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STUDY OF COCOA FERMENTATION AND DRYING KINETICS

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Abstract: This project aimed to determine the fermentation kinetics in different environments, as well as the drying kinetics of post-fermentation almonds. Fermentation kinetics was determined by measuring temperature in a Datalogger, pH, total titratable acidity and soluble solids content. Drying kinetics was determined by the ratio of moisture, moisture, pH, titratable acidity and soluble solids content over time. The temperature reached higher values between the second and the fifth day (48-120 hours). In fermentation, the final pH value for the three treatments was favorable to the formation of chocolate flavor. The values of soluble solids content and acidity were similar. In drying, the three treatments concluded the process with pH values above 5.8, considered high. As for acidity, the curves showed fluctuations, but with similar behavior. Analyzing the drying kinetics, it can be observed that the pile treatment was practically the same as the plastic treatment, where the wood treatment was distinguished only in the initial phase of drying.
Keywords: Almonds, fermentation kinetics, drying kinetics.

INTRODUCTION

Cocoa has in its interior a large number of seeds coated with a white pulp and rich in sugars, which constitute the main raw material for the manufacture of chocolate (FERREIRA et al., 2013). Chocolate quality is influenced by several factors, including good agricultural processing and post-harvest practices. With regard to post-harvest, fermentation and drying are crucial steps for the formation of amino acids and short-chain peptides that are precursors of cocoa flavor (SCHWAN; WHEALS, 2004), being very important for the quality of the product.

According to Minifie (1989) apud Efraim (2004),

> [...] the fermentation and drying of cocoa seeds are of vital importance, and no further processing is capable of correcting failures in these steps. In the fermentation process, the system, the temperature of the environment and the mass, the pH and acidity of the pulp and cotyledon, the process time, the turning over of the mass as well as the microflora present are factors of great importance.

The aim of this work was to determine the fermentation kinetics in different environments (wooden box, plastic box and piles surrounded by banana leaves), as well as the drying kinetics of post fermentation almonds, in order to generate useful information in the optimization of these steps in situations of low production scales.

METHODOLOGY

Cocoa, of different varieties, was obtained in the fruit growing sector of the Federal Institute of Espírito Santo (IFES), campus in Alegre, and from local producers at the correct ripening point. The experiment was conducted on the premises of the Federal University of Espírito Santo.

The experimental design was DBC (Random Block Design), with cocoa from three different harvests constituting the blocks and the fermentation method (wooden box, plastic box and pile) constituting the experimental units.

Fermentations were carried out according to the methodology recommended by CEPLAC (Ferreira et al., 2013) in which the cocoa mass is kept under anaerobic conditions for 48 hours, followed by turning the mucilaginous pulp every 24 hours until the end of fermentation. approximate time is 7 days. Fermentations were carried out in a chamber with a controlled temperature of 33ºC and 80% humidity.

Fermentation kinetics were determined by monitoring temperature, pH, acidity and soluble solids content every 12 hours. The

temperature was determined in a datalogger (Novus brand, FieldLogger model), throughout the fermentation process. Samples of 30 g of cocoa were taken, added with 30 g of distilled water followed by stirring for approximately 2 minutes. Separated in triplicate, the aliquot of pulp solution and distilled water was used to determine pH, total titratable acidity and soluble solids content.

For pH analysis, a direct reading was performed in a potentiometer (Metrohm brand, model 826 pH mobile), at room temperature according to IAL (2008). The total titratable acidity (TT) analysis was performed from an aliquot of the suspension obtained by titration with 0.1 N NaOH, using phenolphthalein as an indicator (IAL, 2008). Acidity was expressed in mEq of NaOH/100 g of cocoa. The analysis of soluble solids content was performed in a refractometer.

Drying was carried out in a tray dryer at a temperature of 40°C under constant air flow until a final moisture content close to 7.5%. The drying kinetics was determined by monitoring the almond weight every hour, moisture, pH and total acidity every hour throughout the almond drying process. Samples of 30 g of dry cocoa were taken, added to 30 g of distilled water, followed by grinding and stirring for 15 seconds in a mixer. After filtered through a sieve, the samples were separated in triplicate, and aliquots were removed from the solution for analysis, and the pH and total acidity were determined according to the same methodology described above (fermentation kinetics step). However, in drying, acidity was expressed on a dry basis (mEq of NaOH/100 g of dry mass).

