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# QUALITY OF NATURAL AND PULPED COFFEE AS A FUNCTION OF TEMPERATURE CHANGES DURING MECHANICAL DRYING

Pedro Damasceno de Oliveira<sup>1</sup>, Marco Antônio Martin Biaggioni<sup>2</sup>, Flávio Meira Borém<sup>3</sup>, Eder Pedroza Isquierdo<sup>4</sup>, Mariana de Oliveira Vaz Damasceno<sup>5</sup>

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ABSTRACT: This research evaluated the sensory quality of processed and dried coffee beans in different ways. Two types of processing were used: dry and wet, besides seven drying methods: drying in yard and mechanical drying with heated air at 50 °C until coffee reached 30% (w.b.) moisture content, followed by drying with air heated to 35 °C until reaching 11% (w.b.) moisture content; drying in fixed-layer dryers with heated air at 45 °C until coffee reached 30% moisture content, followed by drying with heated air at 35 °C until reaching 11% (w.b.) moisture content; and drying in fixed-layer dryers with heated air at 40 °C until coffee reached 30% (w.b.) moisture content, followed by drying with heated air at 35 °C until reaching 11% (w.b.) moisture content; drying in fixed-layer dryers with heated air at 35 °C until coffee reached 30% (w.b.) moisture content, followed by drying with heated air at 50 °C until reaching 11% (w.b.) moisture content; drying in fixed-layer dryers with heated air at 35 °C until coffee reached 30% (w.b.) moisture content, followed by drying with heated air at 45 °C until reaching 11% (w.b.) moisture content; drying in fixed-layer dryers with heated air at 35 °C until coffee reached 30% (w.b.) moisture content, followed by drying with heated air at 40 °C until reaching 11% (w.b.) moisture content. The mechanical drying system consisted of three fixed-layer dryers, allowing the control of temperature and drying flow. Coffee was tasted according to the evaluation system proposed by the Specialty Coffee Association of America (SCAA). Physicochemical composition and physiological quality of the beans were analyzed, involving: grease acidity, potassium leaching, electrical conductivity, color and germination. The results show that pulped coffee is more tolerant to drying than natural coffee, regardless of how it was dried.

**Index terms**: Post harvest, sensory analysis, alternate drying.

# QUALIDADE DO CAFÉ NATURAL E DESPOLPADO EM FUNÇÃO DA ALTERNÂNCIA DE TEMPERATURA DURANTE A SECAGEM MECÂNICA

RESUMO: Objetivou-se neste trabalho avaliar a qualidade de grãos de café processados de duas formas diferentes (via seca e via úmida) e sete métodos de secagem: secagem em terreiro; e secagem mecânica com ar aquecido em secadores: a 50 °C até o café atingir 30% de teor de água (TA), prosseguindo-se com ar aquecido a 35 °C até atingir 11% (b.u) de TA; a 45 °C até o café atingir 30% (b.u) de TA, prosseguindo-se com ar aquecido a 35 °C até atingir 11% (b.u) de TA; a 40 °C até o café atingir 30% (b.u) de TA, prosseguindo-se com ar aquecido a 35 °C até atingir 11% (b.u) de TA; a 35 °C até o café atingir 30% (b.u) de TA, prosseguindo-se com ar aquecido a 50 °C até atingir 11% (b.u) de TA; a 35 °C até o café atingir 30% (b.u) de TA, prosseguindo-se com ar aquecido a 45 °C até atingir 11% (b.u) de TA; a 35 °C até o café atingir 30% (b.u) TA, prosseguindo-se com ar aquecido a 40 °C até atingir 11% (b.u) de TA. A análise sensorial foi realizada utilizando-se a metodologia proposta pela Associação Americana de Cafés Especiais (SCAA). As demais análises realizadas foram composição físico-química e acidez graxa, lixiviação de potássio, condutividade elétrica, cor e germinação. Para a análise estatística utilizou-se o programa computacional Assistat 4.0. A secagem em terreiro proporcionou melhor qualidade fisiológica e sensorial dos grãos de café, quando comparada com a secagem em secadores, mas o uso das temperaturas 35/40 °C e 40/35 °C apresentou resultados semelhantes à secagem em terreiro. Além disso, o café despolpado apresentou melhor qualidade fisiológica que o café natural.

