Enhancing compost quality with bacillus bacteria Leveraging cocoa shells and banana pseudostems

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Abstract. Agricultural waste pollutes natural resources, impacting soil fertility, biodiversity, and CO2 levels. Composting offers an alternative solution. This study evaluated Bacillus albus and Bacillus wiedmannii for composting banana pseudostem and cocoa shell waste. The experiment, divided into 4 treatments, found both residues to be slightly acidic. The pseudostem had higher moisture content (30.5%) compared to the cocoa shell (12.3%). During composting, temperature peaked at 33.7°C and ended at 25.5°C, with a final pH of 7.4 and moisture of 42%. Using statistical



analysis, treatments T3 (cocoa shell/B. wiedmannii) and T4 displayed the best results for various parameters. Additionally, T4 showed significant improvement in NPK content. Germination and root growth tests with cucumber seeds revealed no phytotoxicity, highlighting the effectiveness of composting for waste management and its potential use in agriculture.

1. Introduction

Every agricultural activity generates substantial unused waste. Studies by Sánchez et al. (2019); Salgado (2020) suggest that in developing countries, approximately 80% of this waste is burned, while only a small fraction is used productively. This mismanagement, as highlighted by Velastegui et al. (2017), leads to soil and water contamination, along with the proliferation of bacteria and diseases.

Composting offers a viable alternative to manage agricultural waste. Banana stems and cocoa shells, for example, can be transformed into nutrient-rich compost through controlled decomposition (Pérez, 2020). This not only fertilizes the soil but also promotes overall environmental sustainability. In Ecuador, banana cultivation generates significant waste, with Segarra (2022) estimating a yearly total of approximately 12,402,620.44 tons.

According to Silva et al. (2021), Ecuador's banana production is concentrated in the province of Manabí, making it the country's leader in both banana stem waste and cocoa residue. Manabí, along with Guayas, Los Ríos, Esmeraldas, Santa Elena, and El Oro, are the main cocoa producers (Silva et al., 2021).

Condori (2022) suggests that adding efficient microorganisms like *Bacillus spp.* and subtilis can accelerate composting, reduce unpleasant odors, and increase nutrient availability. These bacteria, as described by Moncayo (2021), secrete proteins and metabolites that benefit plants by counteracting pests and diseases, promoting development, fixing nitrogen, and synthesizing phytohormones for root growth (Bonilla et al., 2021).

This research aims to analyze the effect of Bacillus bacteria on the quality of compost derived from agricultural waste (banana *pseudostem* and cocoa shell). Our goal is to contribute to environmental care by promoting sustainable agriculture through the efficient utilization of organic waste. This aligns with the provisions of Chapter 83, Item 6 of the Constitution of Ecuador, which emphasizes the

responsibility of Ecuadorians to "respect the rights of nature, preserve a healthy environment, and use natural resources rationally, sustainably, and permanently."

2. Methodology

The research was conducted at the Organic Fertilizers Unit (Orchidarium) of the Environmental Engineering career in Escuela Superior Politécnica Agropecuaria de Manabí Manuel Félix López (ESPAM MFL). The unit is situated on the El Limón site, located in the Calceta parish, Manabí province. The specific coordinates are UTM WGS 84: 0°49'28" South latitude and 80°10'54" West longitude (Figure 1). This research employs a combined experimental and bibliographic approach (Ramos, 2021; Arellano, 2023).

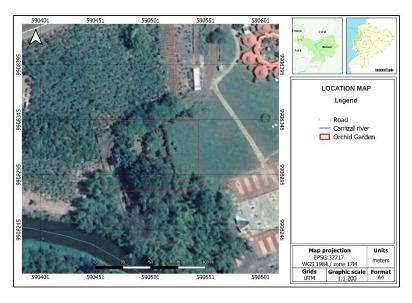


Figure 1. Location of research site.

Experimental design

A two-factor completely randomized design (DCA) was employed for this experiment (Delgado & García, 2023). The research was conducted in a semi-controlled environment with consistent starting materials. Four treatments were established, representing all possible combinations of factor levels (Table 1). Each treatment was replicated four times, resulting in a total of 16 experimental

units. Data analysis followed established statistical methods, including analysis of variance (ANOVA) with a 5% significance level and Tukey's HSD test for multiple comparisons of means (Delgado & García, 2023).

Table 1. Combination of levels and factors.

Treatments	Agricultural residuals	Bacteria
T ₁	Banana pseudostem	B. wiedmannii (Miller et al., 2016)
T ₂	Banana pseudostem	B. albus (Solano-Bastida, 2023)
Т 3	Cocoa shell	B. wiedmannii
Т 4	Cocoa shell	B. albus
Repetitions	<u>4</u>	
Quantity of manure x EU	<u>13kg</u>	
Amount of agricultural waste x EU	<u>13kg</u>	

Composting setup

Each treatment was conducted in a dedicated 40-liter plastic tub. The tubs were filled with a total of 13 kg of waste, with a 50:50 ratio of banana *pseudostem* and cocoa shell (by weight). This balanced composition was chosen because, according to Herrera (2022), incorporating dry manure in equal proportion to the waste accelerates the temperature increase during composting and shortens the processing time.

