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Incorporation of Substrates and Inoculums as Operational Strategies to Promote Lignocellulose Degradation in Composting of Green Waste—A Pilot-Scale Study

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Abstract: Composting is a sustainable alternative for green waste (GW) valorization contributing to the circular bioeconomy. However, the processing time must be reduced and the end-product quality must be improved. This study determined the effect of the incorporation of processed food waste (PFW), unprocessed food (UPFW), sawdust (SW), phosphate rock (PR) and a specific bacterial inoculum on GW-composting process parameters and product quality. Three treatments were evaluated in 120 kg piles: (i) TA: (GW + UPFW + PFW + inoculum), (ii) TB (GW + UPFW + PFW), and (iii) TC (GW). An inoculum of *Bacillus* sp. and *Paenibacillus* sp. was incorporated in the cooling phase for TA. On the other hand, the effect of the inoculum at the laboratory scale (20 kg reactors) was compared with that found at the pilot scale (120 kg piles). The incorporation of FW, SW, PR and the inoculum increased the amount of lignocellulose biodegradation (TA: 29.1%; TB: 22.7%; TC: 18.2%), which allowed for a reduction of up to 14 days of processing time. The product obtained for TA had a similar quality to the other two treatments, although a lower phytotoxicity was determined according to the germination index (TA: 95%; TB: 85%; and TC: 83%). The final product of TA showed the best agricultural characteristics with pH 8.3, TOC of 24.8%, TN of 1.32%, and GI of 98.8%. Finally, the scaling effect with the bacterial inoculum was shown to affect parameters such as the TOC, TN, GI, and, to a lesser extent, temperature and pH. The results obtained in this paper highlight the importance of optimizing the composting of GW, specifically with the use of co-substrates and specific inocula, which can be of interest for composting materials with a high content of lignocellulose such as GW.

Keywords: green waste; composting; bacteria; food waste; lignocellulose

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1. Introduction

The management of green waste (GW) is a challenge due to its heterogeneous composition (i.e., wood, branches, leaves, soil, and grass clippings) and predominance of lignocellulosic compounds such as cellulose (40%), hemicellulose (20–30%), and lignin (20–30%). Although composting can be used to transform GW into a product with potential agricultural value, the difficulty in degrading lignocellulose increases the processing time and reduces the product quality [1].

To optimize GW composting, various strategies have been developed to accelerate the biodegradability of lignocellulosic compounds [1,2]. The incorporation of substrates such as sawdust (SW) and phosphate rock (PR) have been used to provide porosity and phosphorus [3,4]; however, few studies have evaluated the amendment addition of food waste (FW) [5,6], which constitutes more than 70% of municipal solid waste in developing countries [7].

The incorporation of microbial inoculants during the cooling phase is a strategy that represents a key operational change to reduce processing time [8]. However, some studies

have reported that there is no significant effect associated with the use of microbial inoculants due to the potential competition between the exogenous species and the indigenous microbial species of the process [9]. Therefore, further studies are needed to evaluate the synergistic or antagonistic effects of microbial inoculation on the GW-composting process and end-product quality [8]. Bacterial strains from the genus *Bacillus* have been used as inoculants for GW composting due to their capacity to degrade lignocellulose and the formation of humic substances [10]. In contrast, bacteria from the genus *Paenibacillus* have been little studied in the composting of GW with other substrates despite their potential to secrete specialized enzymes for lignocellulose degradation [11].

Additionally, the scaling up of the composting process is considered fundamental for the optimization of the process and the evaluation of its applicability on a large scale [8]. Previously, the effects of inoculation in the cooling phase with *Bacillus* sp. and *Paenibacillus* sp. were preliminarily evaluated at the laboratory scale (500 mL reactors) [12] and in reactors with a capacity of 20 kg for GW and FW co-composting amended with SW and PR [10]. In the present study, the effect of inoculation with *Bacillus* sp. and *Paenibacillus* sp. on GW and FW co-composting was scaled up to a pilot scale of 120 kg composting piles. This study contributes to the search for options to improve GW-composting implementation for urban GW management, which can be implemented at the full scale. Furthermore, this study provides elements for the recovery of waste to generate valuable products within the framework of the circular bioeconomy.