By monitoring the mass of almonds during drying, the moisture ratio (RU) was calculated using Equation 1 and then the drying curve was adjusted according to the Lewis (Equation 2), Page (Equation 3) models) and Henderson and Pabis (Equation 4):

$$RU = (U(t) - Ueq)/(Ui - Ueq) = (m(t) - meq)/(mi-meq)/(mi - meq)$$

(Equation 1)

In which:
U(t)= humidity in time: t
Ueq= equilibrium moisture
Ui= initial moisture
mt= mass in time: t
meq= balance mass
mi= initial mass

$$RU = exp(-kt)$$ (Equation 2)

$$RU = exp(-ktN)$$ (Equation 3)

$$RU = a.\ exp(-kt)$$ (Equation 4)

In which:

k, n and a are model parameters and t is the drying time, in minutes. The adjusted models were compared by the adjusted coefficient of determination, R^2_{adj} and by the standard error of the regression (S). The computer program used to construct the drying curves and adjust the mathematical models was SigmaPlot 11.0.

Humidity was determined by the method of direct drying in an oven (Tecnal brand, model TE-393/1) at 105 °C until constant weight (IAL, 2008). The pH and total acidity were determined according to the same methodology described above (fermentation kinetics step).

At the end of drying, the cutting test was performed according to the methodology described by Efraim et al., 2010. Samples of fermented and dried almonds (containing about 50 almonds each sample) were randomly taken from each fermentation performed, which were sectioned longitudinally and individually evaluated according to color, almond defects and cotyledon compartmentation.

RESULTS

On average, 80 kg of cocoa pods were processed per fermentation, with only about 20% of the weight consisting of seeds with adhered pulp. In all media, the seeds with adhered mucilaginous pulp were covered with banana leaves, left that way until the end of fermentation. The results are expressed as mean values obtained in the three repetitions. Fermentation time was an average of 7 days.

Figure 1 shows the evolution of temperature throughout fermentation.

According to Mattietto (2001), fluctuations in temperature curves occur due to the metabolic activity of microorganisms. According to the graph, it can be seen that the temperature reached higher values between the second and the fifth day (48-120 hours), and the treatment in the wooden trough reached its maximum temperature value (39.2 °C) on the 3rd day (87h of fermentation), the plastic (38.03 °C) on the 4th day, and the pile (41.9 °C) on the 5th day. It is worth noting that the pile treatment presented a different behavior compared to the other treatments. This may have occurred due to the temperature chamber airflow system, which may have compromised the exothermic reactions at the beginning of fermentation, as this medium was more exposed to ambient conditions.

According to Zamalloa (1994), good cocoa fermentations must reach 45 to 48°C in approximately 72 hours after starting the process. However, temperatures vary according to the cocoa mass present in the fermentation media and the environmental conditions.

Figure 2 shows the means of the monitoring curves of the physicochemical parameters analyzed during fermentation for each fermentation medium.

The pH variation during fermentation is related to the changes that occur in the pulp during the process (SOARES, 2001). Low pH values of the pulp in the initial phase is mainly due to the citric acid that is naturally present in the pulp of the cocoa seed. As a result of the activity of microorganisms in the pulp, citric acid is replaced by less dissociated acids such as lactic and acetic acid (SOARES, 2001). In addition, the acetic acid undergoes volatilization and is drained with the liquefaction of the pulp, during the successive turnings that are carried out, in addition to being oxidized to carbon dioxide (EFRAIM, 2009), raising the pH of the medium.

It is noteworthy that during the fermentation period, the pulp undergoes changes in pH, resulting from microbial activity, which causes the seed to die (BATALHA, 2009; MINIFIE, 1989). Dias (1987) reported that cocoa beans whose final pH is less than 4.5, have a low potential in the formation of chocolate flavor while at final pH values above 5.0 this potential is significantly increased. In other words, the final pH values found in this study show that the fermentations carried out resulted in almonds with potential to form the chocolate flavor.

The curves of average acidity in the three treatments showed fluctuations mainly between 50 and 120h of fermentation, with a decline at the end of the process, reaching values very close to the end of fermentation. In healthy cocoa seeds, the high acidity verified initially is due to the pulp and the important presence of citric acid associated with the low level of oxygen in the initial phase of fermentation (SCHWAN & WHEALS, 2004). The production of lactic and acetic acid occurs during the first days of cocoa seed fermentation, mainly between the 3rd and 5th day, and is later absorbed by the seeds (RAMOA JR, 2011). This explains the rise and subsequent decline in the acidity curve obtained, since the parameters were evaluated in the fermentation environment and not inside the seeds.