Material characterization

The initial properties of the banana *pseudostem* and cocoa shell waste were determined through a literature review (Hidalgo, 2019). Following the guidelines established by Ramos (2021), the material was collected in the morning (between 9:00 AM and 10:00 AM) to avoid extreme heat exposure. Sealed, sturdy containers were used for transport to ensure material integrity.

The key physicochemical properties analyzed included moisture content, pH, and the levels of nitrogen, phosphorus, potassium, calcium, and magnesium. These parameters, as highlighted by Hidalgo (2019), provide valuable insights into the nutrient availability of the composted residues. The chemical analysis was conducted at the National Autonomous Institute of Agricultural Research (INIAP) - Pichilingue laboratory located in Quevedo canton, Los Ríos province.

Bacterial incorporation and composting process

Banana *pseudostem* and cocoa shell waste were obtained from the agricultural production areas of ESPAM MFL and transported to the Orchidarium unit. To

expedite degradation, the waste was shredded using an electric forage chopper to achieve a particle size of 1-10 cm (Ramos, 2021).

To introduce *B. albus* and *B. wiedmannii* bacteria, solids from recycled cattle manure (SER) were used. These SER materials, as noted by Diaz and Garcia (2017), harbor abundant bacterial populations. Following the guidelines established by Herrera (2022), the manure was combined with the ground waste in a 50:50 ratio (cocoa shell:banana *pseudostem*). During the mesophilic phase, each experimental unit received 2 liters of bacterial inoculant solution for seven consecutive days, constituting 5% of the total degrading material.

After bacterial incorporation, the compost was manually moistened every two weeks. Digital Soil Tester equipment was used to monitor and control humidity, temperature, and pH. Measurements were taken three times a week, both in the morning (8:30 AM) and afternoon (5:00 PM), to minimize temperature-induced variations (Hidalgo, 2019). To promote optimal temperature increase and expedite organic matter degradation, each EU was covered with black plastic (Delgado & García, 2023). Table 2 details the specific parameters tracked throughout the composting process. This data was crucial for monitoring progress and evaluating the final compost quality in each treatment.

Table 2. Compost parameters during its degradation. Source: García & Delgado (2023)

<u>Parameters</u>	Ideal range that compost should have during its degradation
рН	4.5 – 8.5 Ideal range
Humidity	45%- 60% Ideal range
Temperature	ideal between 50 and 60 °C or up to 65 °C

Composting duration and analysis

The composting process lasted for 20 weeks. During the initial month, the mixture was turned every three days to manage temperature and maintain optimal humidity levels (between 45% and 60%) for efficient nutrient transport (Acosta & Peralta, 2015). Following the degradation period, the compost's physicochemical characteristics were analyzed. These included temperature, pH, electrical conductivity (EC), organic matter (OM), and macroelements (nitrogen, phosphorus, potassium). Table 3 details the specific methods used and the ideal ranges associated with high-quality compost. Physical parameter analyses were conducted at the Bromatology Laboratory within the agroindustrial area of ESPAM MFL. Chemical analyses, consistent with the previous phase, were performed at the INIAP laboratory.

Table 3. Methods for evaluating compost quality indicators. Source: Delgado & García (2023); Hidalgo (2019); Mero & Barreiro (2021).

D	M d 1	Limits		
Parameters	Methods	Class A	Class B	
Temperature	Thermometer (electrode)	5.00	5.00	
рН	Thermometer (electrode)	7.5	8.5	
Electrical Conductivity (EC)	Conductimeter	<3	<8	
Organic Matter (OM)	US EPA SW 846 Method 6010D	≥45	≥25	
C/N Ratio	Organic Matter and Nitrogen	10 - 45	45.1 - 70	
Macroelements (N, P, K)	US EPA SW 846 Method 6010D	≥0,1	≥0,6	

Carbon-Nitrogen ratio (C/N) determination

The carbon-nitrogen ratio (C/N) was evaluated to assess compost maturity. This ratio was calculated based on the percentage of organic matter (%OM) and the percentage of nitrogen (%N). Jackson's constant was employed to estimate the carbon content from the organic matter percentage. The following equation by Mero and Barreiro (2021) was used to calculate the C/N ratio:

$$\left(\frac{C}{N}\right) = \frac{(\%OM)x0.58}{\%N}[100]$$

Phytotoxicity test for compost maturity

The maturity of the compost was evaluated using a phytotoxicity test on cucumber seeds (Cucumis sativus) following the methodology proposed by Urriola et al. (2021). This test assessed seed germination and root elongation over three days.

The compost from the two most promising treatments was selected for this evaluation. Ten grams of each sample were weighed and directly applied to the seeds. Petri dishes lined with three layers of filter paper served as the test containers. Ten cucumber seeds were placed individually in each petri dish, and the date and time of seed contact with the substrate were documented (Urriola et al., 2021).

