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Stabilization of different starting materials through vermicomposting in a continuous-feeding system: Changes in chemical and biological parameters

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ABSTRACT

In this study the feasibility of *Eisenia andrei* to digest great amount of wastes including horse manure (HM), apple pomace (AP), grape pomace (GP), and digestate (DG) was monitored through a continuous-feeding system. New layers of fresh material were gradually added to form an aged-profile of layers in order to understand the interaction between earthworms and microorganisms during vermicomposting. Thus, changes in chemical and biological parameters were evaluated for 240 days. The earthworm population reached maximum values in 120 d-old-layer, which was related to an increase in overall microbial biomass, assayed as dehydrogenase activity, in all of the processed materials. The pH was generally alkaline or neutral in all of the materials. The electrical conductivity did not modify significantly during vermicomposting, except in the case of the processed GP, and DG. The stabilization, in all of the processed materials, was detected after 240 d of vermicomposting, as indicated the decline in the content of dissolved organic carbon (DOC). The N-NO₃⁻ content exhibited an enhanced in the processed HM and AP, while a generalized decreased was found in the GP, and DG materials in 240 d-old-layer. The decline in microbial biomass activity, in all processed substrates, was related to a decrease in the earthworm activity after 240 d of vermicomposting, indicating a high degree of stabilization. However, the β-glucosidase, phosphatase, protease, and o-diphenol oxidase activities were different according to the age of layers and type of processed material. The phytotoxicity test indicated that the end products of the processed AP and DG were chemically stable and enriched with nutrients in comparison with the HM and GP vermicompost. This fact indicates to stabilization (maturation) in the end product, which is important for its safe disposal as an organic nutrient-rich product.

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1. Introduction

The production of solid waste is approximately estimated in 1.3 billion metric tons, and this amount is predicted to double by 2025 (Hoornweg and Bhada-Tata, 2012). Among the total solid waste produced, organic waste is the largest proportion which includes food scraps, yard waste, municipal activities or sewage sludge, agricultural, and industrial waste (Hoornweg et al., 2013). The increase in production and concentration of intense livestock operations along with the increased urbanization of rural regions have resulted in a greater concern for the safe disposal, treatment and

use of livestock materials, such as manure. For instance, horse manure (HM) has largely been used as a suitable organic amendment due to its high amount in plant-available macro and micro-nutrients. However, the overproduction of this material has led to inappropriate management practices which cause detrimental environmental problems including an excessive input of harmful trace elements, salts and pathogens, increased nutrient loss from soils and emission of toxic gasses (Hutchison et al., 2005). Other industries activities, which are those derived from the juice industry, generate a large amount of solid material commonly known as apple pomace (AP). This material has extensively been used in the food industry as a component in fruit tea (Mamma et al., 2009). However, this material has become in a waste as the result of the higher demand from purchasers for components with better aroma and taste than AP. Grape pomace (GP) is a lignocellulosic material remaining after the grape crushing and pressing in wine produc-

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tion which consists of the stalks, skin, pulp and seeds (Flavel et al., 2005). This waste, due to its high amount in macro and micro-nutrients, is considered as a valuable source as a soil fertilizer (Bertran et al., 2004). However, the inappropriate disposal of GP into agricultural soils can produce serious environmental problems, including the excessive release of tannins and phenols in soils, which could inhibit the root growth (Inbar et al., 1991). On the other hand, the anaerobic digestion of materials, such as farm, agro-industrial and organic residues, has become increasingly popular as an alternative for recycling wastes to generate biogas, resulting in the production of a residual material known as digestate (DG) (Insam et al., 2015). The use of DG as an organic amendment in agriculture can therefore represent an important source of plant-available nutrients, enhancing the soil microbial functionality and biomass (García-Sánchez et al., 2015). However, biogas production has been also accompanied by a depletion of organic carbon (C), which may affect the turnover of nitrogen (N). In spite of this fact, digestate appears not to negatively affect the soil organic C status; a post-treatment of digestate has been suggested by several authors as a mean of improving digestate quality as a soil amendment (Insam et al., 2015; Rehl and Müller, 2011).

