Bartha respirometric method to evaluate the biodegradation of mixture of coconut husk and organic waste with and without commercial inoculant

Método respirométrico de Bartha para avaliação da biodegradação da mistura de casca de coco e resíduos orgânicos com e sem inoculante comercial

DOI: 10.55905/oelv21n11-067

Recebimento dos originais: 05/10/2023
Aceitação para publicação: 06/11/2023

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ABSTRACT

The population growth increases the demand for food and goods, generating more waste and socio-economic and environmental impacts. Mitigating these effects is a challenge in the field of sanitation, opportunities for different kinds of approaches. Organic waste treatment and recycling, such as coconut husks, are essential to reduce environmental damage. Composting is a viable option, and the Bartha respirometric method is used to assess waste biodegradability. The experiment evaluated biomass aerobic respiration, estimating the compost stabilization time. The use of commercial inoculant accelerated the degradation process. The results indicate that the Bartha method is efficient in monitoring the coconut husks mixture and other waste biodegradability. The study contributes to clarifying the use of this technique in organic waste management.

Keywords: coconut waste, respirometry, Bartha respirometer.

RESUMO
O crescimento populacional aumenta a demanda por alimentos e bens, gerando mais desperdício e impactos socioeconômicos e ambientais. Mitigar esses efeitos é um desafio no campo do saneamento, oportunidades para diferentes tipos de abordagens. O tratamento e a reciclagem de resíduos orgânicos, como cascas de coco, são essenciais para reduzir os danos ambientais. A compostagem é uma opção viável e o método respirométrico de Bartha é usado para avaliar a biodegradabilidade dos resíduos. O experimento avaliou a respiração aeróbica de biomassa, estimando o tempo de estabilização do composto. O uso de inoculante comercial acelerou o processo de degradação. Os resultados indicam que o método Bartha é eficiente no monitoramento da mistura de cascas de coco e da biodegradabilidade de outros resíduos. O estudo contribui para clarificar a utilização desta técnica na gestão dos resíduos orgânicos.

Palavras-chave: respirometria de coco, respirometria de Bartha.

1 INTRODUCTION

Population and economic growth demand for more food and goods, increasing agro-industrial systems production, and consequently, the technologies involved in the entire production process, to meet the demand (MAZZER; CAVALCANTE, 2004; TEIXEIRA, 2019). With the increase in production, there is a rise in waste generated throughout the production chain and the consumer, becoming a social, environmental, and economic issue. Therefore, it is essential that waste management is carried out properly, otherwise, it can cause pollution and diseases, causing problems for the government and society (COUTINHO et al., 2011).

For an integrated waste management is necessary, coordination between the actions to reduce waste generation at the source and the methodologies for proper treatment and disposal. Reuse and recycling technologies are widely studied in an attempt to solve waste disposal difficulties, with the emphasis being placed on composting (LIMA, 2002; OLIVEIRA, 2019; FILOGÔNIO, 2020).

Brazil is one of the largest coconuts producers, and the consumption of its derivatives generates up to 6.7 million tons of shells per year, making its waste an environmental problem, especially in coastal cities where coconut water is highly consumed and appreciated. After consumption, the primary destination of the coconut shells in their natural state is landfills and sanitary landfills (BRASIL, 2010).

An alternative would be to promote circular economy practices, with reuse and recycling of the waste; furthermore, significant areas are required for the final disposal of the shells. On the other hand, sending waste to landfills should be avoided for several reasons. In these environments, coconut shells can collect water in their concavity and proliferate vectors, causing health issues (ISHISAKI et al., 2006).

Composting could be an alternative for treatment (BRASIL, 2010). Microorganisms present in composting processes can be measured by their respiration using respirometric tests, which are sustainable methodologies for standardizing the stability of organic waste (PEDROTI, 2007; MUNIZ, 2010). Bartha's respirometry can be used to quantify the aerobic biological activity of a waste mixture, determining the respiration of an active biomass (MUNIZ, 2010).

The use of respirometry methodology in determining waste biodegradability conditions, especially coconut waste, contributing to sustainable disposal methods for this residue. The research aligns with the sanitary, environmental, and social aspects, collecting data on the biodegradability of coconut shells, diverting them from landfills, aiming to reduce greenhouse gas emissions.

The article assesses the biodegradability of mixtures of organic food waste, coconut shells, and other sources of carbon (grass clippings and dry leaves), with and without a commercial inoculant, using the Bartha respirometric method. These norms aim to provide general guidance to authors regarding writing instructions and academic organization of these communications.