The plates were maintained at room temperature. Germinated seeds were counted daily at a consistent time for both the compost extract and the control group. After three days, the relative germination percentage (RGP), relative root growth (RRG), and germination index (GI) were calculated using the equations provided by Camacho et al. (2019). Table 4 summarizes the PGR and GI indicators as described by Urriola et al. (2021).

$$RRG = \frac{Average\ root\ length\ in\ the\ extract}{Length\ average} x100\ [2]$$

$$RGP = \frac{Number\ of\ germinated\ seeds\ in\ the\ extract}{Number\ of\ germinated\ seeds\ in\ the\ control}\ x\ 100[3]$$

$$GI = \frac{RGP X RRG}{100} [4]$$

Table 4. RGP and IG indicators. Source: Urriola et al. (2021).

RGP		GI	
77.1	They are	≥ 80%	Indicates that there are no phytotoxic substances, or they are in very low concentration
than 80%		≤ 50%	Indicate that there is a strong presence of phytotoxic substances
	immature	Value between 50% and 80%	It will be interpreted as the moderate presence of these substances

3. Results and discussion

Physical and chemical characteristics of banana pseudostem and cocoa shell residues

Below (Table 5) are the data for humidity, pH, nitrogen, phosphorus, potassium, calcium and magnesium of the banana *pseudostem* and cocoa shell residues.

Table 5. Physical and chemical parameters of the experimental material.

Parameters	Banana pseudostem	Cocoa shell
Humidity (%)	30.5	12.3
Ph	5.8	5.4
Nitrogen (%)	1	1.1
Match (%)	0.61	0.24
Potassium (%)	4.08	3.38
Calcium (%)	1.02	1.13
Magnesium (%)	0.24	0.28

Banana pseudostem

One of the experimental materials studied was the banana *pseudostem*, which had a humidity of 30.5% and a slightly acidic pH of 5.8. It contained 1% nitrogen (N), 0.61% phosphorus (P), 4.08% potassium (K), 1.02% calcium (Ca), and

0.24% magnesium (Mg). These values align with the findings of Quiceno et al. (2014), who reported pH values between 4.5 and 6.5, and Murguetio et al. (2019), who found N=0.850%, P=0.600%, K=5.810%, Ca=2.950%, and Mg=0.480%. However, the humidity value differed from Ordoñez and Sepulveda's (2019) research, which reported a humidity of 15.86%.

The banana *pseudostem* has various characteristics that benefit plant growth and its usage. Pedraza (2019) highlighted that the humidity in the banana *pseudostem* provides a strong base for the plant to support up to 50 kg. Similarly, Hidalgo (2019) emphasized that the banana *pseudostem* contains adequate nutrients for use as a quality compost raw material.

Cocoa shell

The second experimental material used was the cocoa shell, which had a humidity of 12.3% and a pH of 5.4. It contained 1.1% nitrogen (N), 0.24% phosphorus (P), 3.38% potassium (K), 1.13% calcium (Ca), and 0.28% magnesium (Mg). The humidity values differed from those in the research by Vivanco, Matute, and Campo (2017), which reported a humidity of 8.74%. The pH values were similar to those obtained by Castillo et al. (2018), who reported a pH of 6.25. The NPK content was consistent with Puentes et al. (2015), who found N=2.07%, P=0.23%, and K=2.42%.

The cocoa shell provides proteins, carbohydrates, lipids, and minimal quantities of vitamin C (around 0.5 to 2 mg per 100 g of shell). These values can vary slightly depending on the variety of cocoa and growing conditions. Additionally, the cocoa shell contains phenolic compounds, which benefit compost production (Vivanco et al., 2017).

Influence of bacteria of the genus bacillus on the quality of compost

Temperature

The composting piles began with an ambient temperature that fluctuated between 28.3°C and 29.9°C, then varied during the four months of monitoring. T1 (Banana / B. wiedmannii) presented the lowest temperature at 26.4°C in week 8, while in week 14 it reached a maximum temperature of 33.5°C. T2 (Banana / B. albus) had its lowest temperature value in the last two weeks of monitoring, while in week 14 it obtained its maximum temperature of 33.5°C. T3 (Cocoa/ B. wiedmannii) maintained constant temperatures during the four months of monitoring; however, in week 8 it showed a temperature of 27.5°C, this being the lowest, and reached a maximum temperature of 33.7°C. In T4 (Cocoa / B. albus), in week 3 it reached a temperature of 34.1°C, the remaining weeks were

maintained between 30°C and 32°C, ending with a temperature of 25.5°C (Figure 2).

These values agree with what was indicated by Villarreal et al. (2018) that the temperature is between the ranges of 20°C to 45°C, as it is optimal for the growth of the bacteria used, and thus, when turning, the reduction of pathogens and bad odors is achieved. Therefore, Bohórquez (2021) indicates that microbial activity increases the temperature of the composting process. In this way, it is possible to see the evolution in efficiency and the degree of stabilization that the process has reached since there is a direct relationship between temperature and the degradation of organic matter.

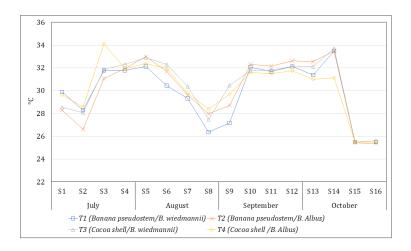


Figure 2. Compost temperature during degradation.

