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\*Corresponding author: Ramírez-Iglesias, E. E-mail address: ec.ramirez@ericonsult.org

# Interaction between organic substrates of the southern region of the Ecuadorian Amazon and the presence of *Eisenia foetida*

Interacción entre sustratos orgánicos de la región Sur de la Amazonía ecuatoriana y la presencia de *Eisenia foetida* 

# Ramírez-Iglesias, E.<sup>a,\*</sup>, Gualán R.<sup>a</sup>, Guayanay V.<sup>a</sup>, Ramírez-Iglesias JR.<sup>b</sup>

<sup>a</sup> Universidad Estatal Amazónica. El Puyo- Ecuador. <sup>b</sup> Universidad Internacional SEK. Quito-Ecuador.

#### A B S T R A C T

This study aimed to evaluate the possible interaction between the chemical properties of some organic substrates of the southern region of the Ecuadorian Amazon and the development and reproduction of the red earthworm (*Eisenia foetida*), to obtain a bioproduct that can be used as organic fertiliser by producers of the region. The treatments used in the experience were: domestic organic waste (DOW), guinea pig excreta (GPE) and sugar cane waste (SCR), the control (CT) was a clay loam soil from the area. The chemical characterisation of the substrates was carried out before and during the introduction of the earthworms. After the pretreatment period (15 days), 30 individuals of *E. foetida* were placed in each bed, totalling 360 individuals. Initially, all the substrates showed low nutritional contents, as well as high C/N ratios, a trend that was maintained until the end of the experiment. After 25 days of composting, it was observed that the GPE treatment showed a pH close to neutrality, as well as greater nutrient availability. GPE and DOW were the organic residues with the best conditions for the development of *E. foetida*. After 45 days of the introduction of the earthworms, the vermicompost obtained could already be used as organic fertiliser, given the availability of nutrients.

#### RESUMEN

La presente investigación plantea evaluar la posible interacción entre propiedades químicas de algunos sustratos orgánicos de la región Sur de la Amazonía ecuatoriana y el proceso de desarrollo y reproducción de la lombriz roja (*Eisenia foetida*), para la obtención de un bioproducto que pueda ser utilizado por los productores de la región. Los tratamientos empleados en la experiencia fueron: residuos orgánicos domésticos (DOW), excretas de cuy (GPE) y residuos de caña de azúcar (SCR), el control (CT) fue un suelo franco arcilloso de la zona. La caracterización química de los sustratos se realizó antes y durante la introducción de las lombrices. Después del periodo de pretratamiento (15 días), se colocaron 30 individuos de *E.foetida* por cantero para un total de 360 individuos. Inicialmente todos los sustratos mostraron bajos contenidos nutricionales, así como altas relaciones C/N, tendencia que se mantuvo hasta el final de la experiencia. A los 25 días de compostación se observó que el tratamiento GPE mostró un pH cercano a la neutralidad, así como mayor disponibilidad de nutriente. GPE y DOW fueron los residuos orgánicos con mejores condiciones para el desarrollo de la *E. foetida*. A los 45 días de introducción de las lombrices, el vermicompost obtenido ya podría ser utilizado como abono orgánico, dada la disponibilidad de nutrientes.

Palabras clave: bioproductos, microorganismos, sustrato-suelo, nutrientes.

# INTRODUCTION

In the Ecuadorian Amazon, the expansion of the agricultural frontier continues to generate large amounts of organic residue (Carvajal Barriga et al., 2013; Solíz Torre,2015; Orejuela-Escobar et al., 2021). In order to access the viability of bioproducts originating from organic residues (Orejuela-Escobar et al., 2021) that can be incorporated into value chains of small producers (Ramírez-Iglesias et al., 2017; Ramírez-Iglesias, 2020), the quality of source substrates need to be determined in terms of C/N ratios (Hu et al., 2021;

Ramakrishnan et al., 2021), decomposition times (Anderson and Ingram, 1989) and nutrient content (Castro-Bedriñana et al., 2020; Das and Deka, 2021).