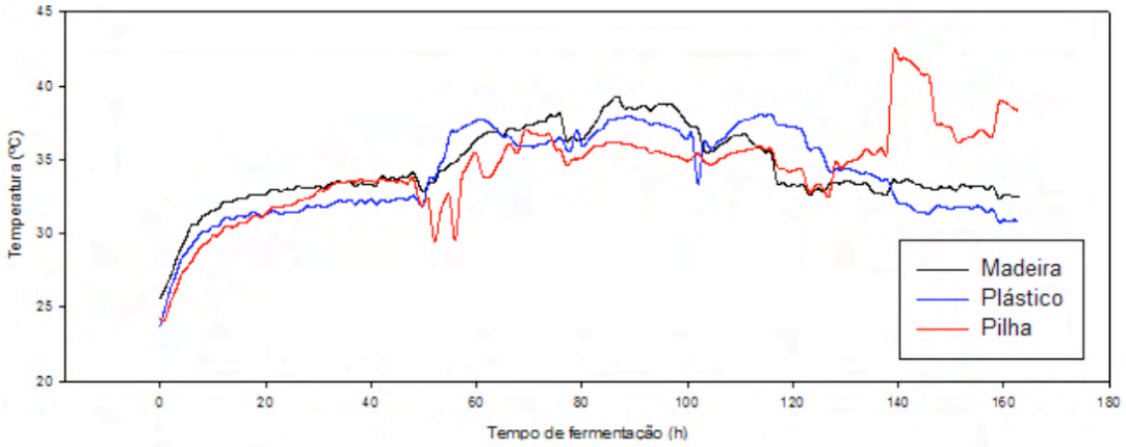
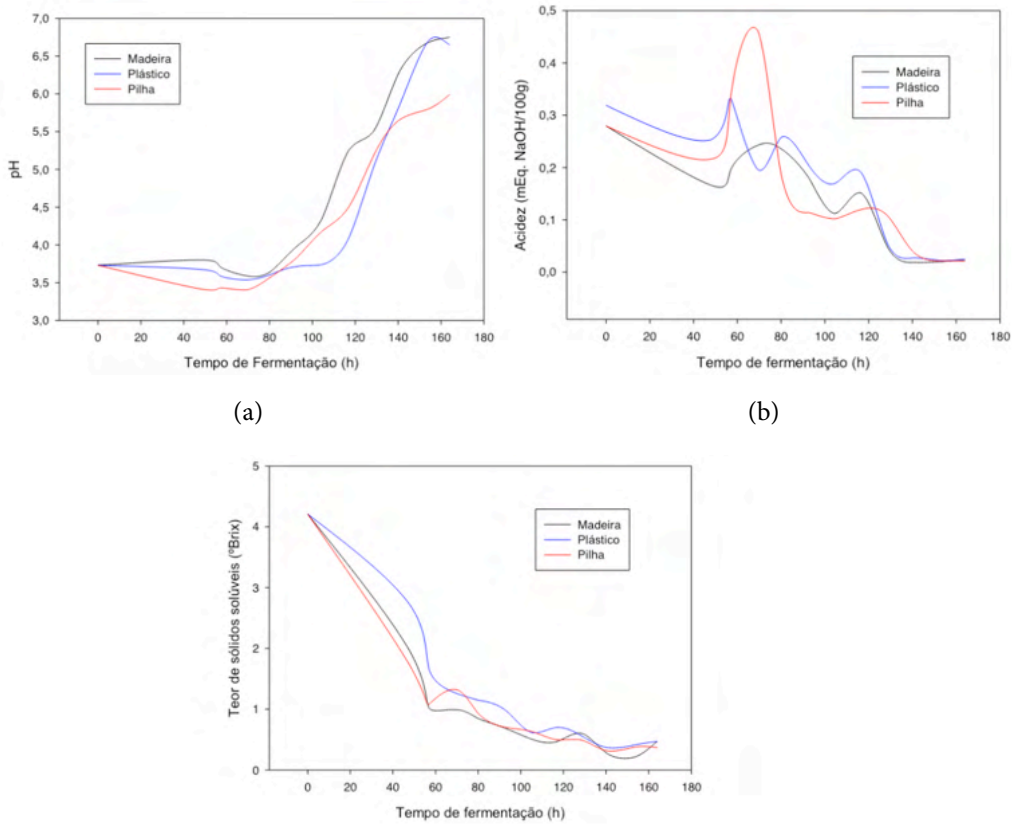


Figure 1. Monitoring temperature during fermentation

Source: Own authorship



(a) (b)

(c)

Figure 2. Values of pH (a), acidity (mEq. NaOH/100g) (b) and soluble solids content (°Brix) (c) obtained during the fermentation processes.

