

Composting-vermicomposting of pigeon dropping waste. A contribution to the reduction of urban contamination

Carolina Elisabet Masin^{1,2} , Alejandra Duran¹ , Cristina Susana Zalazar^{1,3} ,
Maria Emilia Fernandez^{1,*} 

¹Instituto de Desarrollo Tecnológico para la Industria Química (INTEC, UNL-CONICET), Ruta Nacional 168 Km 0, 3000 Santa Fe, Argentina.

²Facultad de Ciencias de la Salud, Universidad Católica de Santa Fe, Echagüe 7151, 3000 Santa Fe, Argentina.

³Departamento de Medioambiente, FICH-UNL, Ruta Nacional 168 Km 0, Ciudad Universitaria, 3000 Santa Fe, Argentina.

*Corresponding author: mefernandez@intec.unl.edu.ar

Original Research

Received:
27 March 2024
Revised:
11 July 2024
Accepted:
16 December 2024
Published online:
20 December 2024

© 2025 The Author(s). Published by
the OICC Press under the terms of
the [Creative Commons Attribution
License](#), which permits use, distribu-
tion and reproduction in any medium,
provided the original work is prop-
erly cited.

Abstract:

Purpose: Without a proper treatment, pigeon dropping waste (PDW) in the urban environment is a sanitary risk for the population because of nasty and irritating odors, a very high content of ammonium, and the presence of pathogens. This study deals with the recycling of PDW from a dovecote, situated in a public city plaza, to achieve its stabilization and eliminate sanitary risks.

Method: Composting of PDW with other locally available lignocellulosic residues (sawdust and chipped tree pruning) and vermicomposting employing *Eisenia fetida* earthworms was applied. Two designs were selected for the vermicomposting stage: (1) Sectorized, with a zone with earthworms and another zone of composted PDW with gradual incorporation to the first one and, (2) Integrated, consisting of the composted PDW, with *E. fetida* in the entire solid.

Results: The composting allowed a partial stabilization of the original mix of PDW, given its highly elevated initial content of ammonium (8693 mg/kg). The combined processes almost eliminated the ammonium present (> 99% reduction) and the action of earthworms shortened the maturation time. Organic matter and electrical conductivity of the solids had important reductions. The treatment affected the resulting characteristics of the solids obtained but the germination index was above 80% in both cases.

Conclusions: Both designs allowed the obtention of two mature, non-phytotoxic vermicomposts. The sectorized vermicompost had better properties and had the advantage of being obtained with fewer initial number of earthworms.

Keywords: *Eisenia fetida*; Pigeon waste; Ammonium; Phytotoxicity; Pathogen elimination

1. Introduction

Urban pigeons are commonly present in almost every city in the world. They have found a satisfactory habitat specially in great metropolises: buildings for the construction of their nests, food, lack of predators and warmer temperatures in comparison to rural zones have led to the growth of their population. Among the problems associated with the coexistence with these birds are those of sanitary type (pigeons are known as bearers of parasites and infectious diseases), air, water and food contamination and structural damage in buildings due to the corrosivity of their depositions. In recreational sites, generation and accumulation of

pigeon dropping waste (PDW), mainly composed of feces and some feathers and food, can intensify the likelihood of occurrence of sanitary problems and their management must be carried out.

“El Palomar” (the dovecote), a place of traditional family recreation built in 1940, stands on one of the main plazas in Santa Fe city (Argentina). Nowadays, an approximate population of 1,200 pigeons nests in this dovecote. The cleaning and general maintenance of this building is the responsibility of the local government, and the ultimate disposition of these wastes without treatment is a problem to address.

Problems associated with the PDW can be mitigated by

stabilizing the organic matter, the content of ammonium and reducing pathogenic agents (mainly fecal coliforms and *Salmonella* sp.). These indicators are the most frequently recommended worldwide to confirm stability of a treated substrate (Leconte et al., 2009). As for other organic wastes, composting and vermicomposting represent low cost alternatives, economically and energetically. Both processes allow the removal of toxic substances and the stabilization of organic wastes, generating solid products which can be used in agriculture for plant growth or applied as soil amendment, depending on their quality (Maharjan et al., 2023). Composting involves the biochemical microbiological degradation of organic wastes while, in vermicomposting, a synergistic action is established between microorganisms and earthworms. The mechanical disintegration and enzymatic degradation of the solid wastes by the earthworms, their mobility within the substrate, the excretion of mucus and the production of bio-aggregates, all of this favors the aeration and the enrichment of the product with nutrients like phosphorus and nitrogen, which ultimately become available for the degrading microorganisms (Yuvaraj et al., 2021).

Specifically, PDW usually presents extreme elevated levels of ammonium, hindering their handling and treatment. Although composting/vermicomposting of birds' wastes have been reported, it has been mostly focused on poultry. Leconte et al. (2009) co-composted rice hulls and/or sawdust with poultry litter and found an increase in N-NH_4^+ content during the first three months and a rapid decrease afterwards, with higher rates of nitrification using sawdust in the mixture. Pizarro et al. (2019) compared raw poultry litter and composted one as lettuce (*Lactuca sativa* L.) safer fertilizer. In a study of combined composting-vermicomposting of poultry litter, rice hulls and/or eucalyptus sawdust, the authors found that the addition of the earthworms action enhanced the quality of the final product (Masin et al., 2020); this conclusion has also been obtained by other authors (Niedzialkoski et al., 2021; Srivastava et al., 2023). In the case of PDW, information is extremely scarce. A characterization of this waste was presented by Villa-Serrano et al. (2010) in terms of organic matter, C/N ratio, P and K contents and micronutrients, and presented as a soil fertilizer without further treatment. Only Singh et al. (2019) reported co-vermicomposting with cow manure and the action of

Eisenia fetida [Savigny, 1826] for the stabilization of this waste. They compared mixtures at different ratios of both wastes and the nutrients (N, P, K, Ca, OM, etc.) changes in the vermicomposts obtained, although neither ammonium nor pathogens' presence was surveyed.

Within this context, and with the aim of solving a particular contamination problem for the inhabitants of the city, this work explores the outcome of composting-vermicomposting PDW from a public dovecote combined with local organic materials. Two types of constructions were designed for the vermicomposting stage, a traditional integrated one and a sectorized one, for the gradual incorporation of composted PDW. As a result, the aim is to obtain a sanitized bioproduct with potentiality to be used as soil amendment for ornamental plants' growth.