*Eisenia foetida* is known to accelerate transformation processes in organic matter (Cataldo et al., 2011; Bellitürk et al., 2020; Hu et al., 2021) and improve nutrient contents by augmenting microbial populations (Prisa, 2020), increasing soil aeration (Hrženjak et al., 1992), water retention capacity, reducing  $H_3O$  concentration (Dulaurent et al., 2020) and nutrient availability, favouring the absorption by plants (Castro-Bedriñana et al., 2020; Hu et al., 2021). However, these benefits may vary depending on the substrate and its treatment.

The oxidation and stabilisation of the residues due to the synergy between earthworm and microorganism activities present in the substrate-soil (Prisa, 2020; Gwenzi, 2021; Khatibi and Hassani, 2021) is carried out by catabolic processes (Grdisa et al., 2001) in the intestines of earthworms. This process begins with the activation of aerobic cellulolytic microorganisms, in the digestion of lignocellulose in the intestine, and later enhances the presence and activity of diverse microorganisms specialised in this medium (Fernández et al., 2009; Dulaurent et al., 2020; Stanis Sagayaraj and Deepan, 2020).

It has been determined that *E. foetida* has a set of digestive enzymes (cellulase, xylanase, acid phosphatase and alkaline phosphatase) (Cataldo et al., 2011; Grdisa et al., 2001) that allow them to digest protozoa, fungi and partially decomposed plant remains (Devi and Khwairakpam, 2020; Dulaurent *et al.*, 2020) enhancing the cycling of nutrients. The activity of decomposer microorganisms showed increased metabolic activity in processing OM when they are in contact with the secretions left by earthworms, containing amino acids, glycosides, and a glycoprotein (Castro-Bedriñana et al., 2020; Ross et al., 2021).

This study aimed to evaluate the relationship between the chemical properties of organic substrates (domestic organic waste (DOW), guinea pig excreta (GPE) and sugar cane residues (SCR), and the development and reproduction process of the red earthworm (*Eisenia foetida*), to obtain a biofertiliser as a bioproduct that can be produced and utilised by small scale agriculture, in an effort to reduce the use of agrochemicals as an agroecological conversion strategy.

# **MATERIALS AND METHODS**

#### Study site

The study was conducted in El Pangui (S 3°39'50.37" W 78°35'56.26"), Ecuador, at an altitude of 855 m.a.s.l., with a mean annual rainfall of 1626 mm and mean temperature range between 22°C and 24°C (Peñaherrera Cabezas, 2016). Regional economic activities carried out by mostly rural population, include the cultivation of crops such as corn (*Zea mays*), sugar cane (*Saccharum officinarum*) (Heredia-R et al., 2020), and the rearing of guinea pigs (*Cavia porcellus*) (Ramírez-Iglesias et al., 2020; Ross et al., 2021).

#### Substrate trial setup

The examined residues, readily available from local agriculture, namely domestic organic waste (DOW), guinea pig excreta (GPE), and sugar cane fibre (SCR), were mixed in a 1:1 ratio with untreated soil, following the methods of Khatibi and Hassani (2021). In this study, the mixtures were 13.5 kg of residue and 13.5 kg of clav loam soil. A control treatment (CT) was composed of 27 kg of clay loam soil, without added OM, the type of soil that predominated locally (Inceptisols), with a pH range between 4 and 5 and low nutrient levels (Moreno, 2017). The soil-substrate mixtures and the control soil were placed in beds with the dimensions length - width - height of 50 x 30 x 30 cm, and with the stability to hold 45 kg (Kiruthika V and Vijayan P, 2020). Thus, three beds were assigned to each substrate and the control. All beds contained a small opening and were placed inclined to allow proper drainage (Fernández et al., 2009). Beds were subsequently conditioned for 15 days.

### Substrate baseline

A baseline of substrate quality was established by taking 10 samples of 200 g from each of the 12 Beds( $t_0$ ), drying them at 40 °C for 3 days in an oven to minimise the volatilisation of the present nitrogen, subsequently measuring pH in water using a substrate : water ratio of 1 : 2; carbon (% C) via wet oxidation without heating (Ciavatta et al., 1989). Wet digestion with H<sub>2</sub>SO<sub>4</sub> and distillation with Microkjeldahl (Ibeto et al., 2019) were used to determine total nitrogen (N). For available P, sodium bicarbonate (Watanabe and Olsen, 1965) was used as an extractant, and determined by the colourimetric method molybdateascorbic acid (Murphy and Riley, 1962) using a Spectronic 20 Thermo Scientific, Genesys 22. The nutrient levels of K and Ca and Mg were determined by atomic absorption, using Mehlich III (Mehlich, 1984) as an extractant and the atomic absorption spectrophotometer IC-3300.