2. Materials and Methods

2.1. Substrates and Composting Process

GW was collected from the campus of Universidad Industrial de Santander (Colombia). FW was obtained from a marketplace where PFW and UPFW were separated at the source. SW and PR were purchased from a commercial establishment. SW provided carbon, porosity, and adjusted moisture content [13], while PR provided phosphorus and porosity [3]. The substrates were manually mixed in the following proportions: 50% GW, 32.5% UFW, 2.5% PFW, 13% SW, and 2% PR. The substrate mixture was defined from an experiment carried out at the laboratory scale in 0.5 L reactors where the substrate mixture and the concentration of the bacterial strains used as the inoculum were optimized [12]. In addition, the mixture guarantees a C:N ratio greater than 25, a value recommended for the composting of green waste [14]. The experiments were carried out at the pilot scale in conical piles of 120 kg. We believe that this study can be easily interpreted and its conclusions are useful for works at the full scale. The piles were set up leveled on concrete and fenced with polyethylene shade cloth to prevent access by external agents. In treatment A (TA), a mixture of substrates plus inoculum was studied; treatment B (TB) corresponded to a mixture of substrates (uninoculated); and in treatment C (TC), only GW was processed. Each treatment had a replicate. Before starting the experiment, the GW, UFW and PFW were manually crushed to a particle size of between 30 and 50 mm [1].

The physicochemical characteristics of the substrates and co-substrates were:

- PFW: moisture of 75.5 ± 7.6%, pH of 4.9 ± 0.4, EC of 3.1 ± 0.4 mS/cm, TOC of 33.5 ± 5.9% (db), TN of 1.2 ± 0.6% (db), and lignocellulose of 17.0 ± 2.6% (db).
- UFW: moisture of 79.1 ± 8.3%, pH of 5.1 ± 0.3, EC of 3.1 ± 0.4 mS/cm, TOC of 33.5 ± 5.9% (db), TN of 1.2 ± 0.6% (db) and lignocellulose of 17.8 ± 3.3% (db).
- TA and TB (including 50% GW, 32.5% UFW, 2.5% PFW, 13% SW, and 2% PR): moisture of 58.2 ± 2.5%, pH of 6.3 ± 0.2, EC of 3.5 ± 0.4 mS/cm, TOC of 47.7 ± 3.1% (db), TN of 1.7 ± 0.3% (db) and lignocellulose of 23.8 ± 1.9% (db).
- TC (100% GW): moisture of 27.3 ± 4.9%, pH of 6.9 ± 0.1, EC of 3.0 ± 0.3 mS/cm, TOC of 26.6 ± 5.8% (db), TN of 11.2 ± 0.5% (db) and lignocellulose of 35.1 ± 6.1% (db).

The bacterial inoculum consisted of *Bacillus* sp. and *Paenibacillus* sp. with concentrations of 4.85×10^5 CFU mL^{-1} and 1.44×10^5 CFU mL^{-1}, respectively. The strains were individually cultivated in a Luria–Bertani medium at 37 °C and 200 rpm, sequentially scaling up in reactors with a capacity of 0.02, 0.06, 0.45, 1, and 7.5 L. The characteristics

of the bioreactor for inoculum scaling are presented in more detail in the work of Oviedo et al. [10]. The inoculum was added to the piles at the start of the cooling phase (i.e., when the temperature was close to 45 °C).

Manual turns were applied depending on the temperature in the piles to maintain the aeration of the piles. When the temperature in the pile remained constant for three days, turning was applied. Furthermore, weekly humidification was applied to maintain a moisture content of between 40% and 60% [4] and stimulate biological activity [15]. The temperature was monitored daily using a digital thermometer with a resolution of 0.1 °C at five characteristic points of the reactors (i.e., centroid and four opposite points of the pile). Manual turnings were performed depending on the temperature reached. The monitoring was performed until all treatments reached ambient temperature.

On the other hand, data from a previous study by Oviedo et al. [10] that was carried out with the same treatments in 20 kg reactors were collected. The reactor configuration is presented in greater detail in the work of Rawoteea et al. [16]. With the information collected in this study on the laboratory scale, it was determined whether there was a scaling effect with the incorporation of the bacterial inoculum.

2.2. Analytical Methods

Five sub-samples of 300 g were obtained from the center and perimetral points of each pile at 30 cm of depth; the combined sample was used for laboratory analysis. The moisture content was gravimetrically measured at 70 °C in an oven until a constant weight was reached. The pH was determined using the potentiometric method (sensIONTM pH meter + MM374) in a suspension of 10 g of the processed composting material in 50 mL of distilled water (i.e., ratio of 1:10, w/v). Electrical conductivity (EC) was determined with the potentiometric method. The Total Kjeldahl Nitrogen (TN) was measured through titration according to Colombian Technical Standard NTC 5167. The ash content was determined with gravimetry at 550 °C, and the total organic carbon (TOC) was estimated from this value [17]. The organic matter losses were determined according to the work of Jiang et al. [18]. The stability of the product was determined with the Rottegrade self-heating test [19] once the process was finished. The germination index (GI) was determined using *Raphanus sativus* seeds, taking one gram of solid sample and diluting it with distilled water in a 1:10 (p/v) ratio [20]. As a maturity criterion, a GI of greater than 80% was adopted.

