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Bioconversion of fruit and vegetable waste into earthworms as a new protein source: the environmental impact of earthworm meal production

Doriana E. A. Tedesco^{1*}, Cecilia Conti¹, Daniela Lovarelli², Elisa Biazzi ³, Jacopo Bacenetti^{1*}

¹Department of Environmental and Policy Science. Università degli Studi di Milano, Via G. Celoria 2, 20133 Milan, Italy.

²Department of Agricultural and Environmental Science. Università degli Studi di Milano, Via G. Celoria 2, 20133 Milan, Italy.

³Council for Agricultural Research and Economics – Zootechnics and Aquaculture Research Center, CREA-ZA, viale Piacenza 29, 26900 Lodi, Italy.

* Corresponding author: doriana.tedesco@unimi.it; Jacopo.bacenetti@unimi.it

Abstract

Food waste is recognized as a global issue affecting the sustainability of the food supply chain. The unnecessary exploitation of natural resources (land, water and fossil energy) and production of greenhouse gas emissions (GHG) make the reduction of food waste a key point. In this context, the use of fruit and vegetable waste (FVW) as growth substrate for fresh earthworms to produce dried meal for feed and food purpose can be recognized as a viable solution.

Therefore, the aim of this study is to evaluate the environmental impact of the bioconversion of FVW into earthworm meal to be used as new food/feed source. This is carried out by adopting the Life Cycle Assessment (LCA) method with an attributional

approach and solving the multifunctionality of the system with an economic allocation between earthworms and vermicompost.

The results show that the main process hotspots are the emissions of methane, dinitrogen monoxide and ammonia taking place during vermicomposting, as well as FVW transport and electricity consumed during fresh earthworm processing. Respect to the one used as feed, the dried meal with food purpose shows a higher impact due to the higher economic value and to the higher electricity consumed during freeze drying compared to the oven-drying process for feed meal production. Enhancing productivity and reducing energy consumption are necessary to improve the sustainability of earthworm meal as food/feed source.

Keywords: Life Cycle Assessment, Circular economy, Fruit Vegetable Waste, Earthworm, Novel food/feed protein, Sustainability

1. Introduction

Food waste is already recognized as an important global issue affecting the sustainability of the food supply chain (Tonini et al., 2018). The losses that occur during the whole lifecycle in terms of food scraps and wasted food in both the agricultural/industrial and domestic phases, can account for up to 60% of the initial weight of the food products (Notarnicola et al., 2017). According to the Food and Agriculture Organization of the United Nations (FAO) about 1.3 billion tons of food produced worldwide every year are wasted along the supply chain (FAO, 2011).

The problem of food waste involves significant environmental, economic and social impacts. Food waste leads to an unnecessary exploitation of natural resources (land, water and fossil energy) and to notable greenhouse gas (GHG) emissions (Pham et al., 2015). Food produced but lost along the chain and wasted accounts for around one quarter of total freshwater resources and of total global cropland area used and lost, and has embedded production-phase emissions that should be counted (Kummu et al., 2012; Porter et al., 2016). The median GWP (Global Warming Potential) value for different food categories were estimated to account 0.37 kg CO₂-eq/kg for field-grown vegetables and 0.42 kg CO₂-eq/kg for field-grown fruit with meat from ruminants having the highest impact 26.61 kg CO₂-eq/kg bone free meat (BFM) (Clune et al., 2016).

Fruit and vegetable waste (FVW) is one of the major categories in the food waste generated, especially in industrialized regions (FAO, 2011; Porter et al. 2016). In EU contribute to almost 50% of the food waste generated by households (De Laurentiis et al., 2018). Moreover, FVW management poses disposal and environmental problems, due to its high biodegradability but also a great potential for reuse, recycling and energy recovery (Plazzotta et al., 2017). On the other hand, the patterns of growth in demand for animal-source foods (Alexandratos and Bruinsma, 2012; United Nations, 2017), poses terrestrial invertebrates as a suitable candidate to supplement other animal-based food proteins (FAO, 2013). A possible strategy is the utilization of FVW as feeding substrate for the rearing of terrestrial invertebrates to be used as potential

protein source for feed and/or food supply chains. Terrestrial invertebrates represent a potential valuable solution to two problems: 1) the increasing amount of food waste, which can cause environmental pollution if not properly managed, 2) the rising global demand for food and feed, necessary to supply human and animal nutrition (Salomone et al., 2017).

Up to now, the attention on alternative protein sources has regarded mainly terrestrial invertebrates from insects both as human food (Oonincx and De Boer, 2012; Halloran et al., 2016) and as animal feed (Smetana et al., 2016; Salomone et al., 2017; Tallentire et al., 2018; Thévenhot et al., 2018). Among others terrestrial invertebrates, earthworms could be an interesting solution to be evaluated. In nature, earthworms grow on a wide variety of organic materials (Edwards, 1988) and are even called "ecosystem engineers" (Jones et al., 1994), as they are the main drivers of the decomposition of organic waste in soil ecosystems (Lim et al., 2016).