# Substrate sampling and examination

After establishing de substrate baseline (t<sub>o</sub>), 30 specimens of the red earthworm Eisenia foetida were introduced in each of the 12 beds, 25  $(t_1)$  and 45  $(t_2)$ days after their introduction, soil properties were again examined as above in 10 samples of 200 g from each bed. In addition, 10 earthworks containing a clitellum from each of the 12 beds were collected for biometric measurements of length and width (Grdisa et al., 2001; Devi and Khwairakpam, 2020) and returned to their respective beds. During the second sampling  $(t_a)$ , a total census of all earthworms and cocoons was carried out in all beds (Fernández et al., 2009; Cataldo et al., 2011; Devi and Khwairakpam, 2020; Heredia-R et al., 2020) to evaluate the reproduction of Eisenia foetida in the evaluated organic wastes (Ramírez-Iglesias et al., 2017).

### Statistical analysis

Descriptive analysis was used for positional measures such as mean and median, and dispersion measures such as standard deviation. Likewise, an analysis of variance (ANOVA) was performed, using the Infostat program (Di Rienzo et al., 2020), to determine statistically significant differences in the variables studied between treatments and a comparison of means with Duncan's Multiple Range method (Duncan, 1974), with significance level of p<0.05.

Additionally, a Principal Component Analysis (PCA) was performed to better discriminate the correlations between the treatments and the measured variables. The PCA was based on the standardised variables related to substrates and earthworms, discriminated by treatment domestic: organic waste (DOW), guinea pig excreta (GPE), sugarcane residues (SCR), and the control treatment (CT). That is, variables with mean 0 and variance 1. This is equivalent to taking the components not from the covariance matrix but from the correlation matrix (in the standardised variables match covariances and correlations). Thus, equal importance is given to all original variables.

#### **RESULTS AND DISCUSSION**

# Chemical characteristics of the substrates in relation to the introduction of red worms (*Eisenia foetida*).

Following the 15d conditioning period, the substrates were characterised as shown in Table 1. GPE resulted to be the most acidic substrate (pH 5.46) as well as the richest in P, K, Ca, while SCR had the highest C/N ratio, followed by the ratios in GPE, DOW and CT, which were residues of plant material with cellulose and lignin, possibly elevating these ratios (Ramírez-Iglesias et al., 2017) in comparison to excreta. All nutrient concentrations were lower in the control soil (CT).

The chemical characterisation of local organic residues provides a quantitative baseline to infer decomposition times (Dulaurent et al., 2020), but also to gauge the potential of agricultural residues for the production of biofertiliser (Ramirez-Iglesias, 2020).

The pH in all organic fertilisers varied according to the substrates used. However, the initial conditions  $t_0$ did not show differences (p > 0.05): GPE (5.46)  $\leq$  SCR (6.04)  $\leq$  CT (6.06) <DOW (6.15). Initially ( $t_0$ ), before the introduction of *Eisenia foetida*, the pH showed tendencies towards neutrality, except for the GPE, which showed a higher degree of acidity compared to the other substrates evaluated. Ramnarain et al. (2019) pointed out that a higher acidity for organic substrates is due to the use of excreta, but the addition of ammonia contributes to the acidity of the substrate, therefore it is advisable to apply a composting period beforehand. This period allows the worms to position in the middle as they seek neutrality (Table 1).

The following figures show the chemical characterisation of the substrates used in relation to the introduction of *Eisenia foetida*, at three different times.

Figure 1a, shows that at  $t_0$  the C/N ratios presented the following trend SCR > GPE > DOW > CT (Duncan p<0.05), being higher in those residues composed of plant material, whose cell wall is mainly composed of cellulose and lignin, which makes the ratios possibly higher (Ramírez-Iglesias et al., 2017), compared to excreta.