All experiments were carried out in triplicate; data were subjected to an analysis of variance (ANOVA), and significantly different means were evaluated using the least significant difference (LSD) at a significance level of α = 0.05. Statistical analysis was performed in SPSS® Version 16.0 (Statistical Package for the Social Science, SPSS, Inc., Chicago, IL, USA).

2.3. Lignocellulose Degradation

The concentration of lignocellulose was determined according to the protocol of the National Renewable Energy Laboratory (NREL) with Soxhlet equipment (Model B-324, Buchi, Spain) considering the moisture, ash content at 550 °C and aqueous and organic extractives of the sample [21]. Additionally, the percentage of lignocellulose degradation was quantified once a week according to Equation (1).

$$\%Lignocellulose\ degradation = 1 - \frac{L_f}{L_i} * 100\% \quad (1)$$

where L_i refers to the lignocellulose content at the beginning of the experiment and L_f is the lignocellulose content at the time of measurement.

2.4. Product Quality

The end-product was manually sieved with a 1.25 cm mesh. An integrated sample of 4 kg was obtained for each treatment by combining sub-samples taken at the perimeter and centroid points of each pile. Quality parameters such as the pH, EC, TOC, TN, stability, and

GI were determined in triplicate. The results obtained in this study were compared with those obtained from a previous experiment with the same treatments but at the laboratory scale (20 kg reactors) [10].

3. Results

3.1. Physicochemical Changes during the Process

The temperature profiles of each treatment during the process are shown in Figure 1. The mesophilic phase in TA and TB was present until day 1 of the process, and it lasted until day 4 for TC. These results were similar to those previously indicated in the composting of UPFW, FW, and GW [22]. The longer time in TC was caused by the greater presence of slow-biodegrading organic carbon [1,23].

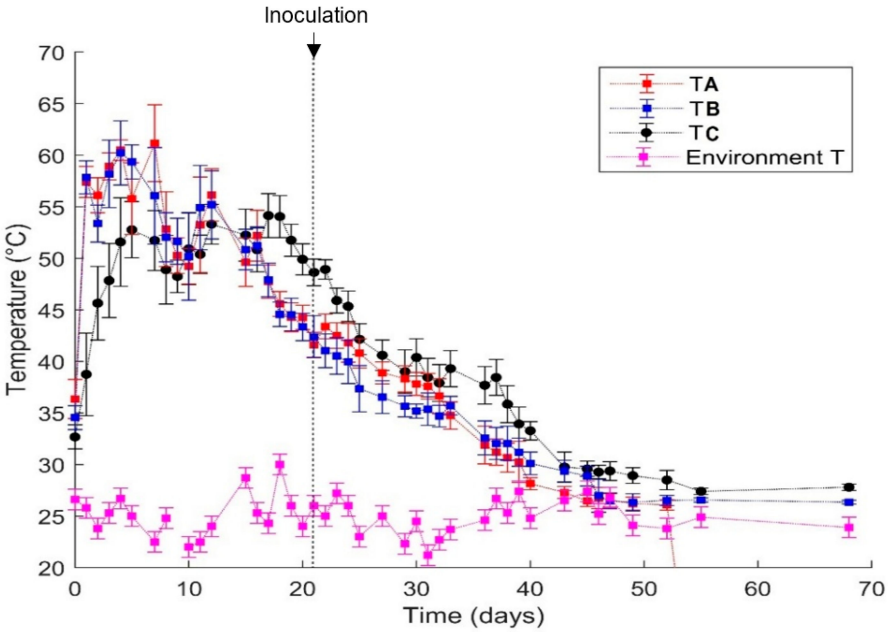

Figure 1. Temperature profiles during the composting process.