2. Materials and methods

Analytical reagents, waste materials and earthworms

All reagents used for the different chemical determinations were of analytical grade. All experiments were carried out at INTEC (Instituto de Desarrollo Tecnológico para la Industria Química) facilities, located within CCT (Centro Científico Tecnológico) Santa Fe property (31°38'6.33"S, 60°40'13.25"O). PDW came from "El Palomar" located in Colón Plaza (31°64'68.07"S, 60°70'39.25"O) in Santa Fe city, Argentina; it was kindly provided by the local government. *Eucalyptus* sp. sawdust (ES) was obtained from a sawmill nearby (31°04'4.5"S, 60°05'47.0"O). Finally, the chipped tree pruning (CTP: mixture of leaves, foliage and small twigs) was collected from CCT Santa Fe landscape area, from where the soil was also extracted. Adult clitellated earthworms of *E. fetida*, employed for the vermicomposting stage, were obtained from INTEC terrestrial oligochaetes bioterium.

Composting and vermicomposting stages

The experimental procedure proposed was a consecutive two-stage process: first, the composting of the aforementioned wastes, followed by a vermicomposting stage. The initial moisture of PDW was 40%, while the other two wastes were dry. PDW, ES and CTP were mixed in a 7:2:1 ratio (Fig. 1) and then composted. ES and CTP were

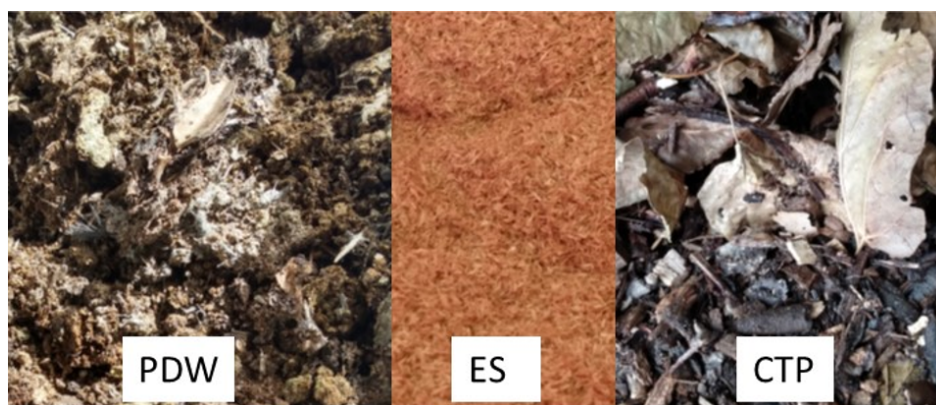


Figure 1. Original waste materials to compost: Pigeon dropping waste (PDW), *Eucalyptus* sawdust (ES) and chipped tree pruning (CTP).

selected given their OM-rich nature and high C/N ratios (e.g. C/N ratio for ES is 131.8). This makes them suitable for mixing with N-rich residues like PDW which presents extremely high content of ammonium. Additionally, CTP contributes to structural support which improves the substrate aeration; and the hygroscopic behavior of both woody materials helps retaining the moisture content of the mixture. They also help reduce the rapid release of nutrients and minimize the pathogen content of PDW.

The composting stage was set in 105 days. It was carried out during the winter-spring season, within a plastic cylindrical vessel (120 L) located outdoors but protected from rainfall and inclement weather. Moisture was kept at 60–70% by manual watering with a pump pressure sprayer and controlled with a hygrometer (TFA). The mixture was turned with a shovel once a week to aerate and homogenate. Temperature was measured in four places within the top 10 cm of the composted substrate's profile and the average value was calculated. The resulting composted material was labelled as PDW-C. Samples were periodically withdrawn for physicochemical and biological characterization.

After the first composting stage, two alternative treatments were designed to compare the vermicomposting of PDW-C. In order to favor the growth of earthworms in the vermicomposting designated area, PDW-C was combined with soil at a proportion 60% PDW-C + 40% soil (S). *E. fetida* earthworms were introduced only in PDW-C+S areas. Each of the two treatments (T1 and T2) were confined in rectangular prism containers (1 × 0.40 × 0.50 m) and subjected to the same outdoor conditions as in the composting stage. The treatments were designed as follows: **T1 (Sectorized treatment)** (28 kg): 30% PDW-C+S + 70% PDW-C (these proportions indicate that the PDW-C+S mix was set within the first 30 cm long of the container and the rest 70% was filled with PDW-C). Weekly, a portion of PDW-C was incorporated to the earthworms' zone in T1, and this continued until the earthworms distributed all along the container. **T2 (Integrated treatment)** (28 kg): 100% PDW-C+S (the complete container was filled with only PDW-C+S).

In every treatment, in PDW-C+S areas, an amount of 900 (T1) and 1500 (T2) clitellated adult *E. fetida* earthworms (mean initial biomass 0.36 ± 0.12 g/ind in wet weight) were inoculated. Containers were covered with a dark plastic net to provide shadow, minimize the loss of moisture and, given the outdoor location, to protect from the intrusion of other animals. The moisture level was maintained at 60–70% the same way as the previous composting stage. The vermicomposting stage lasted 78 days and the vermicomposts obtained were labelled as VC1 (from T1) and VC2 (from T2). The biological parameters of *E. fetida* were studied in each treatment, according to ISO 11268-2 (2023). Earthworm biomass (grams per individual) was surveyed at the beginning and end of this stage, and reproductive activity was also investigated at the end of the vermicomposting, determining reproductive status (number of clitellated individuals) and cocoon and juvenile production.

Physicochemical characterization and pathogens survey

At the beginning and end of every stage, several physicochemical parameters were surveyed. Electrical conductivity (EC) and pH were measured in a 1:10 water suspension of the dried materials (HACH®HQd multiparameter). Organic matter (OM) was obtained by calcination at 550 °C of the dried samples for 2 h. Water soluble carbon (WSC) was quantified by colorimetric method, after wet digestion of a portion of the 1:10 extract (Tognetti et al., 2005). The content of ammonium (N-NH_4^+) was measured periodically throughout both processes by the indophenol-blue method (Laos et al., 2002) with a commercial kit (Wiener®). Total organic nitrogen ($\text{TN}_{\text{Kjeldahl}}$) was quantified by the Kjeldahl digestion-distillation method (Bremner, 1960). Extractable phosphorus (P) was determined by the molybdenum blue method (Murphy and Riley, 1962) on a 0.5 M NaHCO_3 (pH 8.4) extract (Tognetti et al., 2005). Potassium (K) was measured by flame atomic absorption spectroscopy on a 1 M ammonium acetate pH 7 extract (Dionisi et al., 2020). The presence of pathogens (fecal coliforms, *Salmonella* sp. and viable helminth ova) was investigated in PDW, its initial mixture, PWD-C and the two final vermicomposts, following the methods in (USEPA, 2003).