Source: Own authorship

Madeira = Wood

Plástico = Plastic

Pilha = Battery

The refractive index values at the end of the process were 0.47 ºBrix for wood and plastic, and 0.37 ºBrix for pile. The behavior of the curves showed a sharp drop in the first 48h, but without many significant fluctuations in the curves during the rest of the fermentation. This drop in the first hours of fermentation is mainly due to the transformation of sugars into alcohol (ZAMALLOA, 1994). It must be noted that the low values of total solids obtained are due to the fact that the samples in which the readings were taken are diluted.

During the last days of fermentation, cutting tests were carried out on the seeds in order to assess the fermentation stage. Seeds that had purplish cotyledons indicated that fermentation had not evolved enough. As the process progresses, the seeds tend to have a brownish color, with darker edges, and a characteristic aroma, in addition to the appearance of a viscous furrow on the cotyledons. Thus, in addition to temperature control, the cutting test was a factor in evaluating the stage at which the fermentation was at, being an important criterion in deciding when to stop the fermentation process.

Figure 5 shows the means of the monitoring curves of the physical-chemical parameters analyzed during drying for each fermentation medium.

It is noteworthy that the methodologies for obtaining samples for physicochemical analysis were different between the fermentation and drying stages, since the fermentation stage sought to analyze changes in the fermentation medium and in drying the objective was to evaluate the characteristics of the cotyledons (internal part of the seeds).

The treatments started drying with different pH values, but showed similar behavior in the curves. At the end of drying, only the treatment in the pile showed a difference in the final value (5.99) compared to the treatment in wood and plastic (6.75 and 6.65 respectively). Higher pH values for dried almonds, if compared to the values of fermented almonds, was expected due to losses due to volatilization of acetic acid in the drying process (MATTIETTO, 2001).

As for acidity, the curves of the three treatments also showed fluctuations, but with similar behavior. According to Mattieto (2001), it was expected that during drying the almonds presented a drop in the total acidity value, due to the evaporation of acetic acid in the process, which was not observed in the present study, where the behavior of the acidity values were consistent with those of pH. According to the work, artificial drying, being faster than natural drying, does not allow the volatilization of compounds formed during fermentation, such as acetic acid, leading to a decrease in pH and an increase in total titratable acidity, impairing sensorially the products obtained.

As for the soluble solids content, all treatments showed high fluctuations throughout the process, and in the end there was an intense increase in values, which was to be expected, due to water evaporation and consequent concentration of solids present.

Studies with fermented and dried almonds are still scarce, which makes analysis and comparison of results difficult. Furthermore, there is the fact that in solid fermentations there is not so much homogeneity in the samplings, which influences the results of the analyzes carried out both for fermentations and for dryings.

At the end of drying, a cutting test was also performed. Fifty seeds were removed from each medium, and a longitudinal cut was made to carry out the analysis.

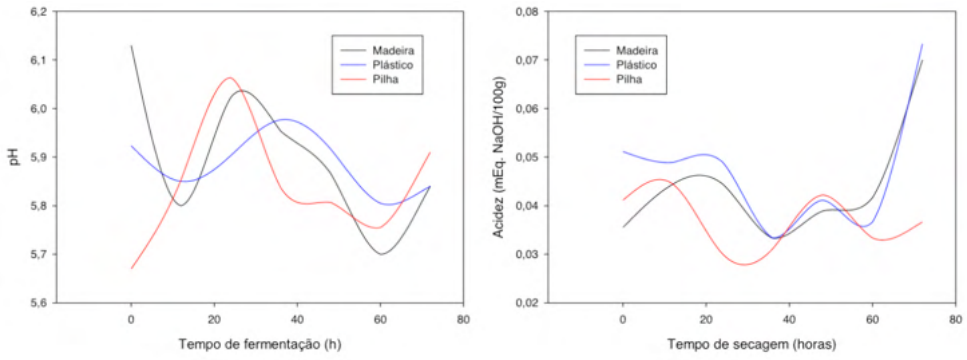
Before fermentation, the cotyledons of cocoa pod seeds have an intense violet color. During fermentation and drying, the various biochemical reactions that occur

(a) (b)

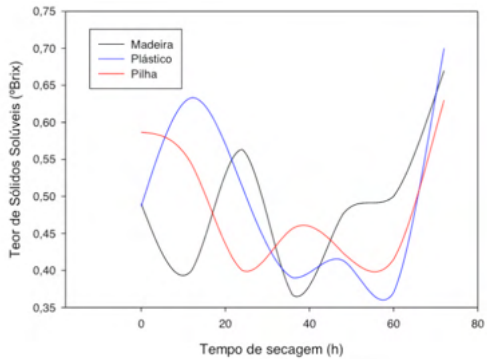
Figure 3. Cutting test at the beginning (a) and last days (b) of fermentation.