In the thermophilic phase, TA reached the highest temperature (64 °C) after 8 days of processing. In contrast, in TB, the highest temperature of 60.7 °C was reached at 9 days, showing no significant differences with TC. Regarding the sanitation of the process, if the temperature was maintained above 55 °C for three days, the material would be free of pathogens and weeds [22]. This condition was achieved with all treatments. The results indicated that the incorporation of FW, SW, and PR in TA and TB had a significant ($p < 0.05$) and synergistic effect on GW composting [13]. This resulted in an advantage compared with what has been documented in the composting of just GW due to the biological stimulus of microorganisms that results in the greater degradation of OM and therefore an increase in temperature. For TC, no temperature differences were observed with respect to what has been documented in the literature. This may have been associated with the predominance of lignocellulose, which is difficult to biologically degrade and therefore limits temperature increases. During the cooling phase, the incorporation of bacterial inocula can affect bacterial communities, as shown in another study [15]. The incorporation of the bacterial inoculum in TA increased the temperature (i.e., Δ 5 °C) between days 19 and 27 of the process. This evidenced that the inoculum stimulated the degradation of organic matter and lignocellulose, possibly due to the secretion of lignocellulolytic enzymes [24]. Similar results were reported with the inoculation with ammonifying bacteria, nitrobacteria, and *Azotobacter* in pig manure composting [24]. Then, the temperature dropped to 27 ± 3 °C on day 38 of the process. In contrast, TB reached this condition on day 43 and TC required 47 days. Therefore, the inoculum reduced the duration of the cooling phase by 13% and

23% compared with TB and TC, respectively. In addition, the duration of the cooling phase between TA and TC (p = 0.0205) showed significant differences.

The results found at the pilot scale showed trends similar to those found in the study of Oviedo et al. on 20 kg reactors [10]. In both cases, the inoculation increased the temperature gradients during the thermophilic phase (3–5 °C). However, the duration of both peaks differed between the scales (i.e., 10 and 6 days for the pilot and laboratory scale, respectively). These differences were probably caused by the mass of the material in the process and the heat-diffusion transport phenomena that prolonged the duration of the temperature peak at the pilot scale. In addition, the possible influence of exogenous microorganisms that could benefit from the incorporation of the inoculum is not ruled out.

Regarding the sanitation product, at both scales, the incorporation of the inoculum did not generate an antagonistic effect on the quality of the final product. Likewise, there was no significant reduction in the processing time with respect to the treatment that only included co-substrates ($p > 0.05$).

Regarding pH, TA and TB started with slightly acidic pH values (i.e., 5.8 and 6.3 units, respectively), as shown in Figure 2. During the thermophilic phase, the pH in all treatments increased to values higher than 8.0, probably due to ammonia volatilization as a consequence of the high temperatures reached [25]. Zhou et al. [26] reported similar pH profiles (7–9 units). In the cooling phase, there were no significant differences between treatments. At the end of the process, the lowest pH value was found in TA, which may have been associated with the nitrification process [26] and the synthesis of phenolic compounds [27]. A similar result was found in a study of the composting of straw residues, when an inoculum consisting of *Aeromonas caviae* sp., *Shinella* sp., *Rhizobium* sp., *Corynebacterium pseudotuberculosis* sp., and *Streptomyces clavuligerus* sp. was applied at the beginning of the cooling phase, which affected the pH dynamics [28]. In contrast, TC presented the highest pH value (i.e., 8.65) at the end of the process in this study.

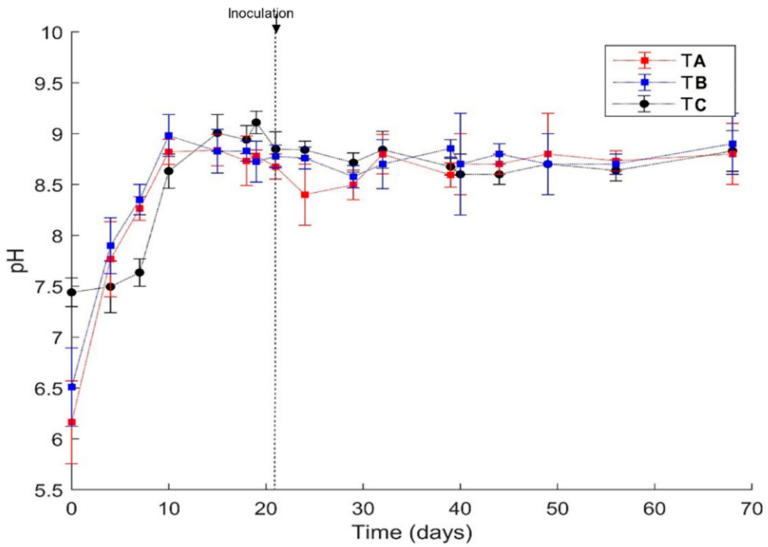

Figure 2. pH changes during the composting process.