Phytotoxicity analysis

Germination bioassays were carried out on both obtained vermicomposts. The bioassay followed USEPA protocol (USEPA, 1996) after an aqueous extraction of the samples. The extracts were obtained by agitating 4 g of sample with 40 mL of distilled water for 1 h, followed by 1 h to settle, at room temperature. Pure extract (100%) and two different dilutions were evaluated (25 and 50%). Twenty *L. sativa*, *L.* seeds were sown over germination paper that was placed at the bottom of a Petri dish embedded with 4 mL of each of the three solutions. Five replicates per treatment were run and a distilled water control was added. After five days of incubation at 24 °C, in the dark, the number of germinated seeds were counted and the length of their roots measured. A Germination Index (*GI*) was calculated as:

$$GI(\%) = \left[\left(\frac{G_s}{G_c} \right) \times \left(\frac{RL}{RL_c} \right) \right] \times 100 \quad (1)$$

where G_s and G_c are the amount of germinated seeds in each treatment and in the control, and RL and RL_c are the average root length in each treatment and in the control, respectively.

Statistical analysis

Physicochemical and earthworm biological parameters, as well as *GI*, are expressed as mean values \pm standard deviation (SD) when corresponding. One-way ANOVA was used to analyze the significant differences (with 95% confidence, $p = 0.05$) between treatments (PDW-C, VC1, VC2) and physicochemical parameters, and the differences between means were compared post hoc using Tukey's test. The significant difference between earthworm biomass during vermicomposting was analyzed using t-test for independent

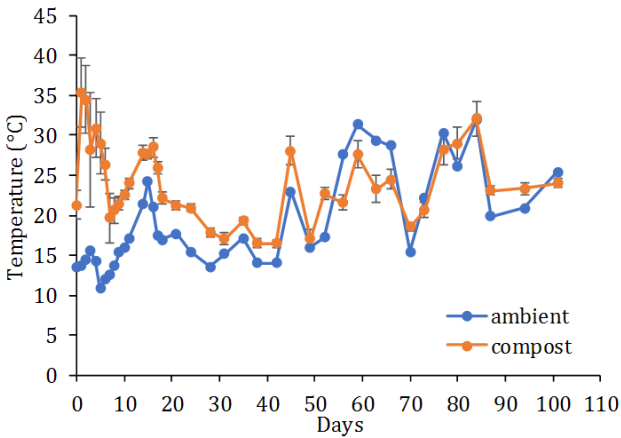


Figure 2. Temperature variation during the composting stage. Error bars correspond to SD.

samples at 95% confidence level. The software used was IBM SPSS Statistics version 25.

3. Results and discussion

Temperature profile during composting

During the composting of the PDW mixture, its temperature was registered and the profile is depicted in Fig. 2. This process was characterized by a prolonged mesophilic stage (10 – 45 °C). The microorganisms developed during this stage have the potential to act in the aerobic oxidation of the organic matter and the biodegradation of both labile and resilient organic compounds, and the nitrification process, depending the type of microorganisms present (Onwosi et al., 2017; Nobili et al., 2022). The lack of a thermophilic phase requires longer composting times to achieve stabilization.

Physicochemical characterization and ammonium profile

The main physicochemical characteristics of PDW, its initial mixture and the products of composting and vermicompost-

ing in T1 and T2 are displayed in Table 1. First, it can be observed that the presence of the earthworms accelerated the organic matter decomposition. During the composting stage, OM was only reduced by 17%, while during vermicomposting, this reduction reached 55.2% in VC1 and 88.9% in VC2 ($p < 0.001$), in comparison to the previous stage. This significant reduction can be related directly to the use of this matter by the earthworms as a source of energy (Singh et al., 2019). Giving these values, VC2 would be considered as a substrate of lesser quality, according to national regulations (SCyMA and SENASA, 2019), as well as international standards (AFNOR, 2005), which recommend $OM \geq 20\%$ for compost and organic amendments, while VC1 is, in addition, fairly closed to the value endorsed by the BOE (BOE, 2013) for vermicomposts as organic amendments ($OM > 30\%$). Even though during composting WSC decreased 42% in relation to PDW, suitable low concentrations were only accomplished after the vermicomposting stage, with both T1 and T2 ($p < 0.001$), with WSC concentrations below 10 g/kg that confirm stability of the products (SCyMA and SENASA, 2019). CO_2 production rate limits, established by the former national guideline ($< 120 \text{ mg } CO_2 \text{ kg}^{-1} \text{ h}^{-1}$) and international standards ($< 2 \text{ mg } CO_2 \text{ gOM}^{-1} \text{ day}^{-1}$) (CCQC, 2001), were achieved at the end of the composting stage. It is worth mentioning that the composting process produces CO_2 and water vapor as the main by-products. However, aerobic decomposition avoids the production of methane (CH_4), a potent greenhouse gas. When organic material is left to decompose naturally in the absence of adequate oxygen, it often undergoes anaerobic decomposition, such as in landfills. The lack of oxygen leads to the growth of anaerobic microorganisms producing CO_2 and CH_4 as by-products. Without composting, organic material emits both gases, with methane being a much more harmful greenhouse gas (Thomson et al., 2022). A recent study highlights that composting processes, particularly when optimized, can effectively reduce CO_2 emissions (Woods et al., 2024). During the composting of the PDW mixture, pH increased up to 9.2 and then decreased to 7.9 at the beginning of the

Table 1. Physicochemical characteristics of PDW, its mixture at the beginning and end of composting and the two vermicomposts (VC1 and VC2).