2 BIBLIOGRAPHIC REVIEW

2.1 URBAN SOLID WASTE (USW): SOCIAL, ENVIRONMENTAL AND ECONOMIC ASPECTS

The United Nations (UN) approved the 17 Sustainable Development Goals (SDGs) in 2015, which have been implemented by member states. The 17 SDGs encompass various themes, including poverty eradication, food security and agriculture, health, education, water and sanitation, as well as sustainable patterns of production and consumption. Some SDGs are related to research, such as SDG 3, that focuses on population health and well-being. Regarding waste, if not properly managed, it can lead to vectors proliferation, compromising people's wellbeing. SDG 12 ensures sustainable production and consumption patterns and is connected to waste treatment and its use throughout its lifecycle. In an attempt to address the issue of USW, treatment methods such as composting and biodigestion have emerged, catering to proper final disposal (OLIVEIRA, 2018; TEIXEIRA, 2019).

In 2010, Federal Law 12.305 was established in Brazil, promoting National Solid Waste Policy (NSWP), in order to achieve sustainable and integrated solid waste management in the country (BRASIL, 2010). The principles of the NSWP, besides attempting to promote integrated management, are also sustainable development, eco-efficiency, and cooperation among public, private, and community sectors, besides fostering a shared responsibility for the environment (INSTITUTO DE PESQUISA ECONÔMICA APLICADA, 2012).

Once waste is seen as a business opportunity and a source of profit, rather than just as "garbage," various economic, social, and environmental benefits can be achieved. One of the gains that can be mentioned includes reduced energy costs, waste transportation costs, and final disposal costs (TEIXEIRA, 2019). USW should preferably be sorted at the source, simplifying the sorting process for recycling and treatments like composting (TEIXEIRA, 2019).

Regarding the generation of USW, approximately 66.64 million tons were produced in 2021, with around 270,000 tons of organic solid waste (OSW) being directed to composting yards (SISTEMA NACIONAL DE INFORMAÇÃO SOBRE SANEAMENTO, 2021).

Considering the earlier-presented OSW generation and the issue of proper disposal, concern arises about waste stemming from coconut consumption (*Cocos nucifera* L.). Being a bulky residue, its disposal without treatment would significantly reduce the lifespan of sanitary landfills. Coconut production and consumption in Brazil are growing each year, with consumption taking place in all states (CÉSAR; SILVEIRA; CUNHA, 2009).

2.2 COCONUT: CULTIVATION IN BRAZIL AND WASTE GENERATION

Coconut originated in India or Sri Lanka and is cultivated in more than 85 countries, covering an area of over 14 million hectares (MARTINS; JESUS JÚNIOR, 2014). It arrived in Brazil in 1553, through the state of Bahia, and is commonly referred to as "coco-da-baía" (ARAGÃO *et al.*, 2010). Its cultivation is suited for a tropical climate, spreading throughout the country's coastal region (SILVA; JERÔNIMO, 2012).

Asia stands out with the largest cultivation areas worldwide; however, Brazil leads in productivity and yield (ARAGÃO et al., 2010). In 1990, the country produced approximately 470,000 tons of coconuts, and by 2004, it ranked fourth among the world's top producers (MARTINS; JESUS JÚNIOR, 2014).

Brazil's production in 2017 was 1.79 million tons of coconuts. The Southeastern state produced 219.9 thousand tons of this total, and Espírito Santo produced around 120.65 thousand tons (INSTITUTO BRASILEIRO DE GEOGRAFIA E ESTATÍSTICA, 2018). In 2020, the harvested area for coconuts was approximately 223 thousand hectares, yielding 1.95 billion fruits (BRAINER; XIMENES, 2020).

With a production area of over 290 thousand hectares, around 2.3 billion coconut shells and 469 million leaves fall from coconut trees, causing negative environmental impacts (SILVA; JERÔNIMO, 2012).

To enhance value and promote recycling of coconut waste, its reuse in agriculture, such as substrates or organic fertilizers, has been considered (CARRIJO; LIZ;

MAKISHIMA, 2002). Coconut shells have fibers that are nearly inert, with high porosity, low cost, and high availability (PIMENTA *et al.*, 2015).

These waste materials could significantly contribute to increased food production and quality (SILVA; JERÔNIMO, 2012). The low cost, high availability, and uncomplicated production make coconut shells suitable as substrates or organic fertilizers (BUENO, 2021). One ton of coconut waste can produce approximately 400 kg of organic compost, implying that Brazil could generate about 1.53 million tons of these byproducts that could even be commercialized (NUNES; SANTOS; SANTOS, 2007).

2.3 COMPOSTING: DEFINITION AND IMPORTANCE

Composting is a technology that gives value to OSW, promoting recycling (OLIVEIRA, 2018). It's a natural decomposition process carried out by aerobic microorganisms to stabilize organic matter (OM) in a controlled manner (KIEHL, 2004).