The results found at the pilot scale were similar to those found at a smaller scale. The differences in terms of pH during the process steps were insignificant. Similarities were found in the rise of pH to alkaline values and a tendency to remain between 8 and 9 units in the cooling stage. These similarities were associated with the contribution of precedent proteins from PFW and UPFW and their mineralization during the ammonia volatilization due to the high temperatures reached [10]. In addition, the bacterial inoculum could affect the behavior of pH over time, as indicated in other investigations [8].

Figure 3 shows the dynamics of the TOC during composting. The concentration of the TOC in all treatments decreased over time. During the thermophilic stage in TA and TB,

a higher degradation of the TOC (19 ± 3%) was achieved compared with TC (11 ± 1.2%). The differences between treatments was associated with the presence of FW, PR, and AS. Bohacz et al. [29] indicated that the incorporation of co-substrates stimulates biological activity, thus allowing for the higher degradation of the TOC and higher temperatures. After inoculation, a more pronounced decrease in the TOC was observed in TA and showed significant differences with respect to TB (p = 0.038). These results were associated with the incorporation of the inoculum that could stimulate the enzymatic activity and the mineralization of organic matter [30]. At the end of the process, the TOC losses for TA were 35.7%. This value was higher than those found in TB (32.1%) and TC (20.1%), indicating a synergistic effect of the inoculum on the process.

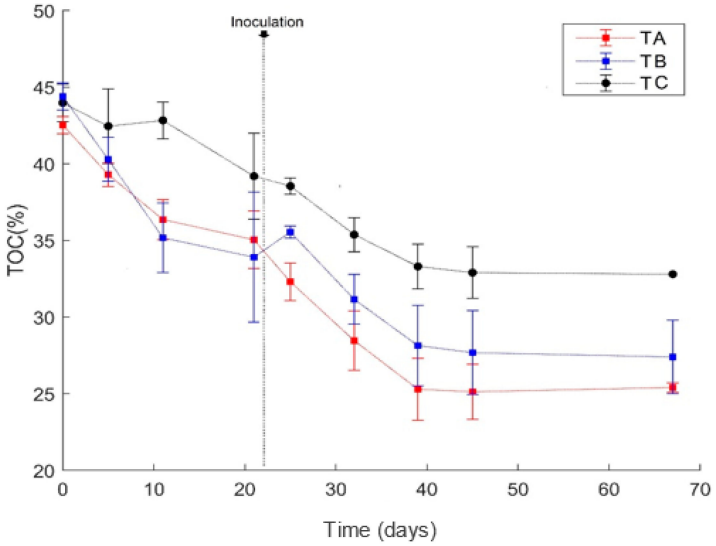

Figure 3. TOC changes during the composting process.

An effect of the inoculum was evidenced in the different evaluated scales. At the laboratory scale, there were greater TOC losses (41.1%) compared with what was found in the present pilot study (35.7%). These differences may be associated with the fact that the effect of the inoculum was enhanced in the more controlled environment and with a lower incidence of exogenous microorganisms due to the environmental and operational conditions of the laboratory-scale reactors. Additionally, oxygen diffusion was better at this scale due to the holes the presented reactors. This could have stimulated biological activity and promoted the degradation of OM. In contrast, at the pilot scale, the effect of compaction and clumping by the inoculum reduced the porosity of the material being processed and may have adversely affected mineralization.

On the other hand, the concentration of TN in all treatments increased over time (see Figure 4). The highest TN concentration was found in TC (2.4 ± 0.2%) and was associated with the availability of this element in the green waste retained by lignin [29]. This was associated with the fact the inoculum utilized the available nitrogen during the cooling phase for their metabolism and secretion of enzymes such as xylanase and cellulase [28,29]. According to Feng et al. [31], these enzymes are key to the degradation of lignocellulose. At the end of the process, the lowest concentration of 1.6 ± 0.3% was obtained in TA, and a concentration of 2.2 ± 0.2% was achieved in TB. The results indicated that the inoculum had an adverse effect on the availability of TN in the final product (i.e., possibly associated with a higher nutrient requirement due to higher biological activity).

The dynamics of nitrogen were similar between the considered scales. In both cases, the incorporation of the bacterial inoculum generated a decrease in the TN concentration, while in the other treatments (i.e., without the incorporation of the inoculum), the NT increased due to the effect of the OM concentration. The main difference was found in terms of NT concentration (i.e., 1.6 and 1.2% for the pilot and laboratory scales, respectively)

during the cooling stage. In both cases, the effect of the bacterial inoculum was antagonistic to the potential to reduce the agricultural quality of the product.