Termos para indexação: Pós-colheita, análise sensorial, secagem alternada.

# 1 INTRODUCTION

Coffee is a product of great importance in global agribusiness. According to the International Coffee Organization - ICO (2016), raw coffee consumption was in the order of 145 million bags in 2016.

There are two processing methods for coffee: dry and wet. In moist processing, three types of coffee can be produced. Peeled coffees, of which the remaining husk mucilage is not removed from the beans; pulped coffees, originating from mechanically peeled fruits, and the remaining mucilage is removed by fermentation; and

¹Syngenta Proteção de Cultivos Ltda. - Rua Alameda das Magnólias, 500 - 37.170-000 - Boa Esperança -MG - damascenoen@yahoo.com.br ²Universidade Estadual Paulista "Júlio de Mesquita Filho" - Faculdade de Ciências Agronômicas de Botucatu - Departamento de Engenharia Rural - Fazenda Experimental Lageado -Cx.P. 237 - 18.603-970 - Botucatu - SP - biaggioni@fca.unesp.br ³Universidade Federal de Lavras/UFLA - Departamento de Engenharia/DEG - Cx.P. 37 - 37.200-000 - Lavras - MG - flavioborem@deg.ufla.br

<sup>&</sup>lt;sup>4</sup>Universidade do Estado do Mato Grosso/UEMT - Departamento de Agronomia - Av. Santos Dummont, s/n - Santos Dummont 78.200-000 - Cáceres - MT - eder.isquierdo@yahoo.com.br

<sup>&</sup>lt;sup>5</sup> Rua Alameda das Magnólias, 500 - 37.170-000 - Boa Esperança - MG - marinavaz 1@yahoo.com.br

those demucilated, in which the mucilage is mechanically removed. In dry processing, intact fruits are submitted to drying, without exocarp removal.

The chemical composition of raw coffee depends on the processing used (BORÉM, 2008; BYTOF et al., 2005; KNOPP; BYTOF; SELMAR, 2006). In general, natural coffees give rise to more full-bodied and sweet beverages compared to pulped coffees, which have more desirable acidity (SANTOS; CHALFOUN; PIMENTA, 2009). Acidity in coffee beans has been pointed out as a good indicator of product quality, in which small amounts of organic acids are necessary to confer essential acidity to the coffee beverage (DA SILVA et al., 2009).

The sensory evaluation method of the Specialty Coffee Association of America has been emphasizing the importance to evaluate the quality of special coffees. The method is based on a quantitative descriptive sensory analysis of the beverage, performed by a selected and trained team of panelists, using an unstructured scale from 6 to 10 for the evaluation of powder fragrance, defects, acidity, bitterness, flavor, residual taste, astringency and beverage body, with final evaluation of the overall quality and quality of coffee according to the terminology presented by Lingle (2011).

In addition to sensory evaluation, the physicochemical evaluation of coffee beans can become a valuable tool for assessing beverage quality. Taveira et al. (2015) observed biochemical changes during processing related to germination metabolism, whose extent depends on the treatment, whether wet or dry. The authors, however, did not correlate it with drying methods.

The drying rate has a significant effect on the quality of agricultural products and, according to Burmester; Eggers (2010), it is mainly influenced by drying air temperature, but also by drying air flow, relative humidity, among other factors. These parameters are not independent, simultaneously influencing the drying process.

Higher drying rates caused by high temperatures can lead to damage to coffee quality due to damage to cell membranes (MARQUES et al., 2008; BORÉM et al., 2008b). Borém et al. (2008b) verified, through ultrastructural scanning electron microscopy, that natural and pulped caffeine endosperm, during drying at 40 °C and in yard, maintained the integrity of the cell membranes and that these membranes were damaged only between moisture contents of 30% and 20% (w.b.), when natural and pulped coffee were dried at a temperature of 60 °C.

High water reduction rates are desirable in reducing the risk of fermentation in early drying stages, as well as energy consumption. However, when using the technology currently available for coffee drying, the increase in drying rate is obtained by increasing the temperature. However, it has been proven by several scientific reports that, when subjected to drying temperatures above 40 °C, coffee suffers serious damage to the cell membrane system of the seed endosperm, with negative reflections on beverage quality, when these are used in the final stages of mechanical drying.