Source: Own authorship.

Figure 4. Cutting test at the end of fermentation for the three treatments. Source: Own authorship.



(a) (b)



(c)

Figure 5. Values of pH (a), acidity (mEq. NaOH/100g) (b) and soluble solids content (ºBrix) (c) obtained during the drying processes.

Source: Own authorship

Figure 6. Cutting test at the end of drying. Source: Own authorship.

		Quantity (%)		
		Wood	Plastic	Pile
Coloring	Brown	67,33	61,33	66,67
	A little brown	19,33	24,67	17,33
	Violet	4,00	5,33	6,67
	White	8,67	8,67	6,00
	Slate	0,67	0,00	3,33
	Total	100,00	100,00	100,00
Defects	Musty	0,00	0,00	0,00
	Infested	0,00	0,00	0,00
	Germinated	0,00	0,00	0,00
	Cuttlefish	0,00	0,00	0,00
	Brittle	100,00	100,00	100,00
Compartmentation	Good	96,67	90,00	97,33
	Partial	3,33	10,00	2,67
	Without	0,00	0,00	0,00
	Total	100,00	100,00	100,00

Table 1. Cutting test of fermented and dried cocoa beans.

inside the seeds lead to the oxidation of phenolic compounds and, consequently, to the darkening of the cotyledons, which start to show a brown color (EFRAIM, 2010) in addition to their compartmentalization. Thus, the high number of measured brown almonds, and the high number of almonds with good compartmentalization, indicate that there was good fermentation in all treatments.

The drying curves fitted in the proposed models are shown in the figure 7 below.

Table 2 shows the values of the adjusted parameters, the adjusted coefficient of determination and the standard error of the regression for each model. The higher the adjusted coefficient of determination (R^2_{adj}) and the smaller the standard error of the regression (S) the better the mathematical model for drying.

Comparing the kinetics of the three treatments using the Page model (figure 5), it can be seen that the piles treatment was practically the same as the plastic treatment. The wood treatment differed from the others only in the initial phase of drying, when it was a little slower. The three treatments had similar behavior in relation to the final drying time.

The almonds were left to dry until moisture was less than 7%. In Figure 9 are found the moisture averages of the 3 repetitions.

According to Normative Instruction No. 57 of 2008, fermented and dried cocoa beans must have a moisture content below 8%, so that the beans used in the study were within the standard.

CONCLUSION

All media presented, at the end of the fermentation process, pH values within a considerable range as ideal for the development of chocolate flavor. The three media also presented acidity curves according to what is found in the literature.

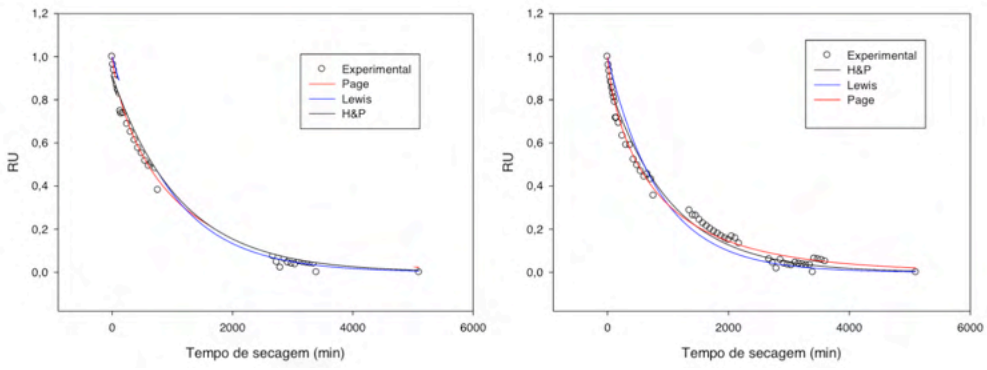
At the end of drying, all treatments had a pH above 5.8. Fluctuations in the curves of the results of physical-chemical analyzes in the drying stage may have been caused by the heterogeneity of sampling, common in solid fermentations.