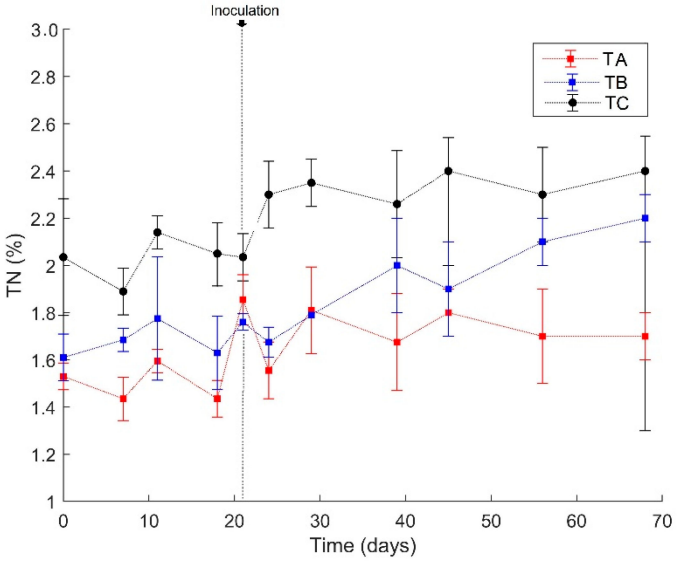

Figure 4. TN changes during the composting process.

3.2. Biodegradation of Lignocellulose

The percentage of lignocellulose in the composting material over time is shown in Figure 5. At the beginning of the process (i.e., mesophilic phase), the lignocellulose degradation was limited. This was mainly observed in TC, in which the predominance of organic compounds of difficult degradation could have limited the biological activity, thus prolonging the duration of the composting process. In contrast, in TA and TB, the biological activity was stimulated, and rapidly degrading carbon compounds were consumed, which increased the temperature and thus enhanced the hydrolysis of hemicellulose and cellulose when the temperature exceeded 60 °C [32].

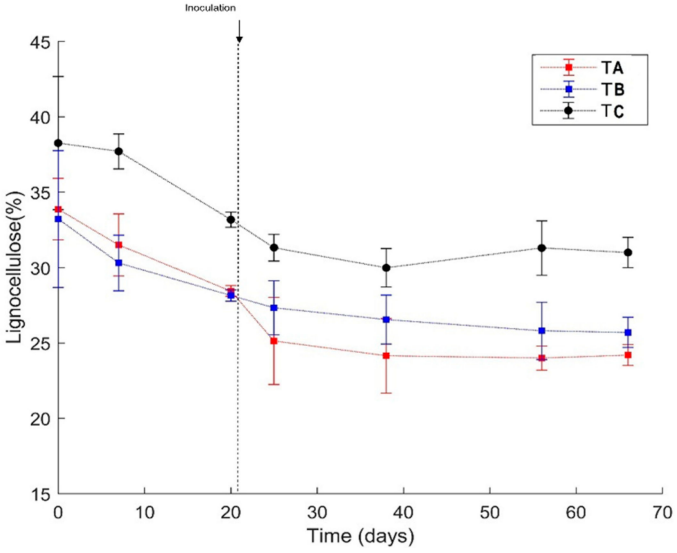

Figure 5. Percentage of lignocellulose in the composting material over time.

The lignocellulose degradation increased in all treatments after the start of the composting process. During the cooling phase, the percentage of lignocellulose degradation was 29.1%, 22.7%, and 18.2% for TA, TB, and TC, respectively. These results are consistent with those reported previously [8] due to the degradation of organic matter, the synthesis

of phenolic compounds, and the synthesis of humic substances. The higher lignocellulose degradation for TA is associated with a higher content of TN that promoted the enzymatic activity of cellulose, xylanase, and phenoloxidase, among other lignocellulolytic enzymes [32,33]. Likewise, the pH values closest to neutrality in TA could have stimulated microbial activity [33]. Furthermore, it was previously reported that delignification takes place faster under neutral pH than under acidic or alkaline pH [34]. The lignin biodegradation observed in this study was higher than that reported previously (27.81%) for GW composting inoculated with a complex, non-defined microbial inoculum [8].

On the other hand, lignocellulose biodegradation was higher at the laboratory scale (31.7%) compared with that obtained at the pilot scale (29.1%). The differences could be associated with the fact that the environmental conditions at the laboratory scale favored the biological activity of the bacterial inoculum, promoting the degradation of lignocellulose. However, the effect of the bacterial inoculum showed a synergistic effect on both evaluated scales, evidencing higher efficiencies (28%) compared with that indicated in other works [8].