Humidity

At the beginning of the monitoring period, the humidity levels were recorded at 63.0% for T1, 63.4% for T2, 62.9% for T3, and 62.8% for T4 (Figure 3). Over the four months of monitoring, the humidity gradually decreased. Between weeks 5 and 8, the humidity levels were consistently around 55.1% for T1, 55% for T2, 55% for T3, and 55.1% for T4. By weeks 13 and 14, the values had further decreased, reaching 43% for T1, 42% for T2, 43% for T3, and 41% for T4. These findings align with the research by Cajusol and Moisupe (2019), who reported humidity values between 42% and 31% after 3 months and 2 weeks.

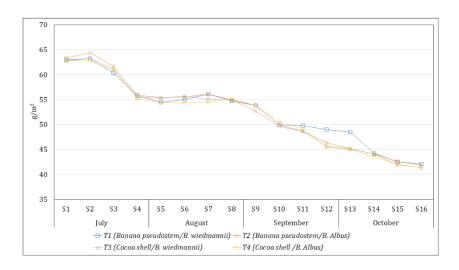


Figure 3. Compost moisture during degradation.

Optimal humidity values for the growth of microorganisms range between 40% and 60%; values lower than this range can decrease microbial activity and delay the composting process (García, 2022). The compost's humidity must be such that water does not completely fill the pores, allowing for the circulation of oxygen and the reaction of other produced gases.

In the final phase of composting, the temperature should be maintained between 50°C and 70°C (García, 2022). Excess humidity can be mitigated with better aeration, which also helps in temperature control. The optimal humidity varies depending on the type of waste, as it is crucial for nourishing microorganisms and transporting nutrients during organic decomposition (Bohórquez, 2021).

pH (Hydrogen Potential)

At the beginning of the composting process, the pH value ranged between 6 and 6.5. In week 2, T2 recorded a pH of 5.8. Between weeks 9 and 12, the pH levels stabilized within a neutral range: T1 ranged from 6.7 to 7.2, T2 maintained a pH of 7, T3 ranged from 6.7 to 7, and T4 ranged from 6.6 to 7. In weeks 13 and 14, T1 showed a slightly alkaline pH of 7.9, and T2 had a pH of 8.2, while T3 presented pH values ranging from 6.3 to 7.4, and T4 from 6.3 to 7.5 (Figure 4).

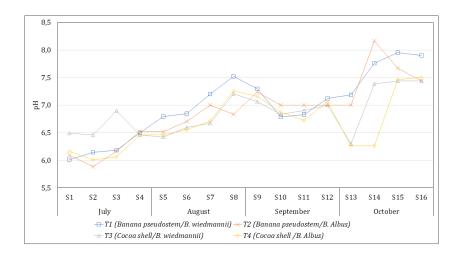


Figure 4. pH of the compost during degradation.

These findings align with Cajusol and Moisupe (2019), who reported pH values between 6.9 and 7.3, emphasizing that increased temperature indicates thermophilic microbial activity. Chávez and Tréboles (2023) state that pH significantly influences the composting process by affecting microbial activity and serving as an indirect indicator of aeration levels. Under anaerobic conditions, organic acids are generated, lowering the pH and inhibiting organic degradation. Therefore, maintaining a pH above 7.5 during the composting process is an indicator of effective decomposition (Bohórquez, 2019).

Likewise, the data from the measurement of humidity, temperature, electrical conductivity (EC), organic matter (OM), macroelements and microelements of the compost as a final product are presented (Table 6).

рΗ

The pH values of the different compost treatments were alkaline, with the highest value being T1 at an average of 9.55 and the lowest being T2 at 9.39. These results were higher than those reported by Rivero (2015), where the pH ranged between 7.03 and 7.43. However, they were comparable to the findings of Monges et al. (2020), who reported values between 8.27 and 9.6.

Table 6. Physicochemical characteristics of compost.

Parameters	T ₁	Т 2	Т 3	Т 4
pН	9.55	9.39	9.53	9.39
Temperature	26.1	26.5	26.85	27.08
Electrical conductivity (EC)	5.53	5.56	3.68	3.38
Organic matter (OM)	50.33	54.88	55.33	59.4
C/N Ratio	14.24	14.47	14.93	17.84
Nitrogen (N)	2.05	2.2	2.15	1.93
Phosphorus (P)	0.61	0.61	0.63	0.63
Potassium (K)	2.76	2.61	2.62	2.52

As shown in Table 7, the ANOVA analysis evaluated the influence of each treatment and the interaction between the bacteria and residue factors on the pH variable. It was determined that the interaction between the two factors is not significant, as the P value was 0.949. Therefore, there is not enough evidence to reject the null hypothesis of equality.

Table 7. Analysis of Variance (SC type III).

	CS	FD	MC	F	O
Bacteria	0.090	1	0.0900	0.96	0.34
Waste	2.25e-4	1	2.25e-4	0.002	0.96
Bacteria* waste	4.00e-4	1	4.00e-4	0.004	0.94
Mistake	11.37	12	0.093		
Total	1.21	15			

The following research provides varied statistical perspectives on the influence of each treatment on pH. Noboa (2021) and Condory and Bravo (2023) found significant statistical differences in the initial and final pH, establishing that the treatments affect the pH at different stages of the composting process. However, Moncayo (2021) did not find significant differences between the treatments, concluding that the application of microorganisms in the composting process does not influence the pH. Instead, it primarily reduces the composting time and provides nutrients to the compost. The results presented by Moncayo are similar to those obtained in the present investigation.