	PDW	Initial mixture of PDW	PDW-C	VC1	VC2
OM (%)	69.2 ^(4.8)	76.1 ^(2.7)	62.9 ^(4.1)	28.2 ^(1.2)	7.0 ^(1.5)
WSC (g C/kg)	28.9 ^(3.5)	25.9 ^(6.3)	17.0 ^(0.1)	5.7 ^(0.2)	2.1 ^(0.1)
NH_4^+ -N (mg/kg)	8693 ⁽²⁷⁶⁾	7127 ⁽⁷⁸⁶⁾	4616 ⁽¹⁴⁴⁾	29 ⁽²⁾	16 ⁽¹⁾
pH	7.5 ^(0.1)	7.4 ^(0.1)	7.9 ^(0.1)	7.3 ^(0.1)	7.1 ^(0.1)
EC (dS/m)	4.55 ^(0.08)	2.14 ^(0.03)	2.88 ^(0.04)	1.39 ^(0.05)	0.49 ^(0.04)
CO_2 production rate ($\text{mg } CO_2 \text{ kg}^{-1} \text{ h}^{-1}$)	348.2 ^(66.3)	287.9 ^(33.9)	34.4 ^(5.2)	29.7 ^(2.8)	5.7 ^(1.7)
Extractable P (g/kg)	–	–	–	2.60 ^(0.18)	0.91 ^(0.12)
K (g/kg)	–	–	–	5.97 ^(0.08)	3.26 ^(0.10)
$TN_{Kjeldahl}$ (g N/kg)	–	–	–	7.4 ^(0.1)	3.1 ^(0.1)
C/N	–	–	–	21.1	12.4

Standard deviation values are reported between parentheses.

vermicomposting stage (Fig. S1 in Supplementary material). The same trend was observed by other authors when composting chicken manure and sawdust, who related this variation to microbial nitrification, OM decomposition and CO₂ production (Gao et al., 2010). During this second process, pH values tended to neutrality, with small variations; this is usually related to the formation of humic compounds which possess buffering properties and stabilize the pH in optimum values (Villar et al., 2017). Earthworms' action is also responsible for this buffering effect on the vermicomposted substrates through the transformation of the organic wastes by the microorganisms within their guts and also through their intestinal secretions (Nobili et al., 2024; Singh et al., 2019). In the same way, the electrical conductivity of PDW was drastically reduced by 47%, only by mixing it with ES and CTP (Table 1). The composted residue PDW-C exhibited a value slightly higher, near below the maximum limit recommended for the safe application of these substrates in soil (< 3 dS/m) (Villar et al., 2017). This increase can be attributed to the degradation of organic matter and the release of soluble salts (Sharma et al., 2022). At the end of vermicomposting, EC showed another significant decrease, especially for VC2 ($p < 0.001$), remaining within the acceptable range for the growth of sensitive plants. Reduction of EC during vermicomposting has also been obtained by other authors and related to the decrease of soluble ions by leaching, immobilization by microorganisms and earthworms or by precipitation as insoluble salts (Domínguez et al., 2018; Masin et al., 2020). Conversely, the pigeon dropping-cow manure vermicomposts obtained by Singh et al. (2019) increased EC, clearly influenced by the manure, as observed from the increasing proportion in their mixtures. This shows the importance of the initial mixture in the products obtained.

The main nutrients present in the two vermicomposts are also reported in Table 1. Extractable P was almost three times higher in VC1. This value was higher than total P obtained by vermicomposting pigeon dropping and manure (Singh et al., 2019). It was also in the same order as extractable P obtained for other composts from poultry manure combined with rice hulls and sawdust (Leconte et al., 2009). A similar content of K was reported for other vermicomposts obtained from poultry litter (Masin et al., 2020) and other organic wastes (Tognetti et al., 2005). However, it was one order of magnitude lower than that from the vermicompost of pigeon dropping and manure obtained by Singh et al. (2019). In this sense, it has been reported that the amount of cattle dung incorporated into the mixture has major influence on the content of K present in the final product (Yuvaraj et al., 2021); hence, a more adequate comparison should exclude this waste. Nitrogen content (TN_{Kjeldahl}) in VC2 was similar to that of the vermicomposts obtained from pigeon and manure (Singh et al., 2019), while VC1 content was slightly higher. C/N ratio of both vermicomposts complies with the California Compost Quality Council (CCQC) maturity index standards ($C/N < 25$) (CCQC, 2001) and national regulations ($C/N \leq 20 - 30$) (SCyMA and SENASA, 2019).

Another beneficial contribution of vermicomposting was the

marked reduction in the content of ammonium at the end of the processes. Birds' droppings, and especially pigeon's ones, contain high quantities of NH₄⁺, and possess a very distinctive strong odor. Variations of this compound during the composting stage are displayed in Fig. 3 (a). The mixing of PDW with the other two wastes caused an attenuation of 17% in its content. The amount of NH₄⁺ during the first two months of composting was such that it doubled the initial already high concentration. This peak at the beginning of the composting process has also been reported by other authors (Leconte et al., 2009; Gao et al., 2010; Gong et al., 2018). Initial increase in ammonium concentration can be explained by the decomposition of nitrogen-containing organic matter during ammonification (Gao et al., 2022). Moreover, both temperature and pH affect nitrification processes and NH₄⁺-NH₃ equilibrium (Song et al., 2021). Due to the absence of a thermophilic phase, temperature might not have been enough to promote a strong volatilization of NH₄⁺ into NH₃; consequently, the conversion progressed slowly. Besides, the increase of ammonia content has been directly related to the increase of pH, which was also observed in this case (Fig. S1 in Supplementary material) and may denote the reduction of ammonium. As pH alters the NH₄⁺ oxidation and NH₃ volatilization, this high NH₄⁺ content may also engender a toxic effect on nitrifying microorganisms, reducing the efficiency of the nitrification process (Manu et al., 2021). As a result of composting PDW mixture, a 35% reduction was achieved in relation to its initial values. As it remained too high according to regulations to be considered a mature compost (< 400 mg N-NH₄⁺ kg⁻¹) (Bernal et al., 2009; SCyMA and SENASA,

