± 0.14) had already reached its maximum peak, while T3 (2.71 ± 0.07) required one more week. T1 (1.81 ± 0.14) took eight weeks to yield its highest value. Similar trends were observed by other researchers, who reported the maximum earthworm biomass amount on weeks 5 and 6 [39, 40] or week 8 [41]. The maximum weight gain followed by weight loss by the time of termination of the experiment was previously reported by other authors as well [40, 42-44]. Such a decline was corroborated by few earlier studies and could be explained by the aging of substrate materials [45], the exhaustion of food [40], the reduction of bioavailable nutrients [46] and the conversion of most of the raw substrate to final products [47]. In the case of HM, authors have attributed this weight loss to the conversion of most of the substrate to vermicompost, which cannot further support the worms growth, in concordance with the results of Suthar and Singh [48] for instance. On the other hand, HW was transformed into a kind of thick and earthy-looking paste, whose degradation was slower. Hence, identical TB has been maintained throughout the process, as can be observed in T1. This may have occurred because of the origin, due to HW coming from plants harvested in a green stage and not withered. Consequently, degradation might be slower and more arduous.

As far as GR is concerned, the maximum values were 126.3 ± 16.4 mg/worm/week in T4, followed by 121.3 ± 18.4 in T5, 115.0 ± 15.4 in T3 and 107.5 ± 18.9 in T2. T1 reached the worst maximum value with 54.6 ± 42.5 mg/worm/week (Fig. 1b). All of them occurred during the first week. In spite of the fact that negative values are observed, the final balance after ten weeks is positive. The best GR mean of E. andrei at the end of the vermicomposting period was 22.38 ± 3.95 mg/worm/week in T5, followed by T3 (18.25 ± 2.07) , T4 (15.29 ± 4.63) T2 (11.17 ± 2.94) and T1 (7.92 ± 1.68) . GR has been considered as an appropriate indicator to assess the earthworm growth in different wastes [44, 49]. The results obtained are far from the great results reported by both Elvira et al. [25], who cited a mean GR for E. andrei of 89,46 mg/worm/week using rabbit manure as feed, and Haimi [50], who informed an E. andrei GR of 75.32 mg/worm/week in batch cultures. Nevertheless, our results have a very close resemblance to earlier studies such as Cluzeau et al. [51], who obtained GR of 31.5 mg/worm/ week in batch cultures for immature E. andrei fed on HM

and peat, as well as o those of Elvira et al. [52], who recorded 8.75 mg/worm/week in pure cultures and values from 12.25 to 15.61 mg/worm/week in mixed cultures, employing E. andrei + Dendrobaena rubida and E. andrei + Lombricus rubellus respectively. The obtained results were also in concordance with the best GR values of González-Moreno et al. [14], who reported 30.73 and 23.59 mg/worm/week employing E. andrei fed by spent coffee grounds and coffee silverskin, both spiked with HM, respectively, although they also informed of negative GR values for various mixtures. The best combination for vermicomposting HW seems to be a 50%HW-50%HM blend. Moreover, the present study revealed that HW on its own cannot provide better growth medium and nourishment for earthworms. The fact that a waste does not work well on its own has been common in previous vermicomposting studies [14, 53, 54], making spiking with some kind of dung necessary.

Sexual development

Earthworms maturity was determined by visualizing the appearance of the clitellum [55]. Hence, a careful observation of each individual was carried out to analyze sexual development. Clitellum is the reproductive gland used for cocoon production which in mature earthworms generally forms an obvious band around the midsection segments [55]. NCE were statistically different between the different mixtures (p-value < 0.05).