Earthworms provide an excellent ecosystem service modifying the physicochemical properties of soil (Singh et al., 2016) maintaining aerobic conditions (Nigussie et al., 2016). They are voracious eaters and biodegraders of waste (Sims and Gerard, 1985; Sinha et al., 2008). Therefore, they can be fed on different types of waste, such as FVW (Chatterjee et al., 2014; Huang et al., 2016; Huang and Xia, 2018). Furthermore, vermicomposting is more effective in FVW disposal techniques to reduce GHG emissions and nitrogen losses than traditional composting (Yang et al. 2017; Swati and Hait, 2018; Colón et al. 2012). From an environmental point of view, vermicomposting process emissions of NH_3 , CH_4 and N_2O are lower by three orders of magnitude than those coming from composting process (Lleò et al., 2013). Moreover, from the vermicomposting process, the derivative vermicompost is an excellent high quality bioactive amendment to improve soil fertility. It is pathogen-free thanks to earthworm gut transit mechanism which classifies vermicomposting as a promising sanitation technique in comparison to composting processes (Soobhany et al., 2017; Yang et al., 2017). Vermicomposting represents an option of FVW valorization, because it is a lowcost biotechnology that turns waste into a high-quality residue, namely vermicompost,

through the joint action of earthworms and microorganisms (Dominguez, 2004; Yang et al., 2017). Currently, earthworms are employed to deal with food waste management in a bioconversion process to mitigate the food waste problem as a sustainable, cost-effective and ecological approach (Fernández-Gómez et al., 2010; Singh et al., 2011; Yadav and Garg et al., 2011; Huang et al., 2016; Lim et al., 2016). Nonetheless, earthworms grown on FVW can contribute to the waste disposal efficiency biotransforming FVW into two valuable products: (i) vermicompost, that can be sold as organic fertilizer, and (ii) earthworms themselves that can be a new food/feed source, thanks to their high protein content.

Like insects, earthworms are rich in proteins, particularly in essential amino acids (Cayot et al., 2009; Zhenjun and Jiang, 2017) and they can contribute to human and animal nutrition (Ncobela and Chimonyo, 2015; Bahadori et al., 2017; Zhenjun and Jiang, 2017). By the way, the production of edible terrestrial invertebrates as food or feed, have to be safe and wholesome. To ensure a high level of protection of human and animal health, terrestrial invertebrates and therefore earthworms need to be considered as "farmed animals" when they represent a food source and fed only with safe feed used as growth substrate. EU framework established restrictions or prohibitions on the feed for farm animals reared for producing food or feed (e.g. prohibited feeding: catering waste or household waste Regulation (EU) No 1069/2009 Regarding animal by-products and Regulation (EC) No 767/2009 on the placing on the market and use of feed, Annex III). Besides, safety rules for food or feed purpose were defined in the Hygiene Package (e.g. Regulation No 852/2004 and Regulation No 853/2004 on the hygiene of foodstuffs; Regulation No 183/2005 laying down requirements for feed hygiene) and on the levels of contaminants (Directive 2002/32/EC on undesirable substances in animal feed and Regulation (EC) No 1881/2006, setting maximum levels for certain contaminants in foodstuffs), according to which earthworm rearing and market as food or feed must respect the legislation in force for farmed animals and for the derivative products.

The aim is to evaluate the environmental impact of the earthworms' meal production for feed and food purpose obtained from earthworms reared on fruit and vegetable waste (FVW) discarded directly from juice and ready-to-eat processing industries. The propensity and willingness towards earthworms as a future food source has already been investigated in a previous study (Conti et al. 2018). In this study the environmental performances of earthworm meal production adopting Life Cycle Assessment (LCA) was analysed, in order to:

- evaluate the real effectiveness and sustainability to produce earthworms as food/feed protein source,
- test the environmental impact of the production system.

Two different scenarios (FEED and FOOD) were evaluated and a sensitivity analysis concerning key parameters, assumptions and methodological choices was performed.

2. Materials and methods

To evaluate the environmental impact of the earthworms' production system, the Life Cycle Assessment (LCA) method was used. LCA is a holistic approach, structured and recognized worldwide that consists of a systematic set of procedures to convert inputs and outputs of the studied system into its related environmental impact. To perform LCA, the ISO standard 14040/44 methodology (ISO, 2006) must be adopted. In this study, the methodological framework of the attributional approach is used. It permits to model the production process of earthworms (*Eisenia fetida*) without considering potential effects on the market due to the use of vegetable waste and to the introduction of protein from earthworm's origin.

According to ISO 14040/44 (ISO, 2006), LCA involves four distinct and interdependent phases, all of which are discussed in detail in the next sections. These phases consist of defining and analysing:

- i) goal and scope, which include the selection of a functional unit and definition of system boundary;
- life cycle inventory, which involves the definition of energy and material flows between the system and the environment and through the different subsystems and operations in the evaluated system;
- iii) impact assessment, during which the inventory data are converted in environmental indicators (i.e. environmental impact categories); and
- iv) discussion and interpretation of the results, where the results from the inventory analysis and impact assessment are summarized, sensitivity and uncertainty analysis are carried out and recommendations are drawn.