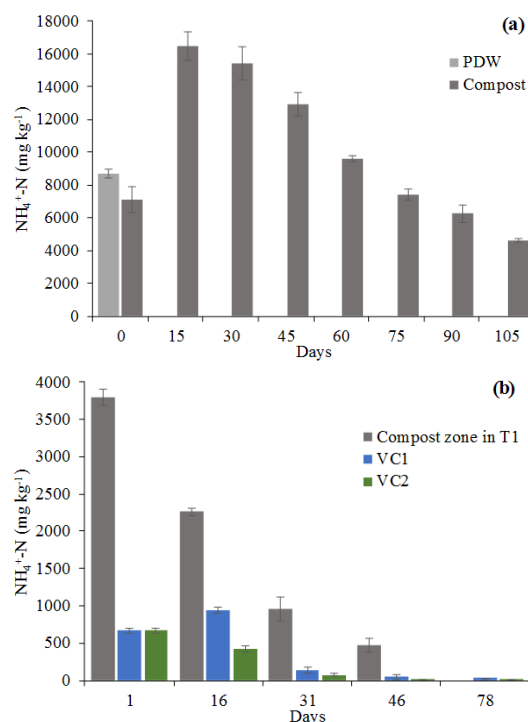


Figure 3. Ammonium content in PDW and evolution during its (a) composting stage and (b) the two vermicomposting treatments and composting zone in T1.

Error bars correspond to SD.

2019), the vermicomposting stage was evidently necessary. Fig. 3 (b) shows the parallel monitoring of both vermicompost treatments T1 and T2, and the composting zone in T1, before being incorporated into the earthworms' zone. During this stage, a very significant decrease of more than 99% ($p < 0.001$) was accomplished, when compared to the original PDW; therefore, the greatest reduction was recorded during vermicomposting, which suggests a high degree of mineralization. After 46 days, all the compost was integrated and a separate measurement for the compost zone in T1 was not possible. According to the CCQC maturity index standards (CCQC, 2001), both vermicomposts are considered as very mature regarding the N-NH_4^+ content ($< 75 \text{ mg/kg}$).

Earthworms biomass and population changes

Mean *E. fetida* biomass changes are shown in Fig. 4. At the end of the vermicomposting stage, a significant variation was observed among treatments. Mean biomass increased 25% in T1 while decreased 17% in T2, in comparison to initial biomass, but only the latter was statistically significant ($p < 0.05$). The increase in T1 was probably due to the quality of the substrate and a better utilization of the organic carbon present in it, affecting the weight of the earthworms (Sharma and Garg, 2017a, 2017b). According to Jayakumar et al. (2022) and Nagannawar et al. (2021), the abundance and quality of easily metabolizable nutrients and the microbial composition in the substrate and associated enzymes, which help breaking down complex organic matter, affect the vital activities of the earthworms. This was reflected on the population in T1, with 96% of the individuals clitellated, while in T2 only 6% of the population presented clitellum, at the end of the assay (78 days of vermicomposting). Likewise, reproductive activity was more active in T1; 85% of the total cocoons and 76% of the total juveniles were registered in this substrate. Another notorious effect in population dynamics in T1 was the fast processing and integration of PDW-C to the PDW-C+S mixture, given a visually homogeneous substrate, with the absence of odors and a fine structure. The excavation, ingestion, trituration, digestion and excretion actions of the earthworms not only stimulate, accelerate and diversify the aerobic microbial activity, but also generate higher porosity in the solid which benefits the hydraulic conductivity of the substrate where they reside (Gutiérrez et al., 2023; Ma et al., 2024).

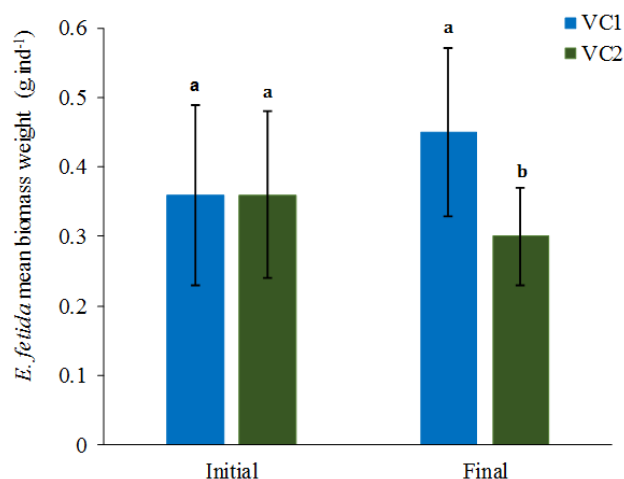


Figure 4. *E. fetida* biomass weight variation during the vermicomposting stage in T1 and T2 treatments. Significant differences between treatments ($p < 0.05$) are designated by different lowercase letters. Error bars correspond to SD.

Pathogens' survey

Table 2 presents the pathogen's count during the assay. At the end of the composting stage, pathogenic microorganisms were reduced more than 90% in comparison to the original PDW and 82.5% when compared to the initial mixture. The presence of coliforms and *Salmonella* sp. was still detected after 105 days of composting; this could be related to the absence of a marked thermophilic phase, given that during composting, the temperature remained in the mesophilic range ($\leq 45^\circ\text{C}$) and the ambient temperature were low ($< 15^\circ\text{C}$) coincident with the first week of composting. No viable helminth ova were detected. Only after vermicomposting PDW-C, the limits established by both national and international regulations (USEPA, 1993; SCyMA and SENASA, 2019) were accomplished by both VC1 and VC2. According to Swati and Hait (2018), plausible mechanisms for elimination of pathogens in organic waste subject to vermi-stabilization may be associated with intestinal activities of earthworms, such as mechanical grinding, enzymatic systems and excretion of coelomic fluids with antibacterial substances; the authors also highlight that the deposition of casts in soil greatly affects microbial communities and favors those that outcompete pathogens.

Phytotoxicity tests

At the end of the vermicomposting stage, phytotoxicity of VC1 and VC2 substrates were assayed. Seeds germina-

Table 2. Pathogens present in PDW, its mixture at the beginning and end of composting and the two vermicomposts (VC1 and VC2).

	PDW	Initial mixture of PDW	PDW-C	VC1	VC2
Fecal coliforms (MPN 100 mL ⁻¹)	16000	9000	1400	500	300
Salmonella sp. (CFU mL ⁻¹)	1620	720	120	ND	20
Viable helminth ova (< 1 viable ovum 4 g ⁻¹ dw)	ND	ND	ND	–	–

MPN: Most Probable Number. CFU: Colony Formation Units. ND: Not Detected. dw: dry weight.