As a widely used technique, composting can be performed with various organic sources and types of waste, and the respirometric method presents certain similar processes (BUENO, 2021). Composting efficiency relates to parameters like carbon-to-nitrogen ratio (C/N), temperature, humidity, pH, material particle size, and others, providing optimal conditions for microorganism development (MASSUKADO, 2008).

Composting stabilizes and reduces the volume of OSW destined for landfills, them value until the end of their life cycle, thus promoting circular economy principles (OLIVEIRA, 2018). In Brazil, the generation of tons of OSW annually highlights the need for reuse to promote sustainable development. Consequently, proper USW treatment is an ongoing concern, and composting is one way of recycling waste, resulting in a stable material suitable for agricultural application (KIEHL, 2004).

2.4 COMPOSTING ACCELERATORS

The use of Effective Microorganisms (EMs) in composting enhances the use of organic matter for agricultural production, reducing the compounds production time (OLIVEIRA, 2018; BUENO, 2021).

Formed by a population of microorganisms, EMs are naturally found in fertile soils and include lactose-fermenting bacteria, yeasts, photosynthetic bacteria, fungi, and actinomycetes (OLIVEIRA, 2018).

The use of EMs can generate income when applied to organic compost production, replacing fertilizers and pesticides containing harmful chemical elements. As a result, EMs reduces environmental and health impacts, enabling healthier production and chemical-free food (BONFIM *et al.*, 2011).

Commercial inoculants are industrialized biological products containing microorganisms, used to expedite the composting process. When added to waste, they promote fermentation, reducing potential odors and the presence of unwanted insects during the process (KORIN AGRICULTURA E MEIO AMBIENTE, 2020).

2.5 BARTHA'S RESPIROMETRIC METHOD

Respirometry estimates the stabilization time of specific waste in the soil, where soil respiration behavior serves as an indicator of microorganism activity. By using respirometry, waste toxicity and biodegradability in the soil can be assessed (CAMPOS, 2008).

The method was developed by Bartha and Pramer in 1965, involving closed systems with 50g of soil, certain substances to be degraded, and titration-based measurements (BARTHA; PRAMER, 1965). Over time, the methodology evolved, utilizing a conductivity measurement for the generated gas (PEDROTI, 2007).

The Bartha respirometric method quantifies aerobic biological activity and aims to estimate the time needed for a waste to stabilize in the soil (COSTA, 2009). Based on carbon dioxide (CO_2) measurement and constructing a gas curve over time, the initial adaptation phase of the curve has a gentle slope, followed by an exponential growth of microorganisms (COSTA, 2009).

The methodology described by Pedroti (2007) employs a closed flask system and analyzes the conductivity of a sodium hydroxide (NaOH) solution.

The primary reactions that occur in the aerobic respirometry system include (RODELLA; SABOYA, 1999):

$$CO_2 + H_2O \rightarrow H_2CO_3 \quad (1)$$

$$H_2CO_3 + 2NaOH \rightarrow Na_2CO_3 + 2H_2O \quad (2)$$

The CO_2 results are measured by conductivity and calculated using the equation (RODELLA; SABOYA, 1999):

$$mg\ de\ CO_2 = M * V * 22 * \left(\frac{C1 - Cx}{C1 - C2}\right) \quad (3)$$

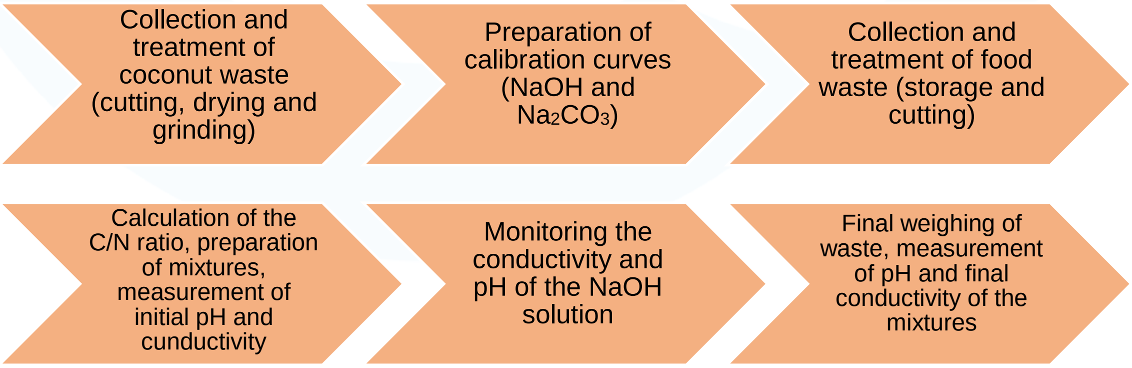
Where:

– M is the molarity of the NaOH solution used (expressed in mol.L^{-1})
– V is the volume used (mL) of the NaOH standard solution in the capture flask
– C1 is the standard concentration of NaOH
– Cx is the conductivity value per sample
– C2 is the concentration of the Na_2CO_3 solution

3 METODOLOGY

The experimental design of the study consists of the steps described in Figure 1.