2.1 Goal and scope definition

Eisenia fetida is a widespread epigeic species of earthworms (Bouché, 1977; Sims and Gerard, 1985) that it is characterized by a high tolerance to a wide range of environmental factors, higher rates of consumption, digestion and assimilation of organic substances and high reproductive rate (Bhat et al., 2018; Domínguez and Edwards, 2010a). Adult earthworms of Eisenia fetida weigh up to 0.55 g (Domínguez and Edwards, 2010a) and reach up to 60-120 mm in length and 3-6 mm in diameter (Sims and Gerard, 1985). They are hermaphrodites (Edwards and Bohlen, 1996), but usually reproduction occurs through copulation and cross-fertilization, after which each of the mated individuals can produce cocoons (Domínguez and Edwards, 2010a). Approximately, the time from newly-laid cocoon through clitellate adult earthworm ranges from 45 to 51 days (Dominguez Edwards, 2010a). Optimum growth conditions include a range of temperature between 25-30 °C, moisture 75-90% (Edwards, 1988) and pH >5 and <9, optimum centered around 7.0 (Kaplan et al., 1980). Given the optimum conditions of temperature and moisture, about 5 kg of worms can vermiprocess 1 ton of waste into vermi-compost in just 30 days (Sinha et al., 2010).

The scope is to investigate the environmental profile of the bioconversion process of FVW into earthworm dried meal as a novel food and feed protein source. The

environmental impact analysis was carried out studying a small-scale production plant of earthworms housed in Northern Italy with LCA approach.

Currently, very few plants are present in Italy, and all of them are small-scaled or lab-scaled. Moreover, also the production processes are quite standardized.

The research questions for this study are as follows:

- What is the magnitude of the environmental impact of the production of earthworms' meal?
- What are the environmental hotspots for the evaluated process?

The outcomes of this study will be useful specially to decision makers, being the first results of a full environmental impact assessment about earthworms' protein production using FVW feed.

2.2 Functional unit

The selection of the functional unit (FU) is crucial to allow fair comparison with other studies and adequate assessments. According to ISO 14040 (ISO 14040, 2006), the FU is defined as the quantified performance of a product system and is used as a reference unit in an LCA.

In the studied context, very few studies were found in literature. Given the function of the earthworm process for novel food/feed protein production, the selected FU in this study was 1 kg of dried meal of earthworm.

2.3 Description of the production process of earthworms meal

The environmental impact analysis was carried out studying a small-scale production system of earthworms located in the province of Lecco (North Italy) (45°55'23" N and 9°19'34" E). The production process of earthworm meal was divided in two subsystems (SS):

- SS1, characterized by the production of fresh earthworms and vermicompost,
- SS2, in which fresh earthworms are used for the meal production.

2.3.1 Subsystem 1: Fresh earthworms and vermicompost production

The present study was approved by the Animal Ethics Committee of Milan University of Study (30.01.17; ethical code number 02/17).

Fresh earthworms and vermicompost were produced on a rearing area of 34 m² made up of a non-woven textile sheet used to avoid water stagnation and earthworms' escape. The area was covered with a net to avoid predators' damage. A mix of young-non-clitellum and adult-clitellate earthworms was provided by the earthworms' producer, added at an initial density of 1 kg/m² and reared on a feeding substrate consisting of FVW.

FVW was provided by a fruit and vegetable producer of ready-to-eat products and had a variable composition dependent on seasonality in vegetable growth and work process. The waste consisted mainly of tropical fruits, such as pineapple, papaya, mango, kiwi as well as of melon, tomatoes and grapes. The fiber components of FVW (e.g., pineapple tufts) were grinded with a gardening shredder to make them biodegradable by earthworms' activity. To reach a C:N ratio optimal for earthworm growth the FVW was mixed with straw (10:1). Fruit and vegetable wastes were left to rot for a few days before being fed to earthworms, which allowed having a narrow range of favorable chemical and environmental conditions more favorable for microbial activity and further decomposition of the growth substrate (Dominguez and Edwards, 2010b). FVW were added to feed earthworms three times a month, by introducing them on the top of the production area. In order to guarantee optimum growth conditions, moisture, temperature and pH of the growth substrate were monitored and kept under control and supplied with water if needed (Moisture 84-88%; Temperature 20-25°C; pH 6.07-8.02; C/N 25.34). These values are in the range of the recommended values of process factors for vermicomposting (Dominguez and Edwards, 2010b).

Earthworms were reared for three months in the most favorable environmental conditions in order to obtain the best conversion efficiency. After 3 months, they were separated mechanically from the vermicompost with the use of a trommel.

Besides earthworms, during the decomposition of FVW also an odor-free and humus-like substance is produced (Suthar, 2009). Vermicomposting is the stabilization of organic material through the joint action of earthworms and microorganisms (Dominguez, 2004) and its final product is the vermicompost. In our study vermicompost represents the co-product of the production system, with possible beneficiations as a valuable replacer of conventional soil fertilizers. Specifically, it is a residue produced by earthworms, characterized by low C/N ratio, high porosity, water-holding capacity and available nutrients (Lim et al., 2015).