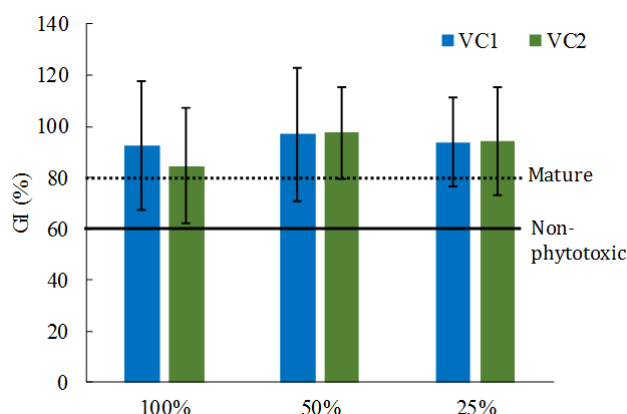


Figure 5. Germination index (*GI*) of VC1 and VC2 extracts (100%) and at two different dilutions (25 and 50%). Values above the full black line denote non-phytotoxicity and above the dotted line indicate also a mature vermicompost. Error bars correspond to SD.

tion and root length are two of the simplest environmental biomonitoring indicators for phytotoxicity evaluation (Lyu et al., 2018), as the germinated seed reflects the effect of contaminants present at the first material exchange interface between the developing plant and the environment (Oliveira et al., 2018). The relation of these two parameters, determined by the *GI*, establishes that values $\geq 60\%$ indicate non-phytotoxic substrates (Zucconi et al., 1985), while values $\geq 80\%$ indicate mature substrates as well as absence of phytotoxicity (Sharma et al., 2022). Fig. 5 illustrates these results. With both dilutions at 25 and 50% and also with the undiluted (100%) extracts of VC1 and VC2 obtained from both treatments, non-phytotoxicity and maturity, of either final substrate, were accomplished; therefore, they are safe to apply to soil. These good *GI* values suggest the presence of some beneficial phytochemicals as well as growth-promoting substances which provide nutrition content for the seeds and have no detrimental effect (Boruah and Deka, 2023). Moreover, the very low ammonium content of VC1 and VC2 could have helped in the non-phytotoxicity of the vermicomposts. As suggested by Nobili et al. (2024), who investigated the stabilization of sludge from a wastewater treatment plant and compared the products obtained with and without the incorporation of *E. fetida*, high NH_4^+ content inhibits germination and root elongation.

4. Conclusion

The PDW-C characteristics were auspicious for the growth and reproduction of *E. fetida* earthworms, despite the extremely elevated content of ammonium at the beginning of the composting stage. This initial stage was crucial for addressing the recalcitrant nature of the residues' mixture, thereby facilitating the subsequent introduction of the earthworms. Regarding the vermicomposts obtained, the sectorized treatment produced a bioproduct (VC1) with improved physicochemical characteristics, obtained with fewer initial numbers of *E. fetida* individuals and the scale of the experimental design and the off-laboratory outdoor location, allowed a more realistic evaluation of the processes' viability to provide a productive use to pigeon wastes. Although the composting stage was long, it could

be shortened by the use of larger volumes of waste. This would allow better conservation of the heat generated, thus achieving a well-defined thermophilic stage. The combined composting-vermicomposting process applied to PDW emerges as a successful, sustainable and viable alternative for safe recycling of a complex waste and the production of a pathogen limits complied and non-phytotoxic bioproduct which can be used as amendment for ornamental plants for the same recreational plaza or as a resource for the city' landscaping. This work reinforces the positive collaboration to provide a simple solution to an existing problem of local waste management. Also, it presents an economic alternative for disposing of the waste from the growing populations of this species in cities that generate worldwide concern.

Acknowledgment

Authors would like to thank for the financial support to Agencia Nacional de Promoción Científica y Tecnológica (ANPCyT), Argentina [grant number PICT 2019-00916], Universidad Nacional del Litoral (UNL), Argentina [grant number CAI+D 2020-50620190100087LI] and Consejo Nacional de Investigaciones Científicas y Técnicas (CONICET), Argentina [grant number PIP 347]. We also acknowledge Santa Fe city government for providing the PDW.

Authors contributions

The authors confirm the study conception and design: CE Masin, CS Zalazar and ME Fernandez; data collection: CE Masin, A Duran and ME Fernandez; analysis and interpretation of results: All authors; draft manuscript preparation: ME Fernandez; and founding acquisition: CS Zalazar. The results were evaluated by all authors, and the final version of the manuscript was approved.

Availability of data and materials

The data that support the findings of this study are available from the corresponding author, upon reasonable request.