Figure 1: Flowchart of experimental steps

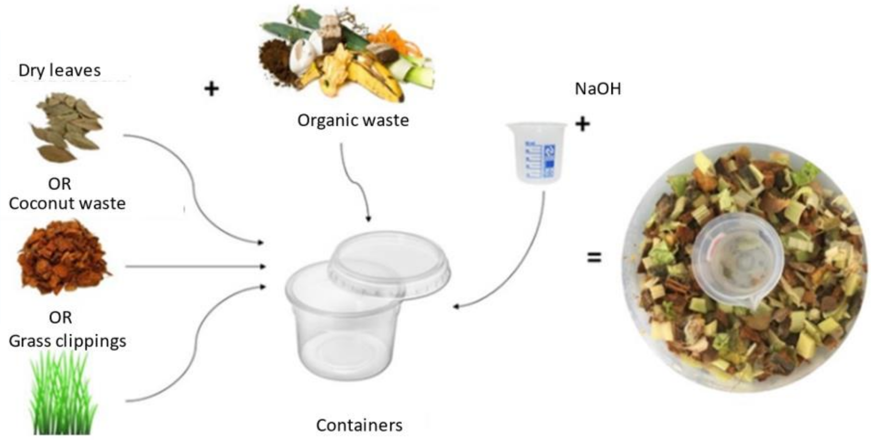


Source: The authors

For the experiment, the adapted Bartha respirometric method, as described by Pedroti (2007), was employed. The coconut shells were cut into 4 pieces, dried for 20 days, and cut againg in 2x2 cm pieces.

For the experimental setup, 28 round 3 L plastic containers with lids were used, and in each of them, a 50 mL polypropylene beaker was added for the NaOH solution. The Figure 2 demonstrates the adapted respirometer.

Figure 2: Adapted respirometer



Source: The authors

Six treatments and one control were conducted, as described in Table 1.

Table 1: Treatment identification and compositions.

Treatments	Composition	Commercial Inoculant
T1	coconut shells + OSW	absent
T2	coconut shells + OSW	present
T3	grass clippings + OSW	absent
T4	grass clippings + OSW	present
T5	dry leaves + OSW	absent
T6	dry leaves + OSW	present
T7 - CONTROL	no waste	absent

Source: The authors

Treatment identified as T7 serves as the control, with no addition of organic waste or inoculant, only the empty container with the NaOH beaker. It is used to verify if the container maintained the conductivity of the NaOH solution or if it was influenced by external factors.

For determining waste proportions, reference values for the C/N ratio from the literature were used, as presented in Table 2.

Table 2: Reference values of the C/N ratio

Waste	C/N ratio	Reference
Coconut shells	132/1	Carrijo, Liz e Makishima (2002)
Fruit remains	35/1	Kiehl (2010)
Vegetable remains	20/1	Massukado (2008)
Leaves	80/1	Massukado (2008)
Grass clippings	32/1	Nascimento *et al.* (2018)

Source: The authors

For the calculation of proportions, the formula proposed by Massukado (2008) was used:

$$\left(\frac{C}{N}\right) = \frac{(Mcc * C{:}Ncc) + (Mrv * C{:}Nrv) + (Mrf * C{:}Nrf)}{Mtotal} \quad (5)$$

Where:

– C/N is the desired carbon-to-nitrogen ratio
– Mcc is the mass of coconut shells/dry leaves/grass clippings
– C:Ncc is the C/N ratio of coconut shells/dry leaves/grass clippings
– Mrv is the mass of vegetable remains
– C:Nrv is the C/N ratio of vegetable remains
– Mrf is the mass of fruit remains
– C:Nrf is the C/N ratio of fruit remains
– Mtotal is the total mass

The proportions of the C/N ratio were calculated for each treatment to achieve an approximate 30/1 C/N ratio. The masses are described in Table 3.

Table 3: Adopted waste proportions for achieving the ideal initial C/N ratio in the experiment considering reference values from the literature

Treatments	Waste Proportions (g)					Final C/N ratio
	Coconut shells	Dry leaves	Grass clippings	Fruit remains	Vegetable remains	
1 e 2	6	-	-	55	89	29,98
3 e 4	-	-	75	40	35	30
5 e 6	-	14	-	29,33	92	30

Source: The authors

The waste materials were individually cut, separated, weighed, and then mixed. A total of 150 g of waste materials were weighed for all treatments except T7 (control), and 10 g of the mixture were removed for pH and conductivity measurement. Thus, all treatments started with a mass of 140 g. The manufacturer's recommendation for the commercial inoculant (CI) was 5 mL of CI for 1000 g of waste materials. The application was performed using a micropipette for each treatment and repetition, proportionally to the quantity in each container. Therefore, 0.75 mL of CI was used for treatments T2, T4, and T6. The parameters analyzed during the research and the equipment used are described in Table 4.