The first clitellate individuals appeared on day 14 in all the combinations except T1, and on day 28 all individuals had the clitellum in T5. Domínguez et al. [39] and our study alike dated 15 days for sexual maturation. Nonetheless, these results, which are shown in Fig. 2, do not resemble other authors' such as Domínguez and Edwards [56], who reported that not all of the worms had developed a clitellum after 48 days employing E. andrei in pig manure. There is a wide disparity among different authors about the number of days for E. andrei to reach maturity, as shown by previous studies, which show ratios ranging from not reaching sexual maturity to beyond a month. Additionally, Elvira et al. [25] did not report clitellum losses during their study of the E. andrei specimens. In our study, clitellum regression took place, in general terms, after the 7th week. This is probably due to the aforementioned depletion of the substrate. Elvira et al. [57] reported clitellum regressed soon after maturity in dairy sludge spiked with cow manure, and furthermore, a notable scarce number of clitellated worms in pure paper sludge and an absence of clitellum in pure dairy sludge treatment. Neuhauser et al. [58] stated that the time needed for clitellum development varies in direct relationship with the nutrient abundance. In fact, Domínguez et al. [39] stated the difficult to compare their results with those of previous

Fig. 2 Evolution of the number of clitellated earthworms (NCE) during the vermicomposting process of hops wastes (HW) and horse manure (HM) in five different combinations. Mean \pm SD values followed by different letters in the same week are different in terms of statistical significance (p-value < 0.05)



works using different organic substrates and taking longer periods of time to reach sexual maturity. Years later, Mikunthan and Piratheban [59] emphasized this difficulty of comparison between studies. In view of the results obtained, the fact that HW does not provide those necessary nutritional conditions for a complete development of the earthworms can be confirmed.

Cocoon production

CP, besides earthworm weight, is a critical indicator for the growth of earthworms [44]. Edwards et al. [49] reported that the important difference of rates of CP in different organic wastes are related to the quality of the waste material used

as feed. In this study, NC and CP were respectively counted and calculated. Figure 3 shows the results.

NC produced by the earthworms were statistically different between the various combinations (p-value < 0.05) along the vermicomposting period, although a great dispersion of the data is evident. Enormous differences between treatments, as well as in terms of the deviations within the same combination in view of the standard deviations (SD), is not surprising since it has also been reported in previous experiments [11, 39]. CP started on the 3rd week in all treatments, except T1. NC increased along the different weeks, reaching their highest value on week 4 for T2, week 5 for T3 and T5, and week 6 for T2. After the maximum peak of these combinations, NC was a constant swing chart, although with a downward trend. It was only at the 7th week that the Fig. 3 Number of the cocoons (NC) produced by the earthworms at each week during the vermicomposting process of hops wastes (HW) and horse manure (HM) in five different combinations. Mean \pm SD values followed by different letters in the same week are different in terms of statistical significance (p-value < 0.05)



first cocoons in T1 were found. The peak value of CP was recorded in T5 (2.00 ± 0.54) in the 5th week, whereas the worse was T1 (0.22 ± 0.38) in the 9th week. T5 yielded the best mean CP during all the vermicomposting period with 0.92 ± 0.13 cocoons/worm/week. The trend was downward to a higher amount of HW, with values from the 0.67 ± 0.21 in T4 to the 0.01 ± 0.02 in T1. More data are available in Table 4.

Except for T1, the results are fully aligned with some of the literature published to date. For instance, Frederickson et al. [60] obtained ranges between 0.46 and 1.56 cocoons/ worm/week vermicomposting green wastes. Elvira et al. [57] reported CP between 0.055 and 1.12 cocoons/worm/ week feeding *E. andrei* with pure cultures and combination

mixtures of cow manure, dairy sludge and paper-mill sludge. Domínguez et al. [39] 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. Nevertheless, other authors reported higher results in earlier studies, obtaining *E. andrei* CP of 1.82 [51], 2.14 [25] or 3.08 cocoons/worm/week [50], which suggests that neither wastes were adequate enough.