The environmental problems associated with the management of these wastes, including HM, AP, GP, and DG, could be significantly reduced by stabilizing them before their use and/or disposal. An environmentally management option could be the vermicomposting which has been reported to be a low-cost technique for the stabilization of hazardous and worthless organic materials of different natures, converting them into safe and valuable end products (Domínguez and Edwards, 2011a). Vermicomposting system is a biological process in which detritivore earthworms interact with microorganisms, accelerating the stabilization of the organic matter and greatly modifying its physical and biochemical properties (Domínguez et al., 2010; Sampedro and Domínguez, 2008). This process consists of two different periods related to the earthworm activity: (i) a vermicomposting period (active phase) in which the earthworm greatly modifies the physico-chemical characteristics of the material as well as its microbial composition (Lores et al., 2006); (ii) a maturation period, following the earthworm displacement toward new layers of fresh substrate, during which microorganisms decompose the processed material by earthworm (Aira et al., 2007a; Gómez-Brandón and Domínguez, 2014). The duration of both, active and maturation phases of vermicomposting, is not fixed and depends on composition of the initial substrates, earthworm species and the rates at which they ingest and process the material (Aira and Domínguez, 2008; Domínguez et al., 2010). Several studies have evaluated the potential use of vermicomposting for the stabilization of different substrates such as manures, AP, GP, and DG (Gómez-Brandón et al., 2011; Hanč and Chadimova, 2014; Hanč and Vasak, 2015; Lazcano et al., 2008; Nogales et al., 2005). Thus, Gómez-Brandón et al. (2013) has investigated the activity of *Eisenia fetida* on the microbial functionality and structure during the vermicomposting of rabbit manure, using a continuous-feeding system. Overall, the stabilization of the organic material was reached within 200–250 days of vermicomposting, as revealed by the lower values of microbial activities compared to the fresh manure. Besides, it was reported a great loss of total carbon in the presence of *E. fetida*, indicating the importance of this epigeic earthworm in the decomposition of organic matter because of their interactions with microorganisms during the process. Hanč and Chadimova (2014) and Hanč and Vasak (2015) have reported the ability of earthworms belong to genus *Eisenia* to transform substrates such as AP and DG into an organic nutrient-rich product, but in a non-continuous system, as indicate the optimal range found in pH, EC, nutrient content and C:N ratio. To this date, less is reported on microbial activities and biochemical changes that occur during

the vermicomposting process of these materials in a continuous-feeding system. On the other hand, studies conducted by Gómez-Brandón et al. (2011) and Nogales et al. (2005) have observed that the combined action of microorganisms and *E. andrei* enhanced the stabilization of materials such as GP using a non-continuous system. This result was evidence by the lower values of labile C pool and overall microbial biomass, measured as the basal respiration and dehydrogenase activity. However, these studies were performed to monitor the effectiveness of the active phase during vermicomposting of GP, but in lab-scale systems. Therefore, there is a need for further studies to evaluate the efficiency of a continuous-feeding system vermicomposting to transform a wider range of different starting materials, and to analyse the stability of the end products as fertilizers. Moreover, it is essential to establish to what extent the relationships between earthworms and microorganisms are crucial in the decomposition of the organic matter and therefore, in the stabilization of organic substrates. To do this, the objective of this study was aimed in investigating the efficiency of *E. andrei* to convert different types of organic materials, including HM, AP, GP, and DG in a continuous-feeding system. With this system, we were able to establish a profile of layers to observe the different phases of interaction between earthworms and microorganisms during the decomposition of the organic matter in the different materials. The evolution of microbial biomass and important enzymatic activities were monitored as well as chemical parameters through an increased aged-profile of layers with different degree of maturation (stabilization), from the top to the bottom of the system. Furthermore, the stabilization of the vermicompost was evaluated during the germination of *Lepidium sativum*, L. seeds to assess the feasibility of using these materials as an agricultural resource. Information regarding this subject will contribute in better understanding the interaction between earthworm activity and microorganisms during the biological transformation and may have important contributions in the large-scale valorisation of organic materials through vermicomposting system.

2. Materials and methods

2.1. Organic materials

HM was collected from a horse farm near Prague (Kladno, Czech Republic), which contained a mixture of solid and liquid horse faeces. AP was provided by the Severofrukt company (Terezin, Czech Republic) and was mainly obtained from apple juice production. GP was composed mainly of pulp and stones of grapes remaining after wine production and was provided by a winery company located in South Moravia (Czech Republic). DG was obtained from the biogas plant of an agricultural cooperative placed in Krasna Hora nad Vltavou (Czech Republic). The composition of the DG was approximately 50% manure slurry, 40% corn silage, and 10% haylage. In order to reduce the worm toxicity factors such as high content of ammonia (HM and DG) and low pH level (AP and GP), the starting organic materials were pre-composted in 70 L capacity laboratory reactors following the same procedure described elsewhere (Hanč et al., 2012). The physico-chemical composition of each organic waste is shown in Table 1.

2.2. Vermicomposting process

The vermicomposting process was carried out in vertical continuous feeding vermireactors (VermiHut Worm Bin). Twelve vermireactors were set up, three per each organic material tested (HM, AP, GP, and D) and were composed of trapezoidal trays (40 × 40 × 18 cm) with a perforated bottom to allow the movement of earthworms from one tray to another. 1 L of cow manure

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