2.3.2 Subsystem 2 – From fresh earthworms to food/feed meal

Once collected, earthworms were repeatedly washed with running tap water to clean the body surface and kept in water until their digestive system were clean. Finally, washing water was removed and earthworms were packaged in plastic bags and stored at - 28° C to let them enter quiescence and kill them.

For the production of dry meal, two technological transformation processes were considered depending on the final destination of the meal: food scenario and feed scenario.

For food scenario the vacuum freeze drying technology method was choose for producing high quality dehydrated earthworm meal because this is a typical technology for food purposes in order to avoid affecting the nutritional characteristics. Earthworms were freeze-dried at a pilot scale level. Finally, freeze-dried earthworms were ground to obtain the meal.

For feed scenario, the dry meal was produced in laboratory by drying earthworms in an oven at 65°C to a constant weight and grinding.

2.3 System boundary definition

A "from cradle to gate" system boundary was considered. More in details, the life cycle of each sub process for both subsystems (SS1 and SS2) was considered. Consequently, the following activities were included: raw materials extraction (e.g.,

fossil fuels, metals and minerals), inputs manufacture (e.g., diesel fuel, electricity, tap water and trucks for FVW transport), inputs use (diesel fuel emissions), maintenance and final disposal of capital goods (e.g., the trucks used for the FVW transport). The emissions into atmosphere (e.g., dinitrogen monoxides, methane, etc.) related to vermicomposting of FVW were also included.

Packaging, distribution, use and end-of-life of the produced meal were excluded from the system boundary.

Figure 1 summarizes the system boundary considered.

Figure 1 - Around Here

2.4 Life Cycle Inventory (LCI)

Inventory data relevant to the production of earthworms' biomass were collected over a three-month experimental test performed in year 2017.

Primary data were collected with questionnaires during interviews with the farmer. These regarded mainly the amount of FVW used as feed, transport, diesel, fossil energy for preparing the feed substrate, water volumes and land occupation for earthworms breeding and water for washing earthworms. Secondary data about electricity for processing earthworms into dried meal and fossil fuel for transport activities were obtained from Ecoinvent database (Weidema et al., 2013).

The main inventory data collected during the experimental trials are reported in **Table 1** for SS1 and in **Table 2** and **Table 3** for the two scenarios in SS2.

Table 1, 2 & 3 – Around here

Vermicomposting is a process that inevitably involves emissions of greenhouse gases (Lleó et al., 2013; Wang et al., 2014; Nigussie et al., 2016; Swati and Hait, 2018), although vermicomposting process emissions are clearly lower than those coming from composting (Colón et al., 2012; Lleó et al., 2013; Nigussie et al., 2016; Yang et al., 2017).

Various factors such initial waste characteristics, process parameters like aeration, moisture content, temperature regime, contribute to fully understand the influence of process parameters in gas emissions during vermicomposting (Swati and Hait, 2018). However, the effects of earthworms on gas emissions are complicated and no consensus has been reached yet (Wang et al, 2014). Notwithstanding all this, an estimation of the gaseous emissions of CH₄, N₂O and NH₃ during SS1 was based on the relationship of growth substrate quality parameters with the gaseous emission as reported by Yang et al. (2017): consequently, the gaseous emissions per kg of fresh earthworm, were assumed equal to 5,056 g CH₄, 1,53 g N₂O and 15,84 g NH₃.

2.5 Allocation

Considering that the production system entails the production of different products, allocation should be dealt with. In some cases, among which this study, the choice of the allocation procedure may be difficult and questionable. However, the present rearing system produces as main product an earthworm biomass growing on a mix of FVW, which is further processed into earthworm meal that leaves the holding. Vermicompost is produced alongside as co-product. Since these two outputs are produced in very different amounts, the sharing of the environmental impact was performed with an economic allocation in order to avoid attributing an unbalanced impact, although commonly the mass allocation is suggested. Thus, economic allocation was selected and based on the estimation of earthworm meal and vermicompost prices as reported in Table 4. In detail, at the time being, no reference prices for food and feed do exist. Therefore, the earthworm meal price for both food and feed was estimated considering the economic sustainability to be achieved by this new food/feed sources in comparison with other animal protein sources currently used. For the feed dried meal, it was estimated 1.1 €/kg dry matter. As reference, the prices of fishmeal (1.46 €/kg of dry matter) (Milan Grain Association, 2018) and of insect dried meal (1.09 €/kg of dry matter) (as proposed by Salomone et al., 2017) were considered. Concerning earthworm food meal, to enable comparison with other animal food

products, meat prices were recalculated to dry matter content. The estimated 15 €/kg of dry matter (22.4 €/kg protein) for food earthworm meal is related to the comparison with animal food products prices such as pork (14-16 €/kg of dry matter, 19 -19.5 €/kg protein), poultry (17-18,3 €/kg of dry matter, 20,4 – 22.0 €/kg protein) and beef (26,6-28,6 €/kg of dry matter, 33,6-37.6 €/kg protein) (Borsa Merci Modena, 2018; ISMEA,2018). The economic value for vermicompost was considered 0.30 €/kg (CONITALO, 2018).