The high number of brown almonds verified in the cutting test indicates that there was good fermentation in all treatments.

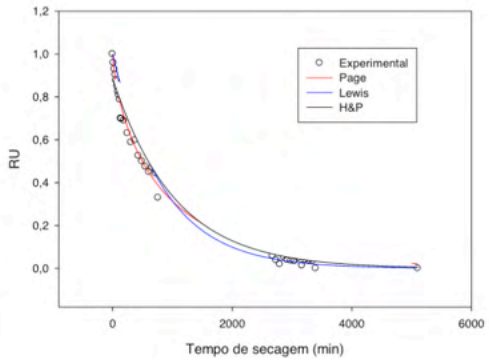
The model that presented the highest value of the adjusted coefficient of determination and the lowest value of the standard error of the regression, in all treatments, was that of Page, indicating that the model is adequate to represent the operation.

By the analysis of variance (ANOVA), presented in Annex I, there was no significant difference between the treatments at 5% probability, when analyzed at the beginning and end of each process.

With the research, it is undeniable that information was generated related to the dynamics of the cocoa fermentation and drying process carried out on a small scale, as well as on the characteristics of the cocoa mass in order to disseminate them in academia and also to cocoa producers in the region of Alegre that will be able, through processing, to add value to their production.



(a) (b)



(c)

Figure 7 - Experimental drying curve of fermented almonds adjusted by the models tested in a wooden trough (a), plastic trough (b), and in piles covered with banana leaves (c).

Source: Own authorship

Middle	Model	k	a	n	R^2_{adj}	S
	Lewis	0,9863			0,9727	0,0531
Wood	H&P	0,0009	0,9113		0,9872	0,0368
	Page	0,0046		0,7851	**0,9909**	**0,031**
	Lewis	0,0012			0,963	0,0608
Plastic	H&P	0,001	0,8932		0,9827	0,042
	Page	0,0069		0,7418	**0,992**	**0,0283**
	Lewis	0,0012			0,9586	0,064
Pile	H&P	0,001	0,8878		0,9803	0,0441
	Page	0,0073		0,7334	**0,9901**	**0,0315**

Table 2. Adjusted parameters, adjusted coefficient of determination and standard error of regression for models of drying kinetics in different fermentation media.

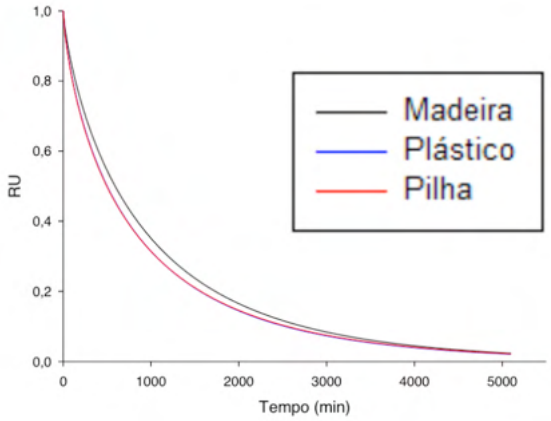


Figure 8. Comparison of the kinetics of all treatments by the Page model

Source: Own authorship

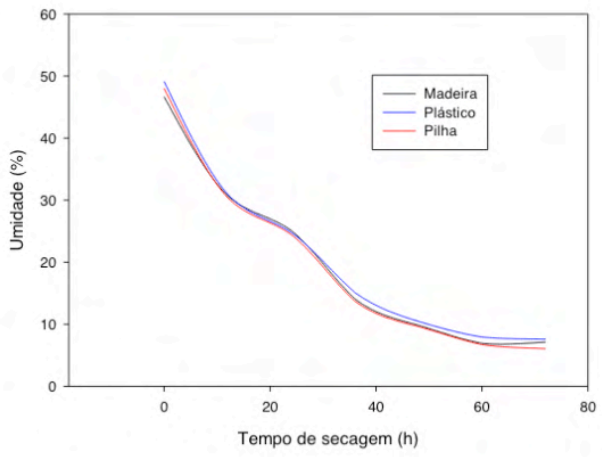


Figure 9. Humidity values (%) during the drying process

Source: Own authorship

Madeira = Wood

Plástico = Plastic

Pilha = Pile

Tempo = Time

Tempo de secagem = Drying time

Unidade = Unit

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