Hatchling formation

All new juvenile earthworms were counted weekly until the 70th day by hand sorting. Formation of Fig. 4 Number of hatchlings (NH) appeared each week during the vermicomposting process of hops wastes (HW) and horse manure (HM) in five different combinations. Mean \pm SD values followed by different letters in the same week are different in terms of statistical significance (p-value < 0.05)



Vermicomposting period (Weeks)

hatchlings were significantly different between the mixtures (p-value < 0.05). Figure 4 shows the results and Table 4 gathers the data.

Hatchlings were observed for the first time in the 5th week in T2, T3, and T5. One additional week was necessary in T4. Hence, fifteen days were necessary between the first appearance of cocoon and hatchling, except for T4. These results are analogous to Kaur et al. [61], who also reported a 15-day period between the first appearance of cocoons and hatchling. Increase in hatchling formation during the present study is also supported by Kaur et al. [61], Chauhan and Singh [62] or Bhat et al. [27], who reported similar upward trends employing *Eisenia fetida* feeding with different wastes.

The maximum NH were observed in T3 (17.67 ± 0.58) on week 7. It was followed by T5 (16.00 ± 4.58) on week 8,

T4 (15.33 ± 5.51) on week 7, and T2 (9.00 ± 6.08) on week 8. The data for total NH and their weight after 10 weeks are shown in Table 3. Moreover, there was no hatchling in T1 and none of the hatchlings developed clitellum in feeds tested at the end of the investigation after ten weeks. Garg et al. [63] informed that there was no hatchling in camel waste in their investigation employing *E. fetida*. and hatchlings had not developed clitellum either. Gupta et al. [64] also reported no *E. fetida* hatchlings observed in water hyacinth treatment. The few cocoons produced in T1 are a direct consequence of the lack of hatchlings. In addition, the trend of a higher biomass to a lower quantity of HW, demonstrates that the residue commented throughout this document is not better than manure.

Mortality

Survival is crucial for the determination of the palatability, suitability and other any impact of wastes in order to carry out a correct and safety vermicomposting [11, 65, 66].

In this study, a 100% of survival of earthworms in all treatments occurred. Hence, hop do not have any toxic compounds that disturb the process and endanger the life of the earthworms.

Conclusions

- A laboratory-pilot-scale assay of vermicomposting of hop wastes have been performed in order to observe *E. andrei* behaviour in biological terms and contributing new knowledge to the scientific community. The results obtained successfully reveals that HW can be vermicomposted since no negative evidence has been observed in this regard.
- The absence of mortality confirms that it is not a dangerous fed material for earthworms neither their survival.
- In addition, moment of maximum splendour of earthworm population build-up and reproduction parameters measured was achieved at around forty or fifty days since the beginning of the test, seeing a clear and widespread decline from that moment.
- 3 weeks were taken necessary to see the first cocoons and five for the hatchlings. Nevertheless, the results also show that large quantities of this waste are not as appetizing as could be predicted initially.
- Combinations with up to 50% waste had better performance. The higher the percentage of HW is, more negative the impact on the earthworm growth and reproduction performance becomes.
- Hence, the authors recommend a 50% blend of HW spiked with HM for the vermicomposting of this waste.

Funding Open Access funding provided by Universidad Pública de Navarra. This research was funded by Government of Navarre and European Regional Development Fund (ERDF) grant number VER-MICOMPOSTAJE 4.0 (0011-1365-2019-000110) research project.

Declarations

Conflicts of interest Authors declare that there is no conflict of interest.