Temperature

The temperatures in the different treatments were as follows: T1 at 26.1°C, T2 at 26.5°C, T3 at 26.85°C, and T4 at 27.08°C. These results differ from those reported by Rivero (2015), where the temperature values of the compost ranged between 7.21 and 7.41°C.

Table 8. Analysis of Variance (SC type III).

	CS	FD	MC	F	Q
Bacteria	0.390	1	0.390	2005	0.182
Waste	17.55	1	17.55	9013	0.011
Bacteria* waste	0.030	1	0.030	0.157	0.699
Mistake	23.37	12	0.1948		
Total	4.51	15			

The ANOVA analysis results presented in Table 8 indicate the influence and interaction between the variables, focusing on the factors analyzed and the temperature variable. The analysis determined that the interaction between the two factors is not significant, as indicated by the P value of 0.699. Therefore, the null hypothesis of equality is accepted. However, it is noteworthy that the waste variable individually showed a significant difference of 0.01, indicating an interaction between the waste factor and temperature in the absence of bacteria.

The Tukey test results for the residual factor and its relationship with temperature are presented in Table 9, revealing significant differences between the residual means across different temperature levels, with agricultural waste exhibiting the highest average.

Table 9. Tukey test for the residual factor, temperature. Means with a common letter are not significantly different (p > 0.05)

Was	te So	cks	n	EE		
1	20	5.30	8	0.16	Α	
2	30	5.96	8	0.16		В

These findings align with prior research; for instance, Moncayo (2021) reported a p-value of 0.977 in the ANOVA test, indicating no significant differences among the treatments evaluated. Similarly, Delgado and García (2023) and Gavilanes (2021) observed no statistical differences in temperature during the composting phases across various treatments. These studies collectively suggest that the treatments did not significantly affect temperature variation during composting.

Electrical Conductivity (EC)

Regarding electrical conductivity, the following results were obtained: T1 with 5.50, T2 with 5.30, T3 with 3.68, and T4 with 3.45. These values differ from those reported by Monges et al. (2020), who obtained values ranging between

1.26 and 2.2. Similarly, Rivas and Silva (2020) reported values varying between 2.8 and 11.4, noting that these values depend on the compost class (Class A and Class B).

Table 10. Analysis of Variance (SC type III).

	CS	FD	MC	F	Q
Bacteria	0.0784	1	0.0784	0.679	0.426
Waste	162.409	1	162.409	140.665	< .001
Bacteria* waste	0.1089	1	0.1089	0.943	0.351
Mistake	13.855	12	0.1155		
Total	17.81	15			

The ANOVA analysis for the electrical conductivity parameter is presented in Table 10, detailing the interactions between the factors and the evaluated parameter. It was found that the interaction between the bacteria and residue factors is not significant, with a P value of 0.351. Thus, there is insufficient evidence to reject the null hypothesis of equality.

However, it is noteworthy that while the interaction between the bacteria and residue factors did not yield significant differences, the residue factor individually influences electrical conductivity. It demonstrates statistical differences regardless of the presence or absence of bacteria.

Table 11 presents the Tukey analysis for the Residuals factor in the electrical conductivity parameter, revealing significant differences between the residual means across the two levels, with agricultural waste showing a higher average.

Table 11. Tukey test for the residual factor, electrical conductivity. Means with a common letter are not significantly different (p > 0.05)

Waste	Socks	n	EE		
1	3.53	8	0.12	Α	
2	5.55	8	0.12		В

The studies referenced below provide various perspectives on the statistical influence of applied treatments on the electrical conductivity parameter. Kaqui (2023) did not find significant statistical differences in this parameter, suggesting that the treatments did not significantly affect electrical conductivity. In contrast, Condory and Bravo (2023) observed over a 30-day period that electrical

conductivity did not exhibit statistical differences. However, Delgado and García (2023) reported finding significant differences in electrical conductivity.

Organic Matter (OM)

T1 presented an organic matter (OM) value of 57.03, T2 had a value of 52.90, T3 obtained 56.71, and T4 recorded 60.76. These values are lower compared to those reported by Rivero (2015), where values ranged between 78 and 66.76. In contrast, these values align more closely with the findings of Monges et al. (2020), who reported values between 15.59 and 52.20.

Table 12 displays the results of the ANOVA analysis investigating the effects of bacteria, residues, and their interaction on the organic matter parameter, providing a statistical evaluation of their interactions. The analysis determined that the interaction between these two factors significantly affects the evaluated parameter, with a P value of 0.001. Hence, there is sufficient evidence to reject the null hypothesis of equality.

Table 12. Analysis of Variance (SC type III).