**Table 1**. Chemical characterisation of substrates after 15 days of conditioning  $(t_0)$ . **Cuadro 1**. Falta texto en español.

Treatment	C/N	Р	К	Са	Mg
		(mg/kg)	(mg/kg)	(mg/kg)	(mg/kg)
t <sub>o</sub>					
DOW	23.18	3.28	0.18	1.56	1.96b
	±4.06	±0.02	±0.02	±0.05	±0.14
GPE	24.69	7.96	1.69	2.24	2.42
	±3.19	±0.62	±0.12	±0.63	±0.16
SCR	33.15	4.02	0.86	1.69	1.86
	±4.19	±0.8	±0.03	±0.09	±0.2
СТ	4.20	1.92	0.19	0.08	0.32
	±0.02	±0.06	±0.06	±0.01	±0.02

Note: results are presented as mean ± SD (n=10). Domestic organic waste (DOW); guinea pig excreta (GPE); sugar cane residues (SCR); control treatment (CT). Total carbon (%C), total nitrogen (%N), and elements such as available phosphorus (P), potassium (K), calcium (Ca) and magnesium (Mg). (Duncan p<0.05).

Total P at  $t_0$  showed higher content in GPE excreta, followed by SCR, DOW, and CT (Fig, 1b). Both K and Ca and Mg (Fig 1c, 1d and 1e) showed higher contents in GPE, followed by SCR, however, in DOW, Mg values were higher when compared to respect to SCR. In all cases, nutrient concentrations were lower in the soil used as control (CT) than in the other treatments. The characterisation of local organic residues that have the potential to become biofertilisers or as substrate for earthworm growth are quantitative indicators to predict decomposition times (Dulaurent et al., 2020), but also indicate the potential of locally generated resources that are a result of agricultural activity (Ramirez-Iglesias, 2020) (Figure 1b, c y d).

When evaluating the substrates that were subjected to conditioning and the direct effect of *E. foetida*, signi-

ficant variations were observed when compared to the initial characteristics.

At t1 (25 days), pH tends to neutrality in all substrates, improving in the case of GPE with the initial acidity (5.44).

In the case of C/N ratios, they tend to be higher in GPE, followed by SCR, DOW and finally in CT. Some authors (Ibeto et al., 2019; Khatibi and Hassani, 2021) point out that *Eisenia foetida* easily processes many organic substrates which benefits their metabolism, translating into a greater production of amino acids and proteins which are released into the environment as a product of their digestion (Grdisa et al., 2001). This is possibly what gradually leads the different substrates evaluated to neutrality, as well as the beginning of equilibrium in the C/N ratios (Paco et al., 2011).



**Figure 1.** Variation of nutrient contents during the vermicomposting process at t0 (15 days of conditioning), t1 (25 days) and t2 (45 days) after the introduction of *E. foetida.* : (*a*) C/N ratio; (b) P content; (c) K content; (d)Ca content (e) Mg content . Different letters at the same time indicate statistically significant differences between treatments (n=10) (p<0.05)

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Figure 1 shows how the semi-decomposed material (at 25 and 45 days of composting) presents significant variations in available P (Fig.1b), showing the following trend: GPE  $\geq$  SCR > DOW > CT. It has been indicated in some studies that a high percentage of the available P applied to the soil becomes immobile and is not available for plant absorption due to the phenomenon of fixation, precipitation or conversion to the organic form, and perhaps some leaching (Bellitürk et al., 2020; Devi and Khwairakpam, 2020), because of the wetting of the substrates, the availability of this element, in this study, tends to increase which is presented as a fertilisation option when using it in these agroecosystems, characterised by the acidity of their soils, where available P is one of the limiting elements for plants (Sánchez et al., 2018).

Higher concentrations of K (Fig. 1e) were found in excreta without any treatment when compared to plant material and CT, as well as for Ca and Mg availability. The improvement of nutrients in the different substrates is possibly due to the increase in microbial biomass (Grdisa et al., 2001) as a result of the synergistic action between earthworms and OM (Kiruthika and Vijayan, 2020).