Table 4: Parameters analyzed during the respirometric test

Parameter	**Frequency**	**Equipments**
pH and waste conductivity	Start and end of the experiment	pH meter and conductivity meter
Temperature	Daily, for 37 days	Thermoometer
Carbon dioxide emission	Daily, for 37 day	Conductivity meter
Mass reduction	End of experiment	Scale

Source: The authors

Calibration curves of NaOH and Na_2CO_3 were prepared before the experiment using the conductivity meter. The same equipment was used for daily measurement of conductivity in the NaOH beakers. For the calculation of the amount of CO_2 generated during the experiment, the initial molarity of the NaOH solution measured in triplicate was considered, with a value of 0.539 mol/L.

Values C1 and C2 (Equation 3) correspond to the calibration curves of NaOH and Na_2CO_3, respectively, considering the molarity 0.539 mol/L (x). Thus, the value of C1 is 85.91 mS/cm, and C2 is 59.37 mS/cm. The daily volume of NaOH used was 30 mL, determining the value of V. With all the data available, Equation 3 can be utilized to calculate:

$$mg\ de\ CO_2 = 0{,}539 * 30 * 22 * \left(\frac{85{,}91 - Cx}{85{,}91 - 59{,}37}\right) \quad (6)$$

Where:

Cx is the conductivity value measured in each sample (mS/cm)

Therefore, to determine the daily CO_2 value, measuring the conductivity of the NaOH solution (Cx value in the equation) is required.

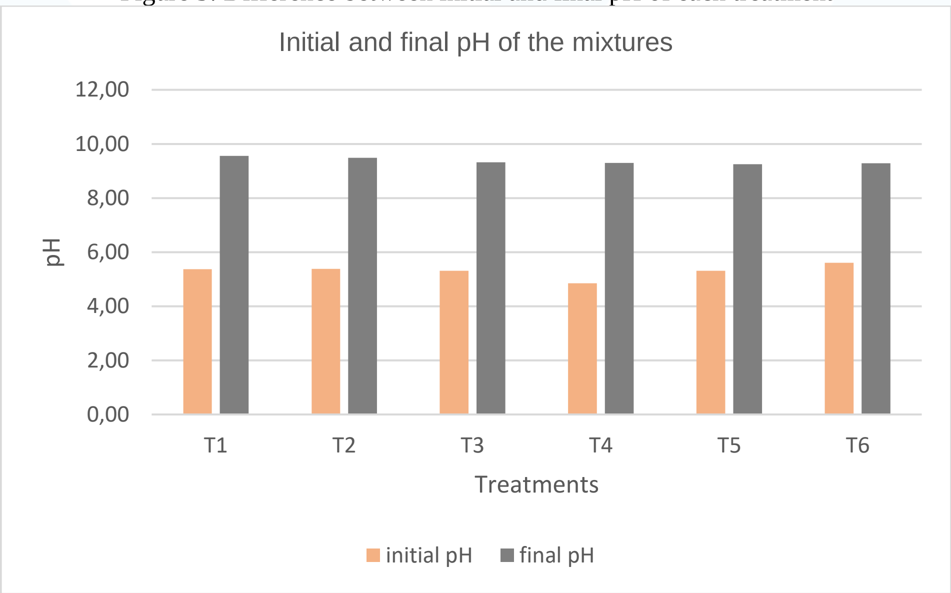
The treatments were conducted in 4 replicates and all parameters were measured in triplicate.

4 RESULTS AND DISCUSSIONS

4.1 PH AND CONDUCTIVITY OF WASTE MIXTURES

Initially, all waste mixtures had pH values in the range of 4 to 6. At the end of the experiment, all of them were in the pH range of 9, in Figure 3.

Figure 3: Difference between initial and final pH of each treatment



Source: The authors

In composting, the desirable final pH of the compost is between 6 and 7.5 (KIEHL, 2004). According to the Ministry of Agriculture, Livestock, and Supply (MAPA), in its Normative Instruction (IN) number 25/2009, the pH of the compost generated after the composting process, classified as Class C, can be safely used in agriculture, and the suitable value must be at least 6.5, concluding that the treatment was favorable for this parameter (MINISTRY OF AGRICULTURE, LIVESTOCK, AND SUPPLY, 2009).

Regarding conductivity, the mixtures initially showed conductivity results in the microSiemens (µS/cm) range, and at the end of the experiment, the mixtures exhibited conductivity values in the milliSiemens (mS/cm) range, with an increase in conductivity across all treatments (Table 5).