The objective of this study was to evaluate the effects of different processing and drying methods on the physiological and physicochemical quality of coffee beans, analyzing their interrelationship with beverage quality.

# 2 MATERIAL AND METHODS

# 2.1 Experimental procedure

The experiment was carried out with parchment coffee (*Coffea arabica* L. cv. Catuai 62), harvested on Santa Clara farm, which has an altitude of 1,270 meters, located 10 km from the city of Carmo de Minas (Latitude: 22 ° 4'14.80 "South; Longitude: 45 ° 7'17.77" West), in the mountain range of Mantiqueira. The harvested fruits were taken to the city of Lavras - Minas Gerais, and transportation took about 2 hours.

After arriving at the Federal University of Lavras, coffee was processed through dry (natural) and wet (pulped) methods, separating only the parchment fruits. After processing, coffee was dried under seven different conditions (Table 1).

Afterdrying, physiological, physicochemical and sensory analyses were carried out at the Post-Harvest Technology Pole Laboratory of the Federal University of Lavras. Figure 1 summarizes the whole experimental process.

# 2.2 Dry processing

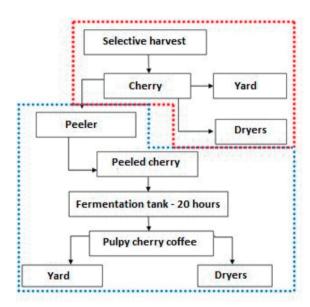
For dry coffee processing, which results in the natural coffees, the fruits were washed and separated hydraulically, by bulk density difference, for the removal of the float and dried fruits present in the plot. Subsequently, the ripe fruits were again manually selected to ensure sample uniformity with respect to the maturation stage. After this procedure, a portion of the natural coffee was taken to the yard for complete drying and the other portion was subjected to mechanical drying in the dryers.

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**TABLE 1 -** Experimental design.

Processing	Drying Method
	OnGround
	35*/40 °C**
	35*/45 °C**
NATURAL/ PULPED	35*/50 °C**
	40*/35 °C**
	45*/35 °C**
	50*/35 °C**

<sup>\*</sup>Initial drying temperature of each treatment.\*\*Drying temperature from the moment the sample reached the mass relative to the moisture content of  $30\% \pm 2\%$  (w.b.), remaining the same until coffee reached 11% (w.b.).



**FIGURE 1** - Flowchart used to obtain the raw material (Dotted lines in red indicate dry processing; Dotted lines in blue indicate wet processing).

# 2.3 Wet processing

For the processing of wet coffee, mature fruits from selective harvesting were again manually selected and mechanically peeled in a green separator (Pinhalense Ltda., Eco – 6). After peeling, the coffee was submitted to fermentation in water to remove the mucilage, under ambient conditions, with an average temperature of 20 °C, for 20 hours. After this period, the parchment coffees were washed with water until the mucilage was completely removed. When the mucilage was completely removed, a portion of the parchment coffee was taken to the yard for complete drying and the other portion was subjected to mechanical drying in the dryers.

# 2.4 Drying on ground

For drying on ground, coffee remained under ambient conditions after processing. One type suspension was used for drying on ground. These coffees were sprayed in fine grain-to-grain layers and, with drying, the layer was folded, according to the methodology proposed by Borém et al. (2008).

During the drying period, the maximum temperature was  $31.8\,^{\circ}$ C, the minimum temperature was  $13.15\,^{\circ}$ C, average room temperature was  $18.85\,^{\circ}$ C, precipitation was  $1.4\,^{\circ}$ cm and relative humidity was  $58.6\,^{\circ}$ %, which lasted from August  $16\,^{\circ}$ to  $27,\,2013$ . Both natural and pulped coffees remained under these conditions until reaching the moisture content of  $11\pm0.2\%$  (w.b.).

# 2.5 Drying in dryer

The mechanical drying plots were made to three dryers (Figure 2) with a fixed layer, which allow flow and temperature (T) control of the drying air with precision, through an electronic panel. The bean layer reached the thickness of 30 cm for natural coffee and 20 cm for pulped coffee.