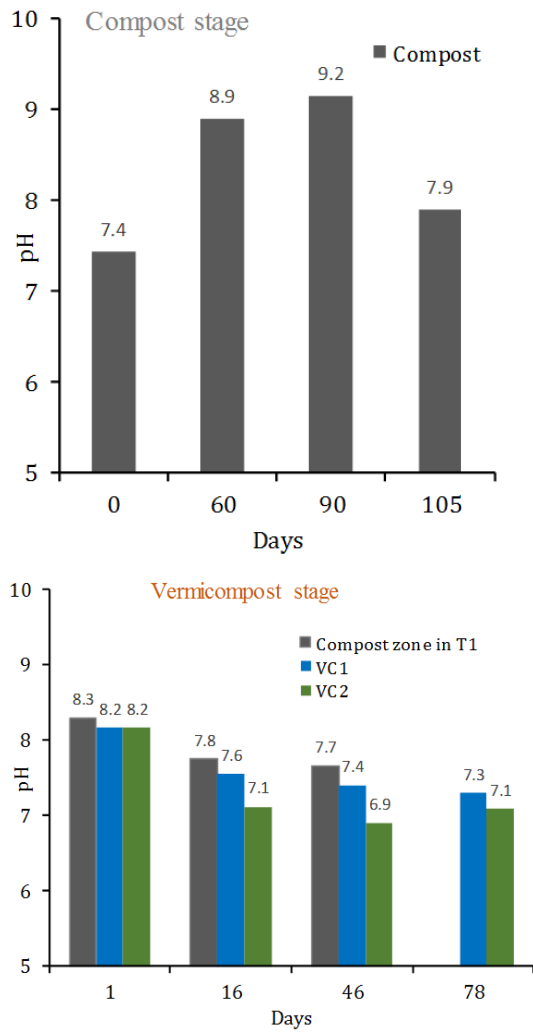
Conflict of interests

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

References

- AFNOR (2005) Association française de normalisation; Norme Française U44-051. Amendements organiques. Dénominations, spécifications et marquage. *Cedex, France*
- Bernal MP, Albuquerque JA, Moral R (2009) Composting of animal manures and chemical criteria for compost maturity assessment: A review. *Bioresour Technol* 100:5444–5453. DOI: <https://doi.org/10.1016/j.biortech.2008.11.027>.
- BOE (2013) Boletín Oficial del Estado (2013) Real Decreto 506/2013 sobre productos fertilizantes. *Official State Bulletin, Ministry of The Presidency, Spain* 164:51119–51207.
- Boruah T, Deka H (2023) Enumeration of synergistic relationship between carbon dioxide evaluation and nutrient budget during vermicomposting of cereal grain processing industry sludge. *Bioresour Technol Rep* 22:101418. DOI: <https://doi.org/10.1016/j.biteb.2023.101418>.
- Bremner JM (1960) Determination of nitrogen in soil by the Kjeldahl method. *J Agric Sci* 55:11–33. DOI: <https://doi.org/10.1017/S0021859600021572>.

- CCQC (2001) California Compost Quality Council. Compost Maturity Index, Technical Report. Nevada, CA, USA. 26.
- Dionisi CP, Mignone RA, Rubenacker AI, et al. (2020) Monitoring of physicochemical parameters of soils after applying pig slurry. Analysis of its application in short and long periods in the province of Córdoba, Argentina. *Microchem J* 159:105545. DOI: <https://doi.org/10.1016/j.microc.2020.105545>.
- Domínguez J, Gómez-Brandón M, Martínez-Cordeiro H, et al. (2018) Bioconversion of scotch broom into a high-quality organic fertilizer: Vermicomposting as a sustainable option. *Waste Manage Res* 36:1092–1099. DOI: <https://doi.org/10.1177/0734242X18797176>.
- Gao M, Li B, Yu A, et al. (2010) The effect of aeration rate on forced-aeration composting of chicken manure and sawdust. *Bioresour Technol* 101 (1): 1899–1903. DOI: <https://doi.org/10.1016/j.biortech.2009.10.027>.
- Gao Y, Zhang C, Tan L, et al. (2022) Full-Scale of a compost process using swine manure, human feces, and rice straw as feedstock. *Front Bioeng Biotechnol* 10:1–12. DOI: <https://doi.org/10.3389/fbioe.2022.928032>.
- Gong X, Cai L, Li S, et al. (2018) Bamboo biochar amendment improves the growth and reproduction of *Eisenia fetida* and the quality of green waste vermicompost. *Ecotoxicol Environ Saf* 156 (3): 197–204. DOI: <https://doi.org/10.1016/j.ecoenv.2018.03.023>.
- Gutiérrez V, Gómez G, Rodríguez DC, et al. (2023) Critical analysis of wastewater treatment using vermifilters: Operating parameters, wastewater quality, and greenhouse gas emissions. *J Environ Chem Eng* 11:109683. DOI: <https://doi.org/10.1016/j.jece.2023.109683>.
- ISO 11268-2 (2023) Soil quality-Effects of pollutants on earthworms-Part 2: Determination of effects on reproduction of *Eisenia fetida*/*Eisenia andrei* and other earthworm species. *International Organization for Standardization*, 36.
- Jayakumar M, Emana AN, Subbaiya R, et al. (2022) Detoxification of coir pith through refined vermicomposting engaging *Eudrilus eugeniae*. *Chemosphere* 291:132675. DOI: <https://doi.org/10.1016/j.chemosphere.2021.132675>.
- Laos F, Mazzarino MJ, Walter I, et al. (2002) Composting of fish offal and biosolids in northwestern Patagonia. *Bioresour Technol* 81:179–186. DOI: [https://doi.org/10.1016/S0960-8524\(01\)00150-X](https://doi.org/10.1016/S0960-8524(01)00150-X).
- Leconte MC, Mazzarino MJ, Satti P, et al. (2009) Co-composting rice hulls and/or sawdust with poultry manure in NE Argentina. *Waste Manag* 29:2446–2453. DOI: <https://doi.org/10.1016/j.wasman.2009.04.006>.
- Lyu J, Park J, Kumar Pandey L, et al. (2018) Testing the toxicity of metals, phenol, effluents, and receiving waters by root elongation in *Lactuca sativa* L. *Ecotoxicol Environ Saf* 149:225–232. DOI: <https://doi.org/10.1016/j.ecoenv.2017.11.006>.
- Ma L, Zhang L, Feng X (2024) Optimization of *Eisenia fetida* stocking density for biotransformation during green waste vermicomposting. *Waste Manage* 187:188–197. DOI: <https://doi.org/10.1016/j.wasman.2024.07.016>.
- Maharjan KK, Noppradit P, Techato K (2023) Potential of *Eisenia fetida* (Redworm) for the conversion of three varieties of organic waste. *Int J Recycl Org Waste Agric* 12:341–350. DOI: <https://doi.org/10.30486/ijrowa.2022.1958871.1466>.
- Manu MK, Li D, Liwen L, et al. (2021) A review on nitrogen dynamics and mitigation strategies of food waste digestate composting. *Bioresour Technol* 334:125032. DOI: <https://doi.org/10.1016/j.biortech.2021.125032>.
- Masin CE, Fernandez ME, Lescano MR, Zalazar CS (2020) Bioconversion of agro-industrial wastes: Combined compost and vermicompost processes using *Eisenia fetida* for stabilization of poultry litter. *Int J Recycl Org Waste Agric* 9:107–118. DOI: <https://doi.org/10.30486/IJROWA.2020.1885790.1011>.
- Murphy J, Riley JP (1962) A modified single solution method for the determination of phosphate in natural waters. *Anal Chim Acta* 27:31–36. DOI: [https://doi.org/10.1016/S0003-2670\(00\)88444-5](https://doi.org/10.1016/S0003-2670(00)88444-5).
- Nagannawar MF, Patil SR, Biradar PM (2021) Growth and reproduction of the epigeic earthworm, *Eisenia fetida* (Savigny, 1826) cultured in various organic wastes. *J Adv Zool* 42:43–59. DOI: <https://doi.org/10.17762/jaz.v42i01.5>.
- Niedzialkoski RK, Marostica R, Damaceno FM, et al. (2021) Combination of biological processes for agro-industrial poultry waste management: Effects on vermicomposting and anaerobic digestion. *J Environ Manage* 297:113127. DOI: <https://doi.org/10.1016/j.jenvman.2021.113127>.
- Nobili S, Masin CE, Zalazar CS, Lescano MR (2022) Bioremediation of hydrocarbon contaminated soil using local organic materials and earthworms. *Environ Pollut* 314:120169. DOI: <https://doi.org/10.1016/j.envpol.2022.120169>.
- (2024) Vermistabilization of excess sludge employing *Eisenia fetida*: Earthworm histopathological alterations and phytotoxicity evaluation. *J Environ Manage* 368:122174. DOI: <https://doi.org/10.1016/j.jenvman.2024.122174>.
- Oliveira GA Rodrigues de, Morais Leme D, Lapuente J de, et al. (2018) A test battery for assessing the ecotoxic effects of textile dyes. *Chem Biol Interact* 291:171–179. DOI: <https://doi.org/10.1016/j.cbi.2018.06.026>.
- Onwosi CO, Igbokwe VC, Odimba JN, et al. (2017) Composting technology in waste stabilization: On the methods, challenges and future prospects. *J Environ Manage* 190:140–157. DOI: <https://doi.org/10.1016/j.jenvman.2016.12.051>.
- Pizarro MD, Céccoli G, Muñoz FF, et al. (2019) Use of raw and composted poultry litter in lettuce produced under field conditions: microbiological quality and safety assessment. *Poult Sci* 98:2608–2614. DOI: <https://doi.org/10.3382/ps/pez005>.
- SCyMA, SENASA (2019) Anexo I: Marco Normativo Para La Producción, Registro y Aplicación De Compost. <https://www.argentina.gob.ar/normativa/nacional/resolucin-1-2019-318692/texto>
- Sharma D, Prasad R, Patel B, Parashar CK (2022) Biotransformation of sludges from dairy and sugarcane industries through vermicomposting using the epigeic earthworm *Eisenia fetida*. *Int J Recycl Org Waste Agric* 11:165–175. DOI: <https://doi.org/10.30486/IJROWA.2021.1922034.1196>.
- Sharma K, Garg VK (2017a) Management of food and vegetable processing waste spiked with buffalo waste using earthworms (*Eisenia fetida*). *Environ Sci Pollut Res* 24:7829–7836. DOI: <https://doi.org/10.1007/s11356-017-8438-2>.
- (2017b) Vermi-modification of ruminant excreta using *Eisenia fetida*. *Environ Sci Pollut Res* 24:19938–19945. DOI: <https://doi.org/10.1007/s11356-017-9673-2>.
- Singh S, Singh J, Kaur A, et al. (2019) Nutrient recovery from pigeon dropping by using exotic earthworm *Eisenia fetida*. *Sustain Chem Pharm* 12:100126. DOI: <https://doi.org/10.1016/j.scp.2019.01.003>.
- Song B, Manu MK, Li D, et al. (2021) Food waste digestate composting: Feedstock optimization with sawdust and mature compost. *Bioresour Technol* 341:125759. DOI: <https://doi.org/10.1016/j.biortech.2021.125759>.
- Srivastava PK, Singh A, Kumari S, et al. (2023) Production and characterization of sustainable vermimanure derived from poultry litter and rice straw using tiger worm *Eisenia fetida*. *Bioresour Technol* 369:128377. DOI: <https://doi.org/10.1016/j.biortech.2022.128377>.
- Swati A, Hait S (2018) A Comprehensive review of the fate of pathogens during vermicomposting of organic wastes. *J Environ Qual* 47:16–29. DOI: <https://doi.org/10.2134/jeq2017.07.0265>.
- Thomson A, Price GW, Arnold P, et al. (2022) Review of the potential for recycling CO₂ from organic waste composting into plant production under controlled environment agriculture. *J Cleaner Prod* 333:130051. DOI: <https://doi.org/10.1016/j.jclepro.2021.130051>.