3.3. Product Quality

The physicochemical parameters of the obtained products are shown in Table 1. pH values in the alkaline range were obtained for the three treatments without significant differences between treatments ($p = 0.15$). The pH values in this study were similar to those reported (i.e., 7.5–9.0) in other lignocellulosic waste composting studies [35,36] This type of product has the potential to be used in soils with acidic characteristics [37]. Likewise, the products comply with the Colombian Technical Standard of Quality (NTC 5167).

Table 1. Physicochemical characteristics of the obtained compost product.

Treatment	pH	EC	TOC	TN	GI
		dS/m	%, db	%, db	%
TA	8.4 ± 0.4 [a]	1.5 ± 023 [a]	25.4 ± 0.3 [a]	1.7 ± 0.8 [a]	95.8 ± 1.4 [b]
TB	8.7 ± 0.3 [a]	1.3 ± 0.3 [a]	27. 4 ± 2.4 [a]	2.2 ± 1.0 [b]	85.4 ± 1.2 [c]
TC	8.6 ± 0.1 [a]	1.4 ± 0.2 [a]	32.8 ± 1.7 [b]	2.4 ± 1.1 [b]	83.1 ± 2.1 [a]

Note: EC: electrical conductivity; TOC: total organic carbon; TN: total nitrogen. TA: 50% GW, 32.5% UFW, 2.5% PFW, 13% SW, and 2% PR with bacterial inoculum. TB: 50% GW, 32.5% UFW, 2.5% PFW, 13% SW, and 2% PR (uninoculated). TC: 100% GW. The same letters indicate no significant differences ($p > 0.05$). Different letters indicate significant differences between treatments for the response parameter ($p < 0.05$).

The EC for the three treatments was in the range of 1.8–1.9 dS/m; because this was lower than 3 dS/m, the products are not considered phytotoxic for seeds [38]. Furthermore, no significant differences in EC were found between the products ($p = 0.17$) of the three treatments; therefore, the inoculation or the presence of the mixture of substrates did not affect this parameter. The EC values obtained in this study were lower than those previously reported in the co-composting of green waste and food waste inoculated with lactic acid bacteria, yeasts, and phototrophic bacteria (i.e., 3.5–4.0 dS/m) [39].

The TOC content in all cases was higher than 20%, the minimum value suggested to increase the content of organic matter in the soil [37]. TC showed the highest TOC content of 32.8%, which was associated with the predominance of organic carbon compounds of difficult degradation in GW, which increased the processing time. There were no significant differences between the TOC of TA and TB ($p = 0.028$); therefore, in this study, bacterial inoculation did not have a significant effect on the TOC of the product. On the other hand, the stability of a product has been identified as a fundamental quality criterion. According to the Rottegrade test, all treatments can be classified as class V and do not represent a potential risk due to the nutrient and oxygen competition between microorganisms in the soil [19].

Regarding the concentration of TN, TB and TC presented the highest concentration values (i.e., 2.2–2.4%) and had significant differences relative to TA (1.32%). However, the TN content for all treatments was higher than 1%, which is the minimum TN value required by Chilean (NCH2880) and Colombian (NTC 5167) regulations. Regarding the maturity of the

product, the GI allows for the detection of potential inhibitory effects on seed germination. The TA and TB treatments had GI values greater than 80%. The GI of TA (98.85 ± 2.9) was the highest, probably due to the increased degradation of lignocellulose and phytotoxic substances stimulated by the bacterial inoculum [40]. Likewise, the presence of mineral additives such as PR can increase the availability of nutrients in a product through sorption mechanisms. However, the GI of the TC treatment was below the recommended value of 80% [37]. This indicates that the product may have had phytotoxic substances that affected the germination of radish seeds.

4. Discussion

The thermophilic temperatures reached in TA and TB were consistent with those of previous experiments and those reported for the composting of GW [13,41]. Regarding sanitation conditions, all treatments had temperatures of above 50 °C for three or more consecutive days. In this regard, different authors have indicated that this is favorable for the elimination of seeds and pathogens [42]. The bacterial inoculum affected the TA temperature during the cooling phase. The increase in the temperature gradient of TA has also been reported in other studies associated with the incorporation of bacterial inocula. Furthermore, the temperature in this treatment decreased because of the mineralization and humidification of the organic matter. The cooling phase in TA was reduced by 3 days compared with TB and 15 days compared with TC. However, no significant differences were found between TA and TB (p = 0.72) regarding the reduction in the cooling phase. These results are comparable to those reported by Yu et al. [8] on *Bacillus* sp. and *Aspergillus* sp., with no significant differences in the reduction in the processing time of the inoculated treatment.