The air flow was controlled at 20 m<sup>3</sup> min<sup>-1</sup> m<sup>-2</sup>, corresponding to a speed of 0.33m s<sup>-1</sup> (SILVA, 2000).

The transition from one temperature to the next, in the case of treatments with heated air at 50/35 °C, 45/35 °C, 40/35 °C, 35/50 °C, 35/45 °C and 35/40 °C, was determined as follows:

The moisture content of the beans during drying was controlled from the initial moisture content of coffee on ground, which made it possible to monitor the mass variation in the respective samples. The moisture content of coffee was determined when the fruits were subjected to a constant temperature of 105 °C for a period of 24 hours (BRASIL, 2009). After this period, their humidity was determined by the difference of the initial and final people.

To determine the time of transition of the air temperature, each tray containing the experimental plot was weighed every hour, and the moisture content was determined by mass difference applying equations 1 and 2. When each drawer reached the relative mass at the moisture content of  $30\% \pm 2\%$  (w.b.), the temperature was changed, remaining until the coffee reached 11% (w.b.).

$$Mf = Mi - \left(\frac{Mi \times PQ}{100}\right)$$

$$PQ = \left[\frac{(Xi - Xf)}{(100 - Xf)}\right] \times 100$$

Where:

Mf: final mass (kg);

Mi: initial mass (kg);

PQ: percentage of break (%);

Xi: initial moisture content (% w.b.); Xf: final moisture content (% w.b.). After drying and cooling, parchment and natural coffee remained stored in polyethylene bags in an environment with a temperature of 10 °C and 50% relative humidity, and it was only used when the physiological, physicochemical and sensory to evaluate the quality of coffee, which happened after a minimum of 90 days of rest, minimum time for coffee to have consolidated its sensory attributes (BORÉM, 2008).

# 2.6 Characterization of coffee quality

# 2.6.1 Sensory analysis

The sensory analysis was performed at the Nutrade Quality Laboratory, located in the city of Varginha/MG. For this purpose, portions of the bean samples classified in the above 16 sieves were used, with discs of malted and defective grains, according to Normative n°8 (BRASIL, 2003).

Sensory analysis was performed by two Certified Special Coffee Panelists (SCAA-Certified Cupping Panelists). The sensory analysis protocol of the SCAA was used, according to the methodology proposed by Lingle (2011), for the sensory evaluation of special coffees, with the assignment of scores, in the range of 6 to 10 points for fragrance, acidity, body, taste, sweetness, uniformity, clean cup, balance and finish. The roast was made with coloration corresponding to 58 points of the Agtron scale, for the whole bean, and 63 points for the ground bean, with a tolerance of  $\pm 1$  point. To obtain the ideal toasting point, the samples were standardized for weight (110 g), bean size (sieve 16 and above), as well as temperature and toasting time (between 8 and 12 minutes), according to Normative n° 8 (BRASIL, 2003).

At each sensory evaluation, five representative cups of coffee were sampled, and a sensory analysis session was conducted for each replicate, totaling three replicates for each treatment. Due to the presence of different sensory characteristics, the sensory analysis of natural and pulped coffee was carried out separately, in order to minimize any possible negative or positive interference. The final results of the sensory evaluation were constituted by the sum of all the attributes.

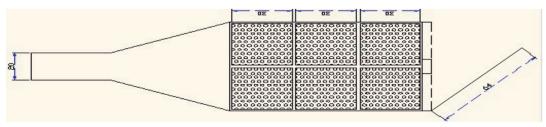


FIGURE 2 - Top view of the dryers used in the experiment.

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# 2.6.2 Physiological and physicochemical analyses

Physiological and physicochemical analyses were performed in the Laboratory of Seed Analysis of the Department of Agriculture of the Federal University of Lavras. For the physiological analyses, four sub-samples of beans with no apparent defects were used, for each replication of the respective treatments.

# 2.6.2.1 Germination test

It was carried out with four sub-samples of 50 seeds, distributed in germinated paper with water equivalent to two and a half times the mass of the dry substrate and placed to germinate at 30 °C. The evaluations were performed 30 days after sowing, according to the Rules for Seed Analysis (BRASIL, 2009), and the results were expressed as a percentage.