Table 4 around here

Because allocation is a key methodological choice and is here based on the estimate of prices subject to variability, sensitivity analysis was performed on this issue (± 30%).

2.6 Life Cycle Impact Assessment (LCIA)

The LCIA consists in transforming inventory data into environmental indicators. This step is achieved by using defined characterization factors that are gathered from characterization methods. Among the available ones, ILCD (International Reference Life Cycle Data System) midpoint characterization method (ILCD, 2012) is endorsed by the European Commission and adopted in this study. According to ILCD, the following impact categories were evaluated: Climate change midpoint (IPPC,2007), Ozone depletion, midpoint (WMO,1999), Human toxicity midpoint, cancer effects and non cancer effects USEtox (Rosenbaum et al 2008), Ecotoxicity freshwater, midpoint USEtox (Rosenbaum et al 2008), Particulate matters, midpoint RiskPoll model (Rabl and Spadaro, 2004 and Greco et al 2007), Photochemical ozone formation, midpoint (Van Zelm et al 2008 as applied in ReCiPe2008), Acidification, midpoint (Seppala et al 2006, Posch et al. 2008), Eutrophication terrestrial, midpoint (Seppala et al 2006, Posch et al 2008), Eutrophication aquatic freshwater/marine, midpoint (ReCiPe2008, EUTREND model - Struijs et al 2009), Resource depletion – mineral and fossil fuels, midpoint (CML 2002, Guinée et al 2002).

The choice of considering these indicators was related to the need of providing a comprehensive evaluation of the environmental impact of *Eisenia fetida* meal production as food and feed supplements.

3 Results

3.1 Fresh earthworm production

Figure 2 shows the environmental hotspots for SS1 (earthworm production). Except for CC, PM, TA and TE, for the other 7 evaluated impact categories, transport of fruit and vegetable waste (FVW) from the food industry to the vermicomposting plant represents the largest contributor to the environmental impact. More in details, the contribution of transport ranges from 1.9% in TA and TE to 95% in MFRD and it is larger than 75% for 6 of the 12 evaluated environmental effects. The emissions during the FVW vermicomposting are the main contributors for CC (78%) due to the emissions of dinitrogen monoxide and methane, PM (94%), TA (97%), TE (98%) and ME (78%) due to the emissions of ammonia. Methane emissions are responsible also of about 4% of POF. Diesel consumption during the partial chopping of FVW is responsible for a share of the environmental impact lower than 5% for all the assessed impact categories, except for POF (17%, mainly due to refinery activities). The impact related to water consumption is little (< 10%) except for FE (37%, due to the emission of phosphate in water) and FEX (12%).

Table 5 reports the absolute impact for producing 1 kg of fresh earthworms in the two scenarios (i.e. FEED and FOOD). The differences between the two scenarios are related to the different allocation factors (see **Table 4**).

Table 5 - Around here

3.2 Earthworms' meal production

Figure 3 shows the comparison between the earthworm meal produced in the two scenarios (FEED and FOOD) as well as the related environmental hotspots. The absolute impact for producing 1 kg of earthworm meal in the two scenarios is reported in **Table 6**. The earthworm meal produced in the FOOD scenario shows an impact higher than the one produced in the FEED scenario. This difference ranges from 2.06 times more for FE to 3.58 times more for TE and is related to the different allocation between meal and vermicompost and, secondarily, to the higher electricity consumption in SS2 of FOOD scenario (where the fresh earthworms are freeze-dried instead of being only dried as in FEED scenario).

Table 6 - Around here

The production of fresh earthworms (SS1) is responsible of more than 75% of the environmental impact for the following impact categories PM, TA, TE and ME in both the scenarios and in MFRD (in FOOD scenario) and of 46% and 62% of CC in FEED and FOOD scenario, respectively. For CC, the impact related to the fresh earthworm production is mainly due to the emissions of CO₂, CH₄ and N₂O that occur during earthworms' rearing. For all the other environmental impact categories (OD, HT-noc, HT-c, FE, FEx, and, even if only for the FEED scenario, also in CC and POF), the SS2 is the main responsible of the environmental impact of the dried meal, with a share of the impact ranging from 54% (CC) to 88% (FE).

Figure 3 – Around here

3.3 Sensitivity analysis

A sensitivity analysis was carried out to investigate the effect of key parameters, assumptions and methodological choices of the study as well as to test the robustness of the achieved environmental results. Thus, the following aspects were considered:

- the electricity consumption during SS2. A variation ± 25% was taken into account for the electricity consumed during drying in the FEED scenario and during freeze-drying in the FOOD scenario.
- The substitution of the Italian electric mix with to renewable electricity produced from a photovoltaic plant.
- the price of earthworm meal. The price of vermicompost kept constant, a variation ± 30% of the price of earthworm meal was considered for both scenarios. Consequently, the allocation factors for vermicompost and earthworm meal in FEED scenario became equal to 83% and 17% and to 73% and 27% at the decrease and increase in price, respectively; in the FOOD scenario, instead, allocation factors were set to 27% and 72% and to 16% and 84% at the decrease and increase in price, respectively;
- the procedure for solving the multifunctionality issue. Considering that the ISO standards suggest to avoid the allocation, instead of the economic allocation, the multifunctionality was solved taking into account a mixed functional unit composed by earthworm meal and vermicompost. More in detail, considering that 6.25 kg of fresh earthworm are needed to produce 1 kg of earthworm meal and 12.8 kg of vermicompost are produced with 1 kg of fresh earthworm the mixed FU is composed by 1 kg of earthworm meal and 80 kg of vermicompost.