With respect to pH, inoculation led to slight increases in pH values due to the volatilization of ammonia caused by lignocellulose degradation. Duan et al. [33] explained that bacterial strains can secrete enzymes such as carboxymethyl cellulose, ammonifying enzymes, and xylanase that can affect pH. This result is similar to that reported by Yu et al. [8]. At the end of the process studied here, the average pH values were between 8 and 9, which were in the range reported in another study (between 7 and 9) [42].

Consistent with the temperature results, the highest level of TOC degradation was found in TA and TB throughout the process. However, the greatest amount of degradation was observed in TA after the incorporation of the bacterial inoculum. This indicated that *Bacillus* sp. and *Paenibacillus* sp. could stimulate the enzymatic activity, mineralization, and humidification processes [43]. Similar results were reported by Jiang et al. [18], who used *Trichoderma* sp. and effective microorganisms. Nevertheless, research with lignocellulosic waste has shown that the inoculum does not have a significant effect on the degradation of the TOC and lignocellulose [44]. The differences in the TOC may be due to different distributions of organic components in the different waste sources and differences in local microbial communities [45]. Vrsanska et al. [46] reported that the enzymatic activity during the process depends on the availability of TN, so at the end of the process, its concentration may decrease. In reports using substrates such as cattle manure with the incorporation of bacterial inocula, similar trends have been observed regarding decreases in TN concentration [47]. However, researchers have also reported that inocula do not adversely affect the concentration of TN but rather increase the inorganic nitrogen [44].

On the other hand, the results obtained in this pilot-scale study (120 kg) had some similarities to what was observed in laboratory experiments (20 kg reactors) [10]. In the case of pH, there were no significant differences between both experimental scales for the product obtained in all treatments. A similar result was reported in the composting of GW with a microbial compound inoculum after a scale-up from the laboratory scale (0.05 L reactor) to 1 m^3 conical piles [8]. The EC values of the products obtained in both experimental scales of this study were lower than 3 dS/m. However, the pilot-scale values (1.3–1.5 dS/m) were lower than those of the laboratory experiment (1.8–1.9 dS/m) due to

the greater leaching of salts in the pilot-scale piles compared with the laboratory reactors that were closed vessels.

At both scales, the lowest TN concentration was found in TA and the highest was found in TC. In the case of TA, this result was probably associated with the fact that the inoculum consumed TN for mineralization processes [40]. In contrast, the greater availability of leaves and lignocellulosic material in TC facilitated the retention of TN at the end of the process. However, this treatment also presented a lower degree of organic matter stability. On the other hand, higher values of TN (1.7–2.4%) were found at the pilot scale compared with the laboratory scale (1.3–1.5%) due to the potential environmental variations of the piles, which could have allowed for the development of different microbial consortia or nitrogen-fixing bacteria that contributed to the conservation of nitrogen in the compost [32]. In contrast, due to their experimental configuration, the 20 kg reactors used in the laboratory had more homogeneous environmental conditions. At both scales, the TOC was lower for the treatments with the substrate mixture (TA and TB) compared with TC. Inoculation did not affect the TOC concentration of the product.

The highest GI values were obtained for the TA product at both scales, with significant differences relative to the uninoculated TB and TC treatments. At the laboratory scale, a GI value of 98.9% was obtained, while at the pilot scale, a value of 95.8% was obtained. However, both scales showed no significant differences in the GI of the TA product.

5. Conclusions

The treatment with a substrate mixture and inoculation during the cooling phase (TA) allowed for a reduction of between 4 and 13 days in processing time compared with the treatment with a substrate mixture and no inoculation (TB) and the treatment with only GW (TC). The results indicated that the bacterial inoculum could affect the native microbiota of the process, although a significant reduction in the process was not achieved. TA showed the highest level of lignocellulose biodegradation (31.7%). The final product of TA was the one with the best agricultural characteristics with a pH of 8.3, TOC of 24.8%, TN of 1.32%, and GI of 98.8%. On the other hand, the scaling effect with the incorporation of the bacterial inoculum was shown to affect parameters such as the TOC, TN, GI, and, to a lesser extent, temperature and pH. The differences between scales were due to diffusion and advent transport phenomena. Likewise, the quality of the product found in this pilot-scale study and that previously obtained from the laboratory experiment with the same treatments had similar values of parameters such as the pH, CE, TOC, and GI. However, the greatest difference was found regarding TN, with higher values for the pilot-scale study. The results obtained in this paper highlight the importance of optimizing the composting of GW, specifically with the use of co-substrates and specific inocula, which can be of interest for composting materials with a high content of lignocellulose such as GW.

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