In the short term, the presence of earthworms in this first phase possibly promoted mineralisation processes, showing a greater availability of nutrients. Some authors (Benitez et al., 1999; Grdisa et al., 2001), indicate that there is a high correlation between OM decomposition and the presence of indicators such as hydrolase and dehydrogenase, both enzymes directly related to OM decomposition and enhanced by the presence of earthworms.

At 45 days after earthworm establishment ( $t_2$ ), substrate conditions presented significant increases in terms of nutrient availability (Figure 1).

The C/N ratios decreased as the earthworms were on the substrates longer. In Figure 1a  $(t_2)$ , it can be observed that vermicompost (GPE) values oscillating between 9 and 11 were expected, which is between the recommended ranges (Benitez et al., 1999; Ramnarain et al., 2019; Vázquez-Villegas et al., 2021) in terms of fertility. Above these values, the amount of carbohydrates would be prevailing, while below there would be a deficit of N, which would limit its transformation into amino acids that can be used by the plant during its development (Porta et al., 2014). Possibly, this is not beneficial for the growth of the earthworms, since there are higher amounts of structural carbohydrates such as lignin in the medium, which compromise the metabolic degradation of OM in a short time, affecting the growth of Eisenia foetida, unlike the GPE, where the C/N ratios show more balanced values (carbohydrates and amino acid formation), which tend to be better compared to the other substrates.

However, the C/N ratio for CT presents a significant increase in relation to the other treatments, which could be related to a prevalence of the carbon content (carbohydrates) over the nitrogen content, for which the fermentation, in this case, will be slow and at a low temperature, taking longer to obtain the final compost. The C/N ratio is an index of the quality of the organic substrate of the soil, where high values imply that the organic material decomposes slowly, as microorganisms immobilise the nitrogen, so it cannot be used (Gamarra et al., 2017).

C/N ratios were more favourable in GPE, possibly due to a higher presence of microbial biomass, which together with the action of earthworms (Hu et al., 2021) promoted the mineralisation of the material, and possibly also higher concentrations of inorganic nitrogen (NH<sub>4</sub><sup>+</sup> and NO<sub>3</sub><sup>-</sup>).

The available P showed a significant increase compared to the evaluation of the same in previous times. When comparing substrates at the same time  $(t_2)$ , there is a higher availability in GPE compared to the rest of the treatments. It should be noted that this element tends to be a limiting element in its organic forms, especially in acid soils, a condition of Amazonian soils (Turbe et al., 2010; Sánchez et al., 2018). In this sense, the joint presence of earthworms and microorganisms possibly enhanced the action of phosphatase enzymes, which hydrolysed P more effectively, and contributed to the transformation and greater availability of P in its chemically assimilable forms by the plant (HPO<sub>4</sub><sup>-2</sup> y H<sub>2</sub>PO<sub>4</sub>) (Singh et al., 2020).

It is important to note that the synergistic action of earthworms tends to vary according to the type of substrate on which *Eisenia foetida* has a longer action time (Devi and Khwairakpam, 2020; Heredia-R et al., 2020), so bacteria such as *Bacillus mesentericus* (Vázquez-Villegas et al., 2021) and *Pseudomonas putida* (Karwal and Kaushik, 2021) solubilise the organic forms of phosphorus (orthophosphate) and transform them into plantassimilable phosphates.

Soil biological indicators include properties associated with biological activity on organic matter, such as microbial biomass, basal respiration, and enzymatic activities; in addition to indices related to abundance, diversity, food chains, stability of microorganism communities, as well as organisms associated with the mesofauna, such as earthworms, annelids, nematodes and arthropods (Idowu et al., 2008). It is important to highlight that the organic matter of the soil contains around 5% of the total N, but it also contains other essential elements for plants such as phosphorus, magnesium, calcium, sulfur and micronutrients, of great importance for mineral nutrition of plants developing in different substrates. In this case, cations such as K, Ca and Mg generally show trends that favour GPE followed by SCR > DOW and finally CT.