The results of the sensitivity analysis are reported in **Table 7**. The change of electricity consumption in SS2 involves an impact variation ranging from -17.5% to + 21.9% in the FEED scenario and from -15.6% to +19.5% in the FOOD scenario. FE is the impact category most affected by electricity consumption, therefore it shows the highest impact variation in both scenarios. On the opposite, PM, TA, TE and ME mainly

affected by emissions occurring during fresh earthworm production, are the impact categories less affected by variations in electricity consumption during drying/freezedrying.

When the electricity is produced from a photovoltaic plant, 9 of the 12 evaluated impacts are reduced (from 1.2 to 67%) but HT-noc, FEx and MFRD increase. These two last impact categories show a remarkable impact increase mainly due to the manufacturing and disposal of the photovoltaic plant. FEx is 3.67 and 2.98 times higher for FEED and FOOD scenario, respectively while MFRD is 12.57 and 7.86 times higher for FEED and FOOD scenario, respectively.

The variation in price of the earthworm meal ($\pm 30\%$) and the consequent variation in the allocation factors involves an impact variation that ranges from $\pm 22.1\%$ for TE to $\pm 2.8\%$ for FE. More in details, when the price varies, the impact grows more for the FEED scenario respect to the FOOD one.

The choice of a mixed FU to avoid allocation, as expected, deeply affect the environmental results. Without allocation, the environmental impact is no more divided between the two products (vermicompost and dried meal) but it is fully attributed to the mixed FU. Respect to the impact of 1 kg of dried meal assessed considering economic allocation, with the mixed FU, the higher impact increase occurs for the FEED scenario where the earthworm price is lower and, consequently, also the allocation factor assessed using the earthworm meal price is low. In the FOOD scenario, where already with economic allocation, the 80% of the impact is attributed to earthworm meal, the use of a mixed FU is less impacting on the environmental indicators.

Table 7 - Around here

4. Discussion

Turning waste into a resource is part of 'closing the loop' in circular economy systems (EU COM, 2015). To this scope, earthworm play an interesting role as they are considered effective in organic waste transformation (Edwards, 1988). The earthworms process of FVW results in two excellent products: the vermicompost, a high-quality bioactive soil amendment, and the earthworms that are grown on FVW that and can be a new food/feed source, thanks to their high protein content. The ecological impacts of this process were estimated with the LCA method.

The environmental impact of the production of 1 kg of fresh earthworm (SS1) from FVW substrate and the subsequent dried process (SS2) to produce 1 kg of earthworm meal for feed purpose shows interesting outcomes; on CC the impact is 0.162 and 2.238 kg CO₂ eq, for 1 kg of fresh earthworm and 1 kg of dried meal, respectively. This is mostly related to the emissions but also to the FVW transport to the plant and to the electricity use for the drying process in SS2. To reduce the role of transport activities on the sustainability assessment, vermicomposting process could optimally take place at the FVW production site. Regarding the reduction of the energy use, a photovoltaic system or other renewable energy sources could be introduced for reducing the impact of oven-dry meal production. In any case, more research on earthworms' meal production for feed purpose using FVW could permit achieving interesting further improvements for alternative protein sources in view of reducing the environmental impact for their production and the food waste (Conti et al., 2018).

Making even more sustainable the earthworms' meal and adopting it in livestock rations would also bring to a second subsequent beneficial effect. In fact, Europe's reliance on imported protein to feed livestock, especially soybean, is inconsistent with sustainability goals. Soybean production is associated, among others, with deforestation, use of pesticides and long-distance transportation (Tallentire et al., 2018b; Thévenhot et al., 2018) which bring to an environmental burden for 1 kg of soybean meal equal to 3.05 kg CO₂ eq (Tallentire et al., 2017a). Another issue to take

into account is the total content of protein (or essential amino acids e.g. lysine) in the comparative emissions evaluation for future feed application of novel ingredients. The protein of earthworm meal ranged from 63.8-72.4% of the dry matter (Tava et al., 2018), higher than the protein of soybean meal, 43-44% of the dry matter (Feedipedia, 2019).