Table 5: Average initial and final conductivity of treatments

Treatments	Composition	Inicial Conductivity (µS/cm)	Final Conductivity (mS/cm)
T1	**Coconut + OSW**	1339,70	3,37
T2	**Coconut + OSW + CI**	1307,33	2,90
T3	**Grass + OSW**	349,34	4,11
T4	**Grass + OSW + CI**	177,18	3,52
T5	**Leaves + OSW**	744,65	2,27
T6	**Leaves + OSW + CI**	895,83	2,42

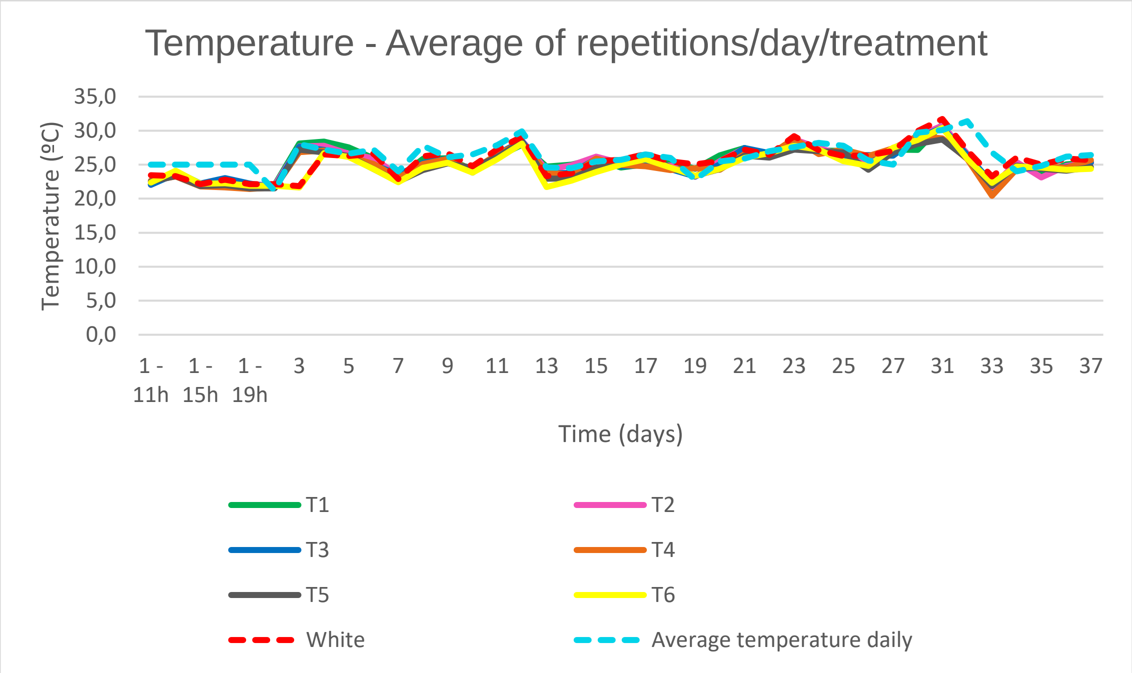
Source: The authors

Electrical conductivity (EC) represents the ability of a solution to conduct electrical current, to transport charges through the diffusion of ions, due to the action of the ionic concentration gradient (WERLANG, 2015). During the degradation of the waste, the generation of organic acids and ions such as phosphate, nitrate, and carbonate occurs, justifying the increase in conductivity (MOREIRA; BRAGA; FRIES, 2009).

For use in agriculture, it is recommended a 2 to 4 mS/cm conductivity (MASSUKADO; SCHALCH, 2010). Therefore, the values found were within the recommended range for soil used in vegetable cultivation, for example.

The temperature of the treatments followed the average local daily temperature (Figure 4). There was no temperature increase in the respirometers due to heat loss to the environment, as observed by Oliveira (2018) and Filogônio (2019) in small-scale composting.