	CS	FD	MC	F	Q
Bacteria	74,563	1	74,563	183,953	< .001
Waste	0.226	1	0.226	0.557	0.470
Bacteria* waste	90,821	1	90,821	224,062	< .001
Mistake	4,864	12	0.405		
Total	170.47	15			

Additionally, individual parameter ANOVA analysis reveals that the bacteria factor independently exerts significant effects on organic matter, with a p-value of 0.01. This underscores that bacteria play a significant role in influencing this parameter.

The Tukey analysis of residuals (Table 13) revealed statistically significant differences. Notably, the *B. albus* treatment within the bacteria factor exhibited the highest mean value. These findings align with Chávez and Tréboles (2023), who reported significant statistical differences. However, Delgado et al. (2018) observed no significant differences in only one treatment, attributing this to reduced carbon levels during composting. Similarly, Muñoz et al. (2020) also found significant statistical differences in their research.

Table 13. Tukey test for the bacteria factor, organic matter. Means with a common letter are not significantly different (p > 0.05)

Waste	Socks	n	EE		
1	52.83	8	0.93	Α	
2	57.15	8	0.93		В

Tukey's test results (Table 14) reveal statistically significant differences between treatment means. Notably, treatment 4 exhibits a considerably higher mean compared to treatments 1, 2, and 3. This suggests that treatment 4 has a significantly greater effect.

Table 14. Tukey test. Means with a common letter are not significantly different (p > 0.05)

Treatments	bacteria	Waste	Socks	N	EE			
1	1	1	50.33	4	0.32	Α		
2	2	1	50.88	4	0.32		В	
3	1	2	55.33	4	0.32		В	
4	2	2	59.41	4	0.32			C

C/N ratio

The C/N ratios of the treatments ranged from 14.24% (T1) to 17.85% (T4). These values fall within the range reported by Barreros (2017) of 14.92% to 20.48%. However, they are lower than the 20% to 30% range observed by Quituisaca (2023). Notably, Mero and Barreiro (2021) suggest an optimal C/N ratio between 10% and 45% for finished Class A compost, indicating that all treatments have the potential to become suitable compost.

Table 15. Analysis of Variance (SC type III).

	CS	FD	MC	F	Q
Bacteria	9,657	1	96,566	306	< .001
Waste	16,954	1	169,538	537	< .001
Bacteria* waste	6,983	1	69,828	221	< .001
Mistake	0.379	12	0.0315		
Total	9,657	15			

A two-way ANOVA was conducted to assess the independent and interactive effects of bacteria and residue factors on the C/N ratio. The analysis revealed a significant interaction effect (p = 0.001), indicating that the influence of one

factor (bacteria or residue) on the C/N ratio depends on the level of the other factor.

Tukey's multiple comparison test (Table 16) revealed no significant difference (p < 0.01) between the C/N ratios of treatments 1 and 2. However, treatments 3 and 4 formed a statistically distinct group with a higher mean C/N ratio, particularly treatment 3. This suggests treatment 3 may be favorable for achieving a higher final C/N ratio in the compost.

Table 16. Tukey test. Means with a common letter are not significantly different (p > 0.05)

Treatments	bacteria	Waste	Socks	N	EE			
1	1	1	14.24	4	0.09	Α		
2	2	1	14.27	4	0.09	Α		
3	1	2	14.97	4	0.09		В	
4	2	2	14.85	4	0.09			С

Table 17 reveals that the compost's pH falls outside the acceptable range for Class A and B composts, indicating a potential issue with high alkalinity. However, the electrical conductivity falls within the permissible limits for Class B compost. Fortunately, the organic matter content, C/N ratio, and macroelement levels (nitrogen, phosphorus, and potassium) all meet the standards for Class A compost, signifying good quality in these aspects. This suggests the compost may be suitable for some applications despite the elevated pH.

Table 17. Compost quality classification.

Parameters	Results (av	erage)
	Туре	
рН	9.47	В
Electrical Conductivity (EC)	4.54	В
Organic Matter (OM)	54.99	Α
C/N Ratio	15.38	Α
N	2.08	Α
Q	0.62	Α
k	2.63	Α

Overall, the C/N ratios of all treatments fall within the range for Class A compost (refer to Table 17 for specific values), indicating a moderate availability of nitrogen and carbon for plant uptake. Interestingly, treatment T4 exhibited a

C/N ratio closest to the ideal range for mature compost, suggesting it may be the most optimal treatment as an organic fertilizer. This aligns with the findings of Quituisaca (2023), who observed a continued release of available nitrogen during crop use as the compost degrades further.

However, it's important to consider the wider context of C/N ratios in compost maturity. Hoseini (2021) reported significant differences in C/N ratios around 30, with relationships between treatments decreasing as the compost matured (evidenced by a lower C/N ratio). In contrast, Condezo (2018) did not observe significant differences in C/N ratios between treatments. These contrasting findings highlight the potential influence of various factors beyond just C/N ratio on compost maturity and suitability for specific applications.

Macroelement content

The analysis focused on three key macroelements: nitrogen (N), phosphorus (P), and potassium (K). Treatment T4 exhibited the highest nitrogen content (2.2%), followed by T3 (2.15%), T1 (2.05%), and T2 (1.93%). Phosphorus levels were fairly consistent across treatments, with both T1 and T2 showing 0.61% and T3 and T4 showing slightly higher values at 0.63%. Potassium content followed a similar pattern to nitrogen, with T4 having the highest value (2.73%) and T2 having the lowest (2.61%).