Considering the earthworm meal produced for food purpose, the environmental impact for 1 kg of fresh earthworm production (SS1) and the subsequent freeze-drying process (SS2) to produce 1 kg of earthworm meal still had interesting results, 0.593 and 5.944 kg CO₂ eq for fresh earthworm and dried meal, respectively. Comparing earthworm meal for food purpose with other different food categories is only partially possible because of the different functional units adopted. Commonly, LCA studies on animal food products such as pork, chicken and beef adopted as functional units the kg edible meat from carcass or kg bone free meat (BFM). Nonruminant livestock had average GWP values equal to 3.49 kg CO₂ eq/kg BFM for fish, 3.65 kg CO₂ eq/kg BFM for chicken and 5.77 kg CO₂ eq/kg BFM for pork. Ruminant livestock show the highest average GWP values in lamb (25.58 kg CO₂ eq/kg BFM) and in beef (26.61 kg CO₂ eq/kg BFM) had (Clune et al., 2017). Referring these data to 1 kg of meat protein production, the highest impact is reported with beef (75-170 kg CO₂ eq), followed by pork protein (21–53 kg CO₂ eq), and finally by chicken protein (18–36 kg CO₂ eq) (de Vries and de Boer, 2010).

Earthworm meal has a high protein content equal to 60-65% in the range of 54.6% to 71.0% dry matter (Zhenjun et al., 1997). Moreover, it is a low-demanding source of land and water use and it can be interesting in contrasting biodiversity loss. Thus, bioconverting FVW to produce earthworm meal has the potential to be considered an environmentally sustainable strategy.

5. Conclusions

Starting from a fruit and vegetable waste material, the goal was to outline innovative and sustainable models to design and develop more efficient regeneration

and re-use systems. In this study, this is achieved by valorising organic waste and transforming it into high value-added products. The turning of FVW into earthworm meal could contribute to make more sustainable the protein production and to meet the future global protein demands..

By means of LCA method, the environmental impact of the production of earthworm meal was quantified with an attributional approach. Both 1 kg of fresh earthworm and 1 kg of dried meal were evaluated. The feed substrate for earthworms is made of FVW so being highly valorized respect to wasting. Given the increasing importance worldwide of issues related to food waste, the transformation into feed and/or food meal is very promising.

Similarly, to other protein sources, earthworm meal currently has high environmental impacts mostly due to emissions during vermicomposting, transport of FVW for fresh earthworm production and energy use during processing. Freeze-drying instead of only oven-drying determines a higher environmental impact for the FOOD scenario respect to the FEED one. To make earthworm meal sustainable and competitive on the market, enhancing earthworm productivity and reducing energy consumption during processing by shifting towards renewable energy sources is essential. From a methodological point of view, the analysis highlighted that the choice made to solve the multifunctionality issue deeply affects the environmental results. In this regard, a comparison with future studies should be drawn using the same criteria for allocation or, even better, avoiding allocation by using a mixed functional unit.

Earthworms as feed protein ingredient can help to replace at least partially the use of soybean and fishmeal in animal nutrition or be used as feed additive, whereas the earthworm grown on a safe feeding substrate, can be an optimal food source or dietary supplement.

Additional research and integration with innovations among different sectors are the key drivers for the near future. However, the outcomes of this study can be useful for the development of a subsidy framework supporting the earthworm dried

meal production chain thanks to the identification of the hotspot stages and their possible mitigations.

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TABLES

Table 1

Main inventory data for SS1.

		Unit	Amount
Input	Fruit and vegetable waste	kg	45.73
	Transport of FVW	km	25
	Diesel	kg	0.196
	Water	m^3	3.66
	Land	m ²	0.41
Output	Vermicompost	kg	12.8
	Fresh earthworms	kg	1

Table 2

Main inventory data for SS2 for the FEED scenario.

		Unit	Amount
Input	Fresh earthworms	Kg	6.25
	Water	m^3	22.41
	Electricity	kWh	2.0
Output	Meal for feed	Kg	1

Table 3

Main inventory data for SS2 for the FOOD scenario.

		Unit	Amount
Input	Fresh earthworms	kg	6.25
	Water	m^3	22.41
	Electricity	kWh	3.67
Output	Meal for food	kg	1

 Table 4

 Prices of product and co-product used in the economic allocation.

Scenario	Product	Amount	Price	Allocation factor
		kg/FU	€/kg	%
FEED	Vermicompost	12.8	0.3	78%
	Meal for feed	1	1.1	22%
FOOD	Vermicompost	12.8	0.3	20%
	Meal for food	1	15.0	80%

 Table 5

 Absolute environmental impact for 1 kg of fresh earthworms in the two scenarios.

	T			
Impact Category	Unit	FEED	FOOD	
CC	kg CO ₂ eq	0.162	0.593	
OD	mg CFC-11 eq	0.0063	0.023	
HT-noc	CTUh	6.07 x 10 ⁻⁹	2.28 x 10 ⁻⁸	
HT-c	CTUh	2.03 x 10 ⁻⁹	7.39 x 10 ⁻⁹	
PM	mg PM2.5 eq	246.8	897.38	
POF	g NMVOC eq	0.293	1.064	
TA	molc H+ eq	0.0107	0.0391	
TE	molc N eq	0.048	0.175	
FE	mg P eq	4.655	16.97	
ME	g N eq	0.410	1.492	
FEx	CTUe	0.140	0.498	
MFRD	mg Sb eq	1.112	4.044	

Table 6Absolute impact for 1 kg of earthworm dried meal produced in the two scenarios.