Figure 4: Average temperature of treatment

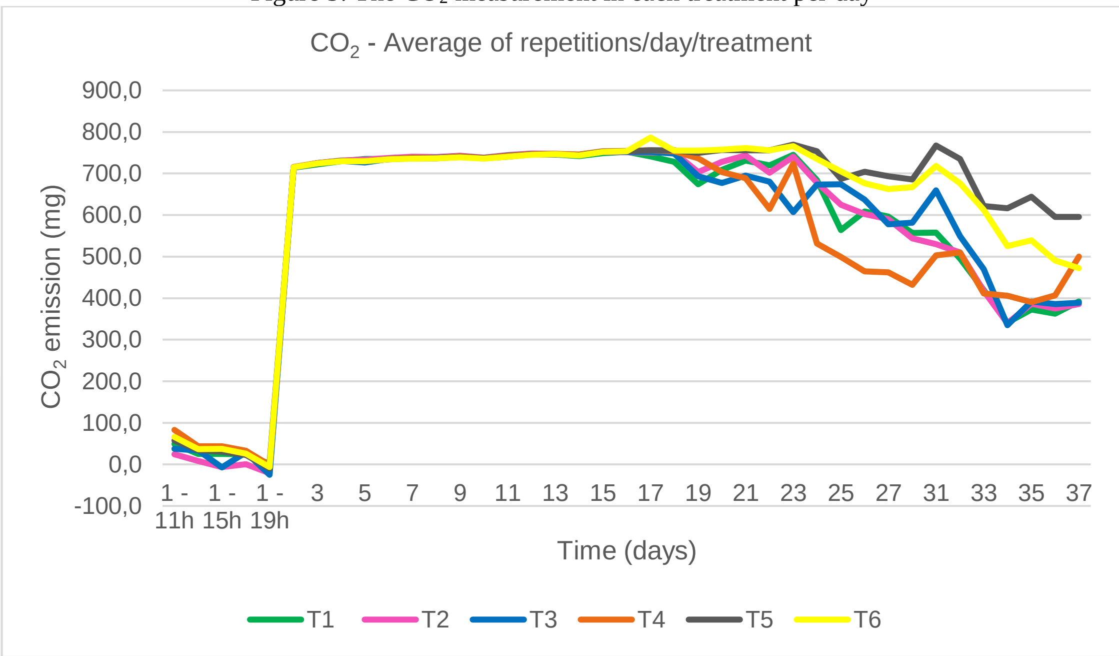


Source: The authors

The daily temperature values are from station A612 in the city of Vitoria, and the data was provided by the National Institute of Meteorology (Inmet) (2021).

For the construction of the CO_2 graph (Figure 5), Equation 4 was used, where Cx is the conductivity value measured daily in the samples.

Figure 5: The CO_2 measurement in each treatment per day



Source: The authors

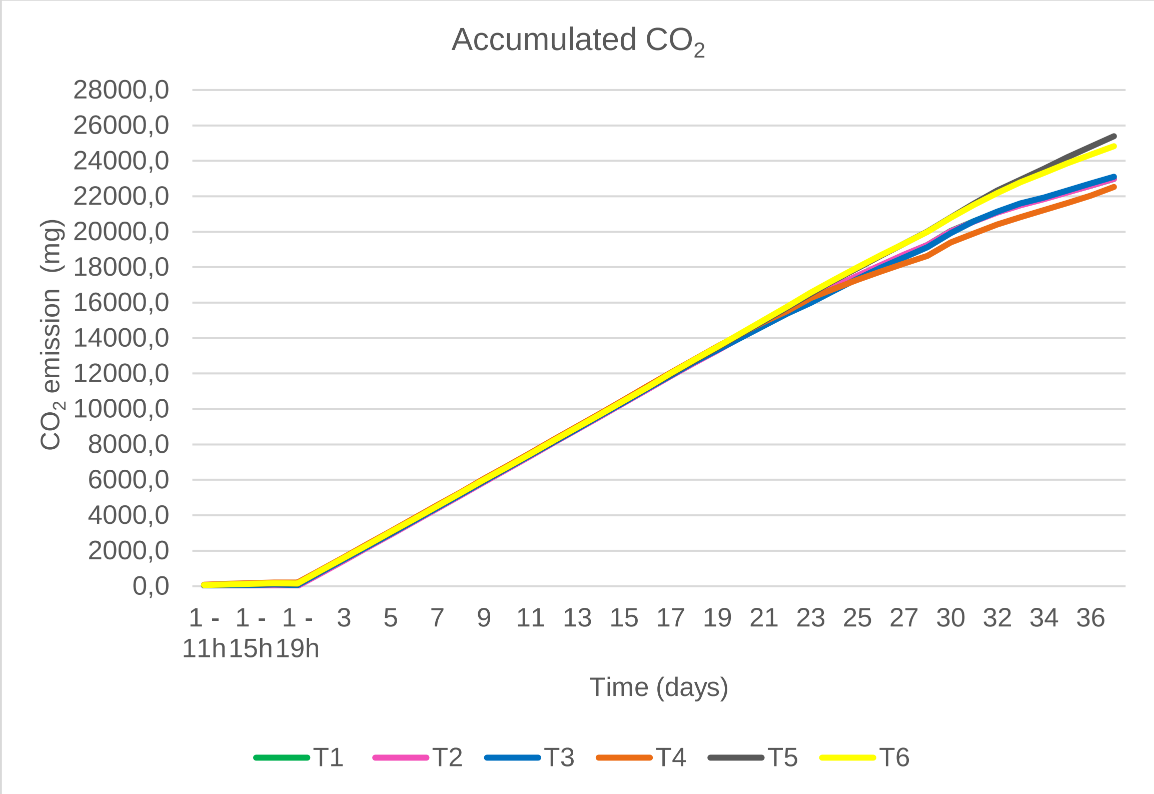
The CO_2 values were less than 100mg/CO_2 on the first day of the experiment and remained relatively low initially, indicating the adaptation phase of microorganisms to the environment. Shortly afterward, there is a peak, and daily production for both treatments is very high in the first 17 days, followed by a decline. Therefore, in the first days of the experiment, the most degradable organic matter broke.