For comparison, the study by Rivero (2015) reported a range of 1.86% to 2.11% for nitrogen and 2.35% to 3.05% for potassium. While the nitrogen content in all treatments falls within this range, the potassium levels in this study are slightly higher than the upper limit reported by Rivero (2015).

Nitrogen

The ANOVA analysis (Table 18) revealed no significant interaction effect (p = 0.337) between the bacteria and residue factors on the macroelement content. This suggests that the influence of one factor (bacteria or residue) on the macroelement levels does not depend on the level of the other factor. It's important to note that these findings differ from those reported by Chávez and Tréboles (2023), who observed a significant interaction effect with values ranging from 0.38% to 2.28%. These discrepancies may be due to differences in experimental design, materials, or specific macroelements analyzed.

The impact of treatments on nitrogen content appears to be context-dependent. Anchundia (2020) observed significant differences between treatments (p = 0.00018), suggesting certain treatments can influence nitrogen content. Conversely, Goya (2013) and Condezo (2018) found no significant differences in

nitrogen (or overall macronutrients) amongst their treatments using ANOVA analysis. These contrasting findings highlight the potential influence of various factors beyond just the treatments themselves.

Table 18. Analysis of Variance (SC type III).

	CS	FD	MC	F	Q
Bacteria	2.50e-5	1	96.56	0.005	0.940
Waste	0.00160	1	169.53	0.379	0.549
Bacteria* waste	2.50e-5	1	69.82	0.005	0.940
Mistake	0.05055	12	0.031		
Total	0.54	15			

The impact of treatments on nitrogen content appears to be context-dependent. Anchundia (2020) observed significant differences between treatments (p = 0.00018), suggesting certain treatments can influence nitrogen content. Conversely, Goya (2013) and Condezo (2018) found no significant differences in nitrogen (or overall macronutrients) amongst their treatments using ANOVA analysis. These contrasting findings highlight the potential influence of various factors beyond just the treatments themselves.

Phosphorus

The ANOVA analysis of the phosphorus content (Table 19) revealed no significant interaction effect (p = 0.940) between the bacteria and residue factors. This indicates that the influence of one factor (bacteria or residue) on phosphorus levels is not dependent on the level of the other factor.

Table 19. Analysis of Variance (SC type III).

	CS	FD	MC	F	Q
Bacteria	2.50e-5	1	2.50e-5	0.005	0.940
Waste	0.001	1	0.001	0.379	0.549
Bacteria* waste	2.50e-5	1	2.50e-5	0.005	0.940
Mistake	0.050	12	0.00421		
Total	0.05	15			

Similar to nitrogen content, the impact of treatments on phosphorus appears to be study dependent. Chávez and Tréboles (2023) reported statistically significant differences in phosphorus content within their compost materials, ranging from 0.07% to 0.56%. These values fall within the range considered suitable for high-quality compost by Castillo (2020), suggesting the applicability of our findings despite potential variations.

However, other studies have shown contrasting results. Condezo (2018) did not observe any significant influence of treatments on phosphorus in their research. In contrast, Condory and Bravo (2023) and Zambrano and García (2023) found significant differences in phosphorus content between treatments using variance tests. These discrepancies highlight the potential influence of various factors beyond just the treatments themselves, such as the type of feedstock materials used.

Potassium

The ANOVA analysis for potassium content (Table 20) revealed no significant interaction effect (p=0.940) between the bacteria and residue factors. This suggests that the influence of one factor (bacteria or residue) on potassium levels does not depend on the level of the other factor.

Table 20. Analysis of Variance (SC type III).

	CS	FD	MC	F	Q
Bacteria	0.058	1	0.058	0.40	0.539
Waste	0.056	1	0.056	0.38	0.547
Bacteria* waste	0.002	1	0.002	0.01	0.893
Mistake	1,761	12	0.146		
Total	1.88	15			

The potassium content in this study did not exhibit a significant interaction effect between the bacteria and residue factors (p = 0.940, Table 20). This is unlike the findings of Chávez and Tréboles (2023) who observed significant differences in potassium content due to both material and size variations. It's important to consider the overall potassium levels in the context of compost quality. Alurralde et al. (2023) suggest a range of 0.3% to 1.0% potassium for good compost. While Condory and Bravo (2023) found statistically significant differences between treatments, they also reported greater uniformity in potassium readings, potentially indicating a material or process factor influencing overall potassium content. In contrast, Condezo (2018) observed no significant differences in potassium content between treatments. These contrasting findings highlight the potential influence of various factors beyond just the treatments themselves, and the importance of considering the overall potassium level for compost quality.

The analysis (Table 20) revealed no significant interaction effect (p > 0.05) between the bacteria and residue factors on potassium content. This suggests that the variations observed in potassium levels are not directly influenced by how these two factors interact with each other. However, it's important to note

that statistically significant differences were found in other parameters like pH, temperature and macroelements (N, P, K) across the treatments.