# 2.6.2.2 Electrical Conductivity

The electrical conductivity of the raw beans was determined by the methodology proposed by Krzyzanowski et al. (1991). Four replicates of 50 beans of each plot were used, which were weighed to an accuracy of 0.001g and immersed in 75 mL of distilled water inside 180 mL plastic cups. These containers were then taken to BOD with forced ventilation regulated to 25 °C for five hours, and the electrical conductivity of the soaking water in BEL W12D was read. With the data obtained, the electrical conductivity was calculated by Equation 3, expressing the result in µS cm<sup>-1</sup> g<sup>-1</sup> of beans.

$$CE = \frac{Value \, read(\frac{\mu S}{cm})}{Mass(a)}$$

# 2.6.2.3 Potassium leaching

The leaching of potassium ions was carried out in the raw beans, according to the methodology proposed by Prete (1992). After the electrical conductivity reading, the solutions were subjected to the determination of the amount of leached potassium. The reading was carried out in a flame photometer (Digimed NK-2002). With the obtained data, the amount of potassium leached was calculated according to Equation 4, and the results were expressed in ppm.

$$LK = \frac{Value \ read \times dilution \times 1,56}{Mass \ (g)}$$

# 2.6.3 Grease acidity

For this analysis, coffee samples stored for 3 months were used (MARQUES et al., 2008) in a cold room with a temperature of 10 °C. The acidity of the grease was determined by titration, according to the method described by the American Association of Cereal Chemists -AACC (1995): 40g of ground coffee samples were weighed and 100 mL of toluene were added to stir for 1 hour and 30 minutes. It was then filtered, using filter paper; 25 mL of the filtered solution were mixed in a conical flask with 25 mL of ethanol (95% v.v-1) over phenolphthalein (0.04% w.v-1) and the solution was titrated with (KOH) at a concentration of 0.025 mol L<sup>-1</sup> until it reached the turning point. The result of the acidity content of the grease was expressed in mL of KOH.100 g<sup>-1</sup> DM, calculated according to Equations 5 and 6.

$$PS = [1 - U(b.u.) \times PC(g)]$$

$$AG = \frac{[V(ml) \times 100]}{PS(g)}$$

Where:

PS: mass of the dry sample (g);

PC: coffee weight (g);

U (w.b.): moisture content on a wet basis (%);

V: KOH volume spent in the titration (extract + indicator), in mL;

AG: grease acidity (mL KOH.100 g<sup>-1</sup> DM).

# 2.7 Statistical analysis

The experimental design consisted of a factorial scheme 2x7, completely randomized, with two processing forms (natural and pulped) and seven drying treatments. Three replicates were performed for each treatment.

The data obtained from the physiological, physicochemical and sensory analysis of coffee were also submitted to analysis of variance, using the Assistat 4.0 software and the means were grouped by the Scott-Knott test, at 1% significance.

# **3 RESULTS AND DISCUSSION**

# 3.1 Characterization of drying conditions

Table 2 shows the average moisture content values at the beginning and at the end of mechanical drying and the total drying time, for dry and wet processed coffee. It can be observed from Table 2 that the drying treatment with higher temperatures at the end of the process had a lower total drying time, when compared to the other mechanical drying treatments. This fact results from the greater difficulty in water removal when the fruits are at lower moisture contents.

**TABLE 2** - Average moisture content values and total drying time, for each drying and processing treatment - Lavras - 2013.

		Average mois	_ Arrama aa derri	natima (h)	
		(% w	.b.)	<ul> <li>Average drying time (h)</li> </ul>	
Drying Treatment	Processing			Before	
		Initial	End	Half dry (w.b.)	Total
35/40 °C	Pulped	46.5	11.6	20.0	32.0
35/40 °C	Natural	65.0	11.4	48.0	113.0
35/45 °C	Pulped	46.5	11.7	20.0	29.0
35/45 °C	Natural	65.0	11.1	48.0	97.0
35/50 °C	Pulped	46.5	11.3	20.0	28.0
35/50 °C	Natural	65.0	11.1	48.0	91.0
40/35 °C	Pulped	48.0	11.5	18.5	37.0
40/35 °C	Natural	64.4	11.2	40.0	139.0
45/35 °C	Pulped	48.0	10.7	15.5	35.5
45/35 °C	Natural	64.4	11.0	28.0	117.0
50/35 °C	Pulped	48.0	10.8	12.0	34.5
50/35 °C	Natural	64.4	11.0	20.0	107.0
On Ground	Pulped	46.5	10.9	-	145.0
On Ground	Natural	64.4	11.8	-	251.0

It can also be observed that the higher total drying times of coffee beans occurred in the treatment on ground, due to the lower exposure time of these coffees to high temperatures and greater relative humidity of the ambient air, to which these coffees were submitted.