# Relationship between the quality of the substrates and the development of *Eisenia foetida*

Figure 2 shows the effect of substrates on earthworm growth at 25  $(t_1)$  and 45 days  $(t_2)$ . At  $t_1$  it is observed that the earthworms show significant differences in

growth in length and diameter (Figure 2a and 2b) according to the substrate use GPE > DOW > SCR  $\ge$  CT.

It is important to mention that when contrasting the C/N ratios (Fig 1a) the treatments with higher relationships are SCR (33.15) and GPE (24.69). However, according to standard deviation, DOW and GPE could be equal.

In the catabolism suffered by the substrates by the earthworms, it is observed that it is favourable to the GPE in the case of nutrient availability, which is reflected in the length and diameter reached by the earthworms during the evaluated period.

Some authors (Fernández et al., 2009; Das and Deka, 2021) mention that there is a synergy between the presence of earthworms and their degradative capacity, which in turn is directly related to the quality of the substrates on which they act. Therefore, being in an environment where nutrients are present, earthworms tend to excrete up to eleven times the level of P, K and Mg present in the medium (Vázquez-Villegas et al., 2021), changing the pH towards neutrality.

At  $t_2$ , diameter (Figure 2b) tends to promote GPE > DOW  $\ge$  SCR  $\ge$  CT. It is observed that there is also a relationship between good development in terms of size and nutrient availability and the metabolic processing of the substrates.

In the decomposition processes of OM, an active phase and maturation or indirect phase are distinguished. In the first phase, the worms modify the physical and chemical properties of the substrate by increasing the microbial populations. In the second phase, it is the microorganisms that take control of the decomposition, previously processed by the worms (Castro-Bedriñana et al., 2020). Then, the passage of OM through the intestines of the worms may contribute to modifications such as particle size decrease (Ramnarain et al., 2019), the addition of sugars, mucus production, and excreted substances such as urea and ammonium that promote greater assimilation and processing of substrates by the microbial populations (Biabani et al., 2018) and in turn contribute to the growth of *Eisenia foetida* on each substrate.

It is possible that the time of evaluation of the presence of the earthworm allowed for a more visible microbial potentiation produced by its secretions (Hu et al., 2021), which in turn favoured its growth (Kiruthika and Vijayan, 2020). The growth of the worms is related to the quality of the food provided, and greater success in the breeding of worms, with a rapid reproduction of the breeding stock, is ensured when the food provided is of optimum quality, thereby increasing development and quantity of worms in a short time, as can be seen in the results obtained. Additionally, certain important characteristics must be gathered in the food substrate, such as a pH value between 7.0 - 8.5; a humidity between 80 to 85%; and organic matter must be biodegradable. The selection of food will depend largely on its availability at the breeding site, this being one of the factors that determine the feasibility and location of the worm crop (Barbados et al. 2003). The differences between the quality of the substrates used in the development of the worms and their chemical composition, as well as the mixtures of waste used for composting, can interfere with the reproduction and mortality rates of earthworms (FAO, 2013).



**Figure 2.** Length (a) and diameter (b) reached by *E. foetida*, at two sampling times  $t_1$  (25 days after introduction) and  $t_2$  (45 days) on substrates: DOW: domestic organic waste, GPE: guinea pig manure, SCR: sugarcane stalk, CT: control treatment; (a). Results are presented as mean ± standard deviation (n=10). For each figure, bars with different letters have mean values that are significantly different between substrates at p < 0.05.

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Regarding the use of excretes as substrate, Guinea pig manure has been considered as one of the best quality, together with horse manure, due to its physical and chemical properties, and is commonly used by farmers as a direct fertiliser (Alva et al, 2107; Sinche-Villafuerte, 2022). In addition, it is easily collected, compared to manure from other animals, since it is normally found in abundance in this area. However, its direct use can constitute an environmental threat due to the emission of gases and the washing of nutrients, since the organic matter derived and the gases produced in the decomposition process have undesirable effects on the environment and health (Wuana and Okieimen, 2011; Villalobos-Maldonado et al., 2017; Das and Deka, 2021). Manure fermentation has a positive effect on the stability of the anaerobic process, due to its buffering capacity and high content of trace elements. In addition, the biodigestion process also decreases the number of pathogens in the excreta used as feedstock for biodigesters. One of the by-products of anaerobic fermentation is biol, which is rich in microorganisms, phytohormones, and nutrients (Benitez et al., 1999; Cataldo et al., 2011; Battacharyya et al., 2015;). The application of these biols to the soil can eliminate contamination, restore bacterial flora, and act as a foliar fertiliser(Cataldo et al., 2011; Devi and Khwairakpam, 2020).