Supplementary Material
Fig. S1. The pH evolution during composting and vermicomposting of PDW mixture.

Tognetti C, Laos F, Mazzarino MJ, Hernández MT (2005) Composting vs. vermicomposting: A comparison of end product quality. *Compost Sci Util* 13:6–13.
DOI: <https://doi.org/10.1080/1065657X.2005.10702212>.

USEPA (1993) 40 CFR Part 257, 403 and 503 Standards for Use or Disposal of Sewage Sludge. US Government Printing Office Fed Regist. 58:9248–9415.

—— (1996) Ecological Effects Test Guidelines: Seed Germination/Root Elongation Toxicity Test. *United States Environmental Protection Agency*, 8.

—— (2003) EPA/625/R-92/013. Control of Pathogens and Vector Attraction in Sewage Sludge. *United States Environmental Protection Agency*, 177.

Villa-Serrano AM, Perez-Murcia MD, Perez-Espinosa A, et al. (2010) Characterization and agronomic use of pigeon manure: a case study in the Northeast Transmontano Region (Portugal). *14th Ramiran International Conference: Treatment and use of organic residues in agriculture: Challenges and opportunities towards sustainable management*

Villar I, Alves D, Mato S (2017) Product quality and microbial dynamics during vermicomposting and maturation of compost from pig manure. *Waste Manage* 69:498–507.
DOI: <https://doi.org/10.1016/j.wasman.2017.08.031>.

Woods E, Rondon Berrio V, Qiu Y, et al. (2024) Biomass composting with gaseous carbon dioxide capture. *RSC Sustainability* 2:79–101.
DOI: <https://doi.org/10.1039/d3su00411b>.

Yuvaraj A, Thangaraj R, Ravindran B, et al. (2021) Centrality of cattle solid wastes in vermicomposting technology – A cleaner resource recovery and biowaste recycling option for agricultural and environmental sustainability. *Environ Pollut* 268 (20): 115688.
DOI: <https://doi.org/10.1016/j.envpol.2020.115688>.

Zucconi F, Monaco A, Forte M, Bertoldi MD (1985) Phytotoxins during the stabilization of organic matter. In: *Composting of agricultural and other wastes. Elsevier Applied Science Publication*, 73–86.