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References

- Castillo-González E, De Medina-Salas L, Giraldi-Díaz MR, Sánchez-Noguez C (2021) Vermicomposting: a valorization alternative for corn cob waste. Appl Sci 11:5692. https://doi.org/10. 3390/app11125692
- European Commission Waste Framework Directive. https://envir onment.ec.europa.eu/topics/waste-and-recycling/waste-frame work-directive_en. Accessed 15 Nov 2022
- Seco A, Espuelas S, Marcelino S et al (2020) Characterization of biomass briquettes from spent coffee grounds and xanthan gum using low pressure and temperature. Bioenergy Res. https://doi. org/10.1007/s12155-019-10069-8
- 4. Moreno Casco J, Moral Herrero R (2008) Compostaje. Mundi Prensa, Madrid España
- Maharjan KK, Noppradit P, Techato K (2022) Suitability of vermicomposting for different varieties of organic waste: a systematic literature review (2012–2021). Org Agric 12:581–602. https://doi. org/10.1007/s13165-022-00413-2
- Blesa Marco ZE, Sáez JA, Pedraza Torres AM et al (2023) Effect of agricultural microplastic and mesoplastic in the vermicomposting process: response of Eisenia fetida and quality of the vermicomposts obtained. Environ Pollut 333:122027. https://doi.org/ 10.1016/j.envpol.2023.122027
- Edwards CA, Arancon NQ (2022) Biology and ecology of earthworms. Springer US, New York NY
- Gómez-Brandón M, Lores M, Domínguez J (2023) Recycling and valorization of distilled grape marc through vermicomposting: a pilot-scale study. J Mater Cycles Waste Manag 25:1509–1518. https://doi.org/10.1007/s10163-023-01627-6
- Sáez JA, Pérez-Murcia MD, Vico A et al (2021) Olive mill wastewater-evaporation ponds long term stored: integrated assessment of in situ bioremediation strategies based on composting and vermicomposting. J Hazard Mater 402:123481. https://doi.org/10. 1016/j.jhazmat.2020.123481
- Kumar A, Mohd M, Dixit J (2023) Smart vermicomposting bin for rapid transformation of Dal lake aquatic weed into fortified vermicompost. Int J Recycl Org Waste Agric. https://doi.org/10. 30486/ijrowa.2022.1955835.1445
- González-Moreno MÁ, García Gracianteparaluceta B, Marcelino Sádaba S et al (2022) Vermicomposting of lavender waste: a biological laboratory investigation. Agronomy 12:2957. https://doi. org/10.3390/agronomy12122957
- Quintela-Sabarís C, Mendes LA, Domínguez J (2022) Vermicomposting as a sustainable option for managing biomass of the invasive tree acacia dealbata link. Sustainability 14:13828. https://doi. org/10.3390/su142113828
- De Medina-Salas L, Giraldi-Díaz MR, Castillo-González E, Morales-Mendoza LE (2020) Valorization of orange peel waste using precomposting and vermicomposting processes. Sustainability 12:7626. https://doi.org/10.3390/su12187626
- González-Moreno MA, García Gracianteparaluceta B, Marcelino Sádaba S et al (2020) Feasibility of vermicomposting of spent coffee grounds and silverskin from coffee industries: a laboratory study. Agronomy 10:1125. https://doi.org/10.3390/agronomy10 081125
- 15. Hu X, Zhang T, Tian G et al (2021) Pilot-scale vermicomposting of sewage sludge mixed with mature vermicompost using

earthworm reactor of frame composite structure. Sci Total Environ 767:144217. https://doi.org/10.1016/j.scitotenv.2020.144217