Additionally, some authors (Alva et al., 2017; Castro-Bedriñana et al., 2020; Stanis Sagayaraj and Deepan, 2020) indicate that according to the diet consumed by the guinea pig, it is possible that the quality of its excreta varies in nutrient availability. This option has been used in anaerobic composting and has been observed as an excellent alternative to be used in biols. The variation in the composition of manure depends on the animal species, its feeding, dry matter content (fresh or dried state) and management. For general practice and use, manure can be considered to contain N (0.5%), P (0.25%) and K (0.5%). When exposed to sun and weather, manure generally loses its value (Blouin et al., 2019; Ramnarain et al., 2019; Dulaurent et al., 2020).

Earthworms enhance the nutritional characteristics of the substrates so that the stabilised product can be applied as organic fertiliser on crops. However, the times when the stabilised vermicompost can be used are different from the times the worms need to increase their populations. This aspect should be considered in case they are to be used as a protein source.

To holistically evaluate the synergy between local substrates and the presence of *Eisenia foetida*, a multivariate analysis was carried out.

The analysis determined the different substrates and the trends favoured in the variables measured, not only in the treatments but also in the earthworms. The treatment as GPE showed greater presence towards the I and IV quadrant, while SCR was like CT located in the II quadrant, and finally, DOW located in the III quadrant.



**Figure 3**. Principal Component Analysis (PC1 and PC2) relative to the variables in the treatments (organic substrates) and the variables measured in *E. foetida*. Variables measured in the substrates: C/N: carbon/nitrogen ratios, total N, available P, K, Ca and Mg. Variables measured in earthworms: length, diameter and population abundance.

**Figure 3**. Principal Component Analysis (PC1 and PC2) relative to the variables in the treatments (organic substrates) and the variables measured in *E. foetida*. Variables measured in the substrates: C/N: carbon/nitrogen ratios, total N, available P, K, Ca and Mg. Variables measured in earthworms: length, diameter and population abundance.

PC 1 accounted for 81.1% of the variability and the second component for 16.3%. The largest number of variables related to substrate quality were collected by CP1, while variables such as worm length, C/N ratios and pH were collected by PC 2.

The organic residue formed by GPE was the one that showed the highest correlation with variables measured in the earthworms such as diameter, length, number of cocoons, and abundance. Likewise, variables oriented towards substrate quality such as available P, total N, K, Ca and Mg, also showed a higher clustering in the I and IV quadrant, and towards GPE, followed by DOW, then by SCR and finally by CT.

The organic residue formed by GPE, showed the highest correlations with the number of cocoons (p < 0.05). It is possible that the time in which the presence of the earthworm on the substrates was evaluated, allowed a more effective microbial potentiation product of the secretions of the earthworm (Hu et al., 2021), which in turn promoted its growth (Devi and Khwairakpam, 2020) in GPE, but probably the time was insufficient to observe a clearer trend of the cocoons in different substrates. The quality conditions of the GPE treatment from the beginning were nutritionally beneficial for earthworm establishment (Gudeta et al., 2021). However, the product obtained by the earthworm-substrate synergy could be employed as a bioproduct that allows the reuse of locally generated waste, according to the determined chemical characteristics.

### CONCLUSIONS

The results obtained in this study showed that Guinea pig excreta (*Cavia porcellus*) (GPE) and domestic organic waste (DOW) exhibit a greater synergy in the association with *E. foetida*, showing a pH close to neutrality, as well as a greater availability of nutrients. This supports the potential use that could be given, through composting and vermicomposting, to a stabilised bioproduct which can be marketed or used locally for planting crops, or in the case of worms as a source of protein.

The research perspectives point to the characterisation and use of organic waste generated locally in the Ecuadorian Amazon, which should be considered as an agroecological strategy by producers to minimise the use of external inputs and fertilisers.

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