The treatments with dried leaves, T5 and T6, had the highest CO_2 production, and the treatments with coconut, T1 and T2, should have shown the same results due to their high lignin content, but the result was similar to grass treatments.

The experiment happened for 37 days and showed CO_2 production, indicating the progress of degradation in the mixtures. Thus, the biodegradability of the different treatments, reflected in CO_2 generation, followed the same trend over time.

Figure 6 shows the cumulative CO_2 generation among treatments, i.e., the sum of the daily values produced.

Figure 6: Average accumulated CO_2 of treatments



Source: The authors

Regarding the reduction in waste mass, the initial weighing was performed in all respirometers, and the final weighing was carried out on the last day of the experiment (Table 6).

Table 6: Difference between initial and final masses of treatments and respective percentage of mass loss

Tratamentos	Massa inicial (g)	Média da massa final (g)	Porcentagem restante de resíduos (%)	Redução da massa (%)
T1		93,75	66,96	33,07
T2		100,8	72,00	28,00
T3		111,5	79,64	20,36
T4	140	113	80,71	19,29
T5		104,3	74,50	25,50
T6		102,3	73,07	26,93

Source: The authors

ISSN: 1696-8352

The reduction in waste mass occurs due to the aerobic degradation of organic matter, being converted into volatile compounds, CO_2, and water (PIMENTA et al., 2016). Treatment T1 showed the highest percentage of mass reduction, followed by treatment T2, both with coconut. Therefore, despite having compounds that are difficult to degrade, coconut shells had a higher mass reduction compared to other carbon sources used in this research.

5 CONCLUSION

In this study, the Bartha respirometric method measured through conductivity proved to be a suitable and effective approach for monitoring the biodegradability of waste materials that are not commonly used in composting processes. The experimental results demonstrated several key findings.

First, the initial pH of all waste mixtures ranged from 4 to 6, and by the end of the experiment, they had all shifted to a 9 pH range. This shift towards a more alkaline pH was favorable for composting, aligning with the desired pH range of 6 to 7.5 for composted materials. The compliance with recommended pH levels indicated that the treatment process was conducive to composting.

Regarding conductivity, initial measurements fell within the microSiemens (µS/cm) range, while final measurements showed an increase to the miliSiemens (mS/cm) range. This rise in conductivity was attributed to organic acids and ions generation, such as phosphate, nitrate, and carbonate, during the degradation process. The observed increase in conductivity aligned with typical composting trends and remained within the suitable range for agricultural utilization.

Throughout the 37 day experiment, the measurement of CO_2 production provided insights into the degradation progress. The initial days showed relatively low CO_2 production, indicative of an adaptation phase for microorganisms. Subsequently, a peak in CO_2 production was observed in the first 17 days, followed by a decline. This pattern suggests that the most degradable organic matter can break easily in the initial phase. While treatments involving dry leaves showed the highest CO_2 production, surprisingly, treatments involving coconut, which contains significant lignin, produced CO_2 levels similar to grass-based treatments.

Furthermore, the reduction in waste mass over the course of the experiment indicated successful aerobic degradation. The coconut-based treatment (T1) showed the highest reduction in waste mass, followed by the treatment with coconut and inoculant (T2). This result was intriguing, as coconut waste contains challenging-to-degrade compounds yet exhibited greater mass reduction compared to other carbon sources.

In the context of the presence or absence of the commercial inoculant (IC), no significant differences were observed between the treatments with and without IC. This suggests that the inoculant did not exert a pronounced impact on the biodegradation process under the experimental conditions.

To explore these findings further, it is recommended that future studies extend the experimental duration to observe the stability of the CO_2 production curve. Additionally, conducting trials with a wider variety of waste materials and conditions could provide more comprehensive insights into the application of the Bartha respirometric method for assessing the biodegradability of diverse waste streams.

In conclusion, this study demonstrated the feasibility of employing the Bartha respirometric method with conductivity measurements to assess the biodegradability of unconventional waste materials in composting processes. The results show the importance of monitoring pH, conductivity, and CO_2 production as indicators of successful composting and waste degradation.

ACKNOWLEDGEMENTS

The authors would like to thank Ifes (Federal Institute of Technology of Espirito Santo) and Fapes (Fundação de Apoio à Pesquisa do EspíritoSanto) for the financial support.

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