- Sharma K, Garg VK (2018) Comparative analysis of vermicompost quality produced from rice straw and paper waste employing earthworm Eisenia fetida (Sav.). Bioresour Technol 250:708–715. https://doi.org/10.1016/j.biortech.2017.11.101
- Machado JC, Faria MA, Ferreira IMPLVO (2019) Hops: new perspectives for an old beer ingredient. In: Grumezescu AM, Holban AM (eds) Natural beverages. The science of beverages, vol. 13, pp 267–301. Academic Press, ISBN 9780128166895. https://doi. org/10.1016/B978-0-12-816689-5.00010-9
- Agriculture and Rural Development. European Commission Hops. https://agriculture.ec.europa.eu/farming/crop-productions-andplant-based-products/hops_en. Accessed 12 Dec 2022
- Kopeć M, Mierzwa-Hersztek M, Gondek K et al (2020) Biological activity of composts obtained from hop waste generated during the brewing. Biomass Convers Biorefinery. https://doi.org/10. 1007/s13399-020-00746-6
- Butt KR (1993) Utilisation of solid paper-mill sludge and spent brewery yeast as a feed for soil-dwelling earthworms. Bioresour Technol 44:105–107. https://doi.org/10.1016/0960-8524(93) 90182-B
- Saba S, Zara G, Bianco A et al (2019) Comparative analysis of vermicompost quality produced from brewers' spent grain and cow manure by the red earthworm Eisenia fetida. Bioresour Technol 293:122019. https://doi.org/10.1016/j.biortech.2019.122019
- Afonso S, Arrobas M, Pereira EL, Rodrigues MÂ (2021) Recycling nutrient-rich hop leaves by composting with wheat straw and farmyard manure in suitable mixtures. J Environ Manage 284:112105. https://doi.org/10.1016/j.jenvman.2021.112105
- Luskar L, Polanšek J, Hladnik A, Čeh B (2022) On-farm composting of hop plant green waste—chemical and biological value of compost. Appl Sci 12:4190. https://doi.org/10.3390/app12094190
- 24. Bouché MB (1972) Lombriciens de France: écologie et systématique. Ann Zool Ecol Anim 72:671
- Elvira C, Dominguez J, Briones MJI (1996) Growth and reproduction of Eisenia andrei and E. fetida (Oligochaeta, Lumbricidae) in different organic residues. Pedobiologia 40:377–384
- González-Moreno, MÁ, García-Gracianteparaluceta, B, Espuelas-Zuazu, S, et al (2020) A small-scale study of optimization of vermicomposting of coffee industry wastes focused on growth rate. In: 5th International Online Conference on Reuse and Recycling of Materials (ICRM 2020). Kottayam, Kerala India
- Bhat SA, Singh J, Vig AP (2015) Potential utilization of bagasse as feed material for earthworm Eisenia fetida and production of vermicompost. Springerplus 4:1–9. https://doi.org/10.1186/ s40064-014-0780-y
- Esmaeili A, Khoram MR, Gholami M, Eslami H (2020) Pistachio waste management using combined composting-vermicomposting technique: physico-chemical changes and worm growth analysis. J Clean Prod 242:118523. https://doi.org/10.1016/j.jclepro.2019. 118523
- Singh D, Suthar S (2012) Vermicomposting of herbal pharmaceutical industry waste: earthworm growth, plant-available nutrient and microbial quality of end materials. Bioresour Technol 112:179–185. https://doi.org/10.1016/j.biortech.2012.02.101
- Yadav A, Garg VK (2011) Vermicomposting-an effective tool for the management of invasive weed parthenium hysterophorus. Bioresour Technol 102:5891–5895. https://doi.org/10.1016/j.biort ech.2011.02.062
- Parthasarathi K, Balamurugan M, Prashija KV et al (2016) Potential of Perionyx excavatus (Perrier) in lignocellulosic solid waste management and quality vermifertilizer production for soil health. Int J Recycl Org Waste Agric 5:65–86. https://doi.org/10.1007/ s40093-016-0118-6