Based on these findings, treatment T3 appears to be the most effective overall. Treatment T4 also exhibited significant statistical differences in macroelements, suggesting some comparability in terms of nutrient content. However, further evaluation considering all the parameters (pH, temperature, etc.) might be necessary to definitively determine the optimal treatment for specific applications.

Compost maturity assessment: phytotoxicity test with cucumber seeds

Table 21 summarizes the results of the phytotoxicity test on cucumber seeds, which was conducted to evaluate the maturity of the two most promising treatments identified based on the physicochemical properties analyzed in the previous phase. The test measured three key parameters:

Table 21. CRR, PGR and GI results of the two best treatments.

Species	Treatment	RRG	RGP	GI
	T 3 R 1	85.94	71.67	61.65
	T_3R_2	90.28	67.22	60.85
	T_3R_3	91.18	72.69	66.38
	T 3 R 4	93.67	80.15	74.89
Cucumber	Control	80.55	68.25	60.00
Cucumber	T_4R_1	87.72	71.96	63.27
	T_4R_2	89.92	69.64	62.70
	T_4R_3	89.34	71.96	62.43
	T_4R_4	95.04	61.27	59.58
	Control	81.33	70.55	61.00

The phytotoxicity test revealed a positive effect of treatment T4R4 on cucumber root development. This treatment exhibited the highest relative root growth (RRG) of 95.04 compared to the control and other treatments (Table 21). Conversely, treatment T3R1 displayed the lowest RRG value (85.94). These findings suggest that T4R4 may have low concentrations of phytotoxic substances or even contain compounds that stimulate root growth, leading to its superior performance in the RRG test.

The t-test analysis (Table 22) revealed no statistically significant differences (p > 0.05) between the treatments for RRG, relative germination percentage RGP, and GI. This suggests that neither treatment significantly impacted root development, germination, or disease presence in cucumber seeds. However, it's important to consider the individual treatment responses within the observed

range. While all treatments achieved an RRG above 80% (similar to Urriola et al., 2021 and Camacho et al., 2019), some variations were observed. Treatment T3R4 exhibited the highest RRG (95.04), potentially indicating minimal phytotoxicity or the presence of root-stimulating compounds.

Table 22. T-student test for the parameters RRG, RGP, GI.

Classification	Variable	n	Half	p-value
Treatments	RRG	4	72.93	0.296
Treatments	RGP	4	65.94	0.194
Treatments	GI	4	92.54	0.330

The RGP data revealed complete seed germination across treatments, with some variation in percentage. Treatments T3R2 and T4R4 showed the lowest germination percentages (67.22% and 61.27%, respectively), while T3R4 had the highest (80.15%). The control group achieved a germination rate of 68.25%. These findings suggest that some level of phytotoxic agents might still be present in the compost, potentially affecting germination in some treatments.

The GI values also suggest potential variations in compost maturity. T3R4 displayed the highest GI (74.89), followed by the control (60.00) and T4R4 (59.58). While these values fall below those reported by Huerta et al. (2015) who suggested a minimum of 100% for mature compost, they are still above the threshold for immature compost proposed by Urriola et al. (2021) (below 80% GI).

Conclusions

The initial characterization of the banana *pseudostem* (30.5% humidity, pH 5.8) and cocoa shell (12.3% humidity, pH 5.4) revealed key differences. The *pseudostem* had higher moisture content and slightly lower acidity compared to the cocoa shell. The cocoa shell, however, stood out for its content of calcium, magnesium, vitamins, and proteins, while the *pseudostem* offered a higher concentration of macronutrients. During the composting process, the temperature peaked at 33.7°C before stabilizing at 25.5°C. The initial pH (6.0-6.5) increased to a range of 7.4-7.5 within four months, indicating a shift towards a more neutral environment.

The presence of Bacillus bacteria and its potential impact on compost quality were also investigated. A decrease in overall humidity was observed, dropping from an initial range of 62.9%-63% to around 42% by the end of the process.

This decrease can be attributed to both the decomposition activity of the bacteria and the initial moisture content of the agricultural waste. Interestingly, treatments T3 and T4 (including cocoa shell) showed a greater decrease in humidity, suggesting a potential influence of the feedstock on moisture dynamics. On the other hand, treatments containing *Bacillus albus* (T2 and T4) appeared to be more efficient in overall decomposition. Statistical analysis using ANOVA and Tukey's Test revealed significant differences between treatments. Treatment T3 (cocoa shell with *B. wiedmannii*) stood out in terms of pH, temperature, electrical conductivity, and organic matter content. Treatment T4 excelled in terms of macroelement content (N, P, and K).

The phytotoxicity test with cucumber seeds was used to evaluate compost maturity. Treatment T4R4 exhibited the highest RRG at 95.04, suggesting the presence of beneficial compounds and minimal phytotoxic substances. Conversely, treatment T3R1 displayed a lower RRG (85.94), potentially indicating higher concentrations of unmetabolized phytotoxic agents. However, it is important to note that the t-student test did not detect statistically significant differences between treatments for RRG, RGP or GI.

We believe that our research demonstrates the effectiveness of the procedures tested for enhancing the quality of compost through bacillus bacteria, with significant potential for contributing to waste management and its potential use in agriculture.

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