Impact category	Unit	FEED	FOOD	
СС	kg CO2 eq	2.238	5.944	
OD	mg CFC-11 eq	0.187	0.414	
HT-noc	CTUh	1.46 x 10 ⁻⁷	3.37 x 10 ⁻⁷	
HT-c	CTUh	3.96 x 10 ⁻⁸	9.55 x 10 ⁻⁸	
PM	g PM2.5 eq	1.960	6.374	
POF	g NMVOC eq	4.635	11.801	
TA	molc H+ eq	0.073	0.255	
TE	molc N eq	0.309	1.108	
FE	g P eq	0.234	0.482	
ME	g N eq	3.441	10.936	
FEx	CTUe	3.698	8.328	
MFRD	mg Sb eq	11.397	33.436	

Results of the sensitivity analysis: Impact variation respect to the values reported in Table 6 considering different electricity consumption in SS2, different dried meal price, electricity produced from a photovoltaic plant (PV) and a mixed FU to avoid allocation.

Table 7

	FEED					FOOD						
Impact category	EE - 25%	EE +25%	Low price	High price	PV EE	Mixed FU	EE -25%	EE +25%	Low price	High price	PV EE	Mixed FU
CC	-10.9%	13.6%	-10.4%	10.4%	-33.6%	161.5%	-7.5%	9.4%	-5.5%	5.5%	-49%	15.6%
OD	-15.8%	19.8%	-4.7%	4.7%	-55.4%	74.0%	-13.1%	16.4%	-3.0%	3.0%	-67%	8.6%
HT-noc	-14.8%	18.5%	-5.9%	5.9%	36.5%	91.8%	-11.8%	14.8%	-3.6%	3.6%	46%	10.2%
HT-c	-13.6%	17.0%	-7.3%	7.3%	-15.1%	113.7%	-10.3%	12.9%	-4.2%	4.2%	-20%	12.1%
PM	-4.3%	5.3%	-17.9%	17.9%	-8.5%	279.1%	-2.4%	3.0%	-7.7%	7.7%	-15%	22.0%
POF	-12.1%	15.1%	-9.0%	9.0%	-35.4%	140.0%	-8.7%	10.9%	-4.9%	4.9%	-49%	14.1%
TA	-1.6%	2.0%	-20.9%	20.9%	-3.4%	326.0%	-0.8%	1.1%	-8.4%	8.4%	-6%	23.9%
TE	-0.6%	0.7%	-22.1%	22.1%	-1.2%	344.2%	-0.3%	0.4%	-8.6%	8.6%	-2%	24.6%
FE	-17.5%	21.9%	-2.8%	2.8%	-32.8%	44.1%	-15.6%	19.5%	-1.9%	1.9%	-37%	5.5%
ME	-5.1%	6.4%	-17.0%	16.9%	-11.9%	264.3%	-2.9%	3.7%	-7.5%	7.5%	-21%	21.3%
FEx	-15.4%	19.2%	-5.3%	5.3%	298.9%	82.1%	-12.5%	15.7%	-3.3%	3.3%	367%	9.3%
MFRD	-7.8%	9.8%	-13.9%	13.9%	786.1%	216.2%	-4.9%	6.1%	-6.6%	6.6%	1257%	18.9%

Highlights

- Fruit and vegetable waste as growth substrate to produce earthworm as feed and food
- The environmental impact of earthworm meal as feed and food was evaluated with LCA
- Two scenarios were considered for the dried meal: FEED or FOOD
- Climate change is 2.24 and 5.94 kg CO2 eq respectively for FEED and FOOD scenarios
- The allocation choices affect the results of the environmental impact indicators

System Boundary

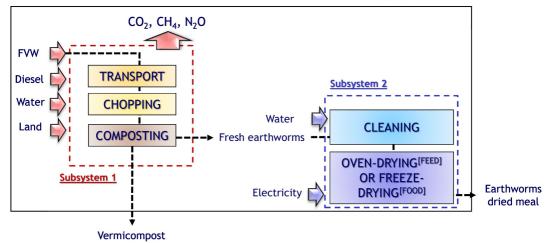


Figure 1

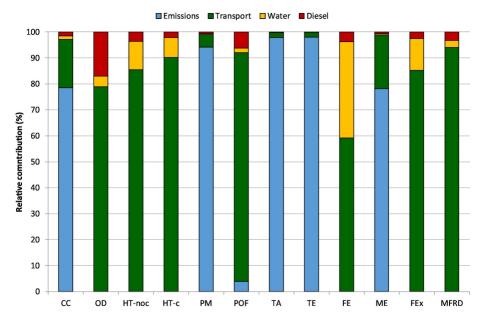


Figure 2

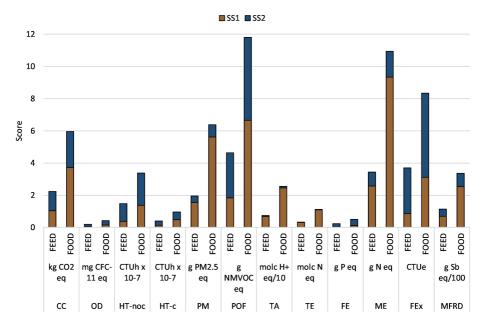


Figure 3