- 32. Karmegam N, Vijayan P, Prakash M, John Paul JA (2019) Vermicomposting of paper industry sludge with cowdung and green manure plants using Eisenia fetida: a viable option for cleaner and enriched vermicompost production. J Clean Prod 228:718–728. https://doi.org/10.1016/j.jclepro.2019.04.313
- Coulibaly SS, Bi IAZ (2010) Influence of animal wastes on growth and reproduction of the African earthworm species Eudrilus eugeniae (Oligochaeta). Eur J Soil Biol J 46:225–229. https:// doi.org/10.1016/j.ejsobi.2010.03.004
- Garg VK, Suthar S, Yadav A (2012) Management of food industry waste employing vermicomposting technology. Bioresour Technol 126:437–443. https://doi.org/10.1016/j.biortech.2011.11.116
- Yadav A, Suthar S, Garg VK (2015) Dynamics of microbiological parameters, enzymatic activities and worm biomass production during vermicomposting of effluent treatment plant sludge of bakery industry. Environ Sci Pollut Res 22:14702–14709. https:// doi.org/10.1007/s11356-015-4672-7
- Suthar S (2007) Production of vermifertilizer from guar gum industrial wastes by using composting earthworm Perionyx sansibaricus (Perrier). Environmentalist 27:329–335. https://doi.org/ 10.1007/s10669-007-9032-9
- Balachandar R, Baskaran L, Yuvaraj A et al (2020) Enriched pressmud vermicompost production with green manure plants using Eudrilus eugeniae. Bioresour Technol 299:122578. https:// doi.org/10.1016/j.biortech.2019.122578
- Negi R, Suthar S (2018) Degradation of paper mill wastewater sludge and cow dung by brown-rot fungi Oligoporus placenta and earthworm (Eisenia fetida) during vermicomposting. J Clean Prod 201:842–852. https://doi.org/10.1016/j.jclepro.2018.08.068
- Domínguez J, Edwards CA, Webster M (2000) Vermicomposting of sewage sludge: effect of bulking materials on the growth and reproduction of the earthworm Eisenia andrei. Pedobiologia 44:24–32. https://doi.org/10.1078/S0031-4056(04)70025-6
- Gupta R, Garg VK (2008) Stabilization of primary sewage sludge during vermicomposting. J Hazard Mater 153:1023–1030. https:// doi.org/10.1016/j.jhazmat.2007.09.055
- 41. Malińska K, Zabochnicka-Światek M, Cáceres R, Marfà O (2016) The effect of precomposted sewage sludge mixture amended with biochar on the growth and reproduction of Eisenia fetida during laboratory vermicomposting. Ecol Eng 90:35–41. https://doi.org/ 10.1016/j.ecoleng.2016.01.042
- Suthar S (2008) Microbial and decomposition efficiencies of monoculture and polyculture vermireactors, based on epigeic and anecic earthworms. World J Microbiol Biotechnol 24:1471–1479. https://doi.org/10.1007/s11274-007-9635-9
- Suthar S (2007) Nutrient changes and biodynamics of epigeic earthworm Perionyx excavatus (Perrier) during recycling of some agriculture wastes. Bioresour Technol 98:1608–1614. https://doi. org/10.1016/j.biortech.2006.06.001
- 44. Li W, Bhat SA, Li J et al (2020) Effect of excess activated sludge on vermicomposting of fruit and vegetable waste by using novel vermireactor. Bioresour Technol 302:122816. https://doi.org/10. 1016/j.biortech.2020.122816
- Gómez-Brandón M, Lazcano C, Lores M, Domínguez J (2011) Short-term stabilization of grape marc through earthworms. J Hazard Mater 187:291–295. https://doi.org/10.1016/j.jhazmat. 2011.01.011
- 46. Gunadi B, Edwards CA (2003) The effects of multiple applications of different organic wastes on the growth, fecundity and survival of Eisenia fetida (Savigny) (Lumbricidae). Pedobiologia 47:321–329. https://doi.org/10.1078/0031-4056-00196
- Suthar S, Mutiyar PK, Singh S (2012) Vermicomposting of milk processing industry sludge spiked with plant wastes. Bioresour Technol 116:214–219. https://doi.org/10.1016/j.biortech.2012.03. 101