According to Borém et al. (2006) and Ribeiro et al. (2003), the exposure time, the drying air temperature and flow, the initial and final moisture content of the product, the ambient air temperature, are factors that affect the drying dynamics, and have a significant effect on the quality of agricultural products. The removal of exocarp and mesocarp in the wet processing of coffee contributes to the drying time of these coffees.

The increase in air temperature results in a greater difference between the vapor pressure of the drying air and the product, making the water easier and faster to remove (SIQUEIRA; RESENDE; CHAVES, 2012). The increase in temperature reduces the viscosity of the water, directly influencing the resistance of the fluid to the flow. The decrease in viscosity facilitates the diffusion of the water molecules in the capillaries

of the product, besides providing an increase in the vibration level of the water molecules, which also contributes to the increase in the drying rate (CORRÊA et al., 2010).

# 3.2 Sensory analysis

Considering this type of evaluation, the analysis of variance of the data was performed for the overall score.

The overall score for each type of processing is shown in Table 3 and the overall score for each drying method is in Table 4.

It is observed that processing had no significant effect on the overall score. However, if each attribute that comprises the overall score is analyzed, it was observed by panelists that natural coffees have higher scores, in terms of body and sweetness. Pulped coffees presented higher scores for acidity and finishing. This fact is directly related to the type of processing used. As the overall score is the sum of all attributes, there were no significant differences for these coffees, even though it was known that, for our taste, the coffee was different.

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**TABLE 3** - Mean values of the overall score for each drying treatment - Lavras - 2013.

Processing	Overall score
Natural	80.50 A
Pulped	79.60 A

Means followed by distinct letters, upper case in columns, belong to the same cluster by Scott-Knott's test at 1% probability.

**TABLE 4** - Mean values of the overall score for each drying treatment - Layras - 2013.

Drying treatment	Overall score	
On Ground	82.70 A	
35/40 °C	81.20 A	
35/45 °C	78.83 B	
35/50 °C	77.55 C	
40/35 °C	81.54 A	
45/35 °C	79.42 B	
50/35 °C	80.04 B	

Means followed by distinct letters, upper case in columns, belong to the same cluster by Scott-Knott's test at 1% probability.

Regarding drying treatments 50/35 °C and 35/50 °C, there were differences between the average values of the overall score, regardless of the type of processing used, whether it is pulped or natural. The highest overall score values were found in the drying treatment 50/35 °C. It is noted that the use of heated air at 50 °C after half-dry was extremely detrimental in maintaining its sensory characteristics, indicating a higher sensitivity of these coffees to the increase in the drying temperature at the end of the drying process. According to Borém et al. (2008) and Saath et al. (2012), the cell membranes of coffee beans are especially damaged when the moisture contents of coffee are between 30% to 20% (w.b.), using a constant drying temperature of 60 °C for pulped and natural coffee.

The highest values for the total score were found in the coffees dried on ground, 35/40 °C and 40/35 °C, when compared to the other drying treatments with heated air. This fact indicates a possibility of new managements that reduce costs, since there will be a greater final quality of the product. For specialty coffees, some producers are using lower temperatures.

It can also be observed that the increase in drying temperature, before or after half-dry, was detrimental to the sensory attributes of these coffees. Borém et al. (2006), Coradi et al. (2007) and Marques et al. (2008), studying the effect

of bean mass temperature on sensory quality, reported that the increase in drying temperature was detrimental to the maintenance of sensory quality of parchment and natural coffee.

# 3.3 Physiological and physicochemical analyses

# 3.3.1 Electrical conductivity, potassium leaching and germination

Table 5 reveals that the type of coffee processing and drying had a significant influence the physiological evaluations. Lower germination values were found in natural coffees, indicating that more intense physiological damage occurred in these coffee beans, due to the longer exposure period to high temperatures. A similar result was observed by Taveira (2009), indicating the higher tolerance of these coffees to high drying temperatures, when compared to natural coffees. For natural coffees, only the coffees dried in yard presented values indicative of the presence of physiological activity in the beans. For the drying treatments with heated air, the values were low, indicating embryo death of natural coffee beans during the drying process, reinforcing the sensitivity of these coffees to drying at high temperatures (TAVEIRA, 2009).

ralues of physiological and physing, data in percentage (%) - Lavr	icochemical evaluations for the ir ras - 2013.	nteraction drying treatment
 Electrical conductivity	Potassium leaching	Germination

	Electrical	conductivity	Potassium	leaching	Germination	
Drying Treatment	Natural (μS.cm <sup>-1</sup> .g <sup>-1</sup> )	Pulped (μS.cm <sup>-1</sup> .g <sup>-1</sup> )	Natural (mg.kg <sup>-1</sup> )	Pulped (mg. kg <sup>-1</sup> )	Natural (%)	Pulped (%)
On Ground	2.06 aC	1.38 aC	15.85 aB	8.68 bA	70.25 aA	72.75 aB
35/40 °C	4.82 aA	3.75 bA	20.26 aA	12.02 bA	50.50 bB	93.34 aA
35/45 °C	5.10 aA	3.91 bA	14.00 aB	14.80 aA	8.17 bD	92.67 aA
35/50 °C	5.21 aA	4.12 bA	15.18 aB	12.02 aA	$0.00~\mathrm{bD}$	76.33 aB
40/35 °C	3.47 aB	2.32 bB	12.13 aB	10.99 aA	35.33 bC	94.5 aA
45/35 °C	3.66 aB	2.71 bB	8.81 aC	13.64 aA	37.33 bC	92.66 aA
50/35 °C	3.85 bB	3.10 bB	9.13 aC	9.87 aA	31.00 bC	81.50 aB

Means followed by same lowercase letters in the rows and capitals in the columns belong to the same cluster by Scott-Knott's test at 1% probability.

The drying treatments 35/40 °C, 40/35 °C, 35/45 °C and 45/35 °C for pulped coffees, are presented as a good alternative for seed drying of wet processed coffee, in order to maintain their physiological quality (germination and EC). According to Taveira et al., (2015) wet processed coffee has a greater tolerance to drying than those which underwent the dry process, due to the greater activity of antioxidant enzymes, found in wet processed coffee.

For natural coffees, the increase in drying temperature before reaching the moisture content of 30% or later, significantly reduced the germination percentage, being more intense in the drying treatments 35/45 °C and 35/50 °C.

The use of high temperatures allows faster drying; however, it can lead to a very large moisture content difference between the periphery and the bean center, generating a high pressure gradient, which can cause disruption in the cellular membranes of coffee beans, resulting in a reduction in seed vigor, related to the emergence and potential development of normal seedlings, or even the total loss of viability, defined as the ability to produce normal seedlings.

It can be confirmed from Table 5, that there were significant differences between the types of processing and drying used in the experiment, in relation to the electrical conductivity. The highest values of electrical conductivity, regardless of the drying treatment, were found in natural coffees, when compared to pulped coffees, indicating that this form of processing contributed to the electrical conductivity values being smaller, with consequent maintenance of the cellular structures

and product quality. Another fact that may have contributed would be the shorter exposure time of these coffees to the high temperatures, when compared to the natural coke exposure times (PRETE, 1992).

Regarding drying treatments, it was noticed that the increase in drying temperature resulted in higher electrical conductivity values, both for coffees processed dry and wet. This fact corroborates what was reported by Borém et al. (2008) and Coradi et al. (2007), who verified that the increase in drying temperature causes damage to the cell membrane system of coffee beans, increasing the electrical conductivity of the bean exudate.

It is also seen in Table 5 that the drying treatment that caused the least damage to cell structures was on ground. This fact may be related to the lower exposure time to high temperatures, lower bean mass temperatures and lower drying rates. Regarding the mechanical drying