- Suthar S, Singh S (2008) Feasibility of vermicomposting in biostabilization of sludge from a distillery industry. Sci Total Environ 394:237–243. https://doi.org/10.1016/j.scitotenv.2008.02.005
- Edwards CA, Dominguez J, Neuhauser EF (1998) Growth and reproduction of Perionyx excavatus (Perr.) (Megascolecidae) as factors in organic waste management. Biol Fertil Soils 27:155– 161. https://doi.org/10.1007/s003740050414
- Haimi J (1990) Growth and reproduction of the compost-living earthworms Eisenia andrei and E. fetida. Rev DÉcologie Biol Sol 27:415–421
- Cluzeau D, Fayolle L, Hubert M (1992) The adaptation value of reproductive strategy and mode in three epigeous earthworm species. Soil Biol Biochem 24:1309–1315. https://doi.org/10.1016/ 0038-0717(92)90110-J
- Elvira C, Domínguez J, Mato S (1997) The growth and reproduction of Lumbricus rubellus and Dendrobaena rubida in cow manure mixed cultures with Eisenia andrei. Appl Soil Ecol 5:97– 103. https://doi.org/10.1016/S0929-1393(96)00120-5
- Gong X, Li S, Carson MA et al (2019) Spent mushroom substrate and cattle manure amendments enhance the transformation of garden waste into vermicomposts using the earthworm Eisenia fetida. J Environ Manage 248:109263. https://doi.org/10.1016/j.jenvman. 2019.109263
- Varma VS, Kalamdhad AS, Khwairkpam M (2016) Feasibility of Eudrilus eugeniae and Perionyx excavatus in vermicomposting of water hyacinth. Ecol Eng 94:127–135. https://doi.org/10.1016/j. ecoleng.2016.05.058
- Smetak KM, Johnson-Maynard JL, Lloyd JE (2007) Earthworm population density and diversity in different-aged urban systems. Appl Soil Ecol 37:161–168. https://doi.org/10.1016/j.apsoil.2007. 06.004
- Nogales R, Thompson R, Calmet A et al (1998) Feasibility of vermicomposting residues from olive oil production obtained using two stage centrifugation. J Environ Sci Health Part A 33:1491– 1506. https://doi.org/10.1080/10934529809376800
- 57. Elvira C, Sampedro L, Nogales R (1999) Suitability of sludges from dairy and paper industries for growth. Pedobiologia 43:766–770

- Neuhauser EF, Hartenstein R, Kaplan DL (1980) Growth of the earthworm Eisenia foetida in relation to population density and food rationing. Oikos. https://doi.org/10.2307/3544730
- Mikunthan G, Piratheban S (2011) Performance of epigeic earthworm species in different solid wastes for compost making. Proc 22nd Int Conf Waste Technol Manag 195–205
- Frederickson J, Butt KR, Morris RM, Catherine D (1997) Combining vermicultura with traditional green waste composting systems. Soil BiolBiochem 29:725–730
- Kaur A, Singh J, Vig AP et al (2010) Cocomposting with and without Eisenia fetida for conversion of toxic paper mill sludge to a soil conditioner. Bioresour Technol 101:8192–8198. https:// doi.org/10.1016/j.biortech.2010.05.041
- Chauhan HK, Singh K (2013) Effect of tertiary combinations of animal dung with agrowastes on the growth and development of earthworm Eisenia fetida during organic waste management. Int J Recycl Org Waste Agric 2:1–7. https://doi.org/10.1186/ 2251-7715-2-11
- 63. Garg VK, Yadav YK, Sheoran A et al (2006) Livestock excreta management through vermicomposting using an epigeic earthworm Eisenia foetida. Environmentalist 26:269–276. https://doi. org/10.1007/s10669-006-8641-z
- Gupta R, Mutiyar PK, Rawat NK et al (2007) Development of a water hyacinth based vermireactor using an epigeic earthworm Eisenia foetida. Bioresour Technol 98:2605–2610. https://doi.org/ 10.1016/j.biortech.2006.09.007
- Ečimović S, Velki M, Mikuška A et al (2022) How the composition of substrates for seedling production affects earthworm behavior. Agriculture 12:2128. https://doi.org/10.3390/agricultur e12122128
- Rusănescu CO, Rusănescu M, Voicu G et al (2022) The recovery of vermicompost sewage sludge in agriculture. Agronomy 12:2653. https://doi.org/10.3390/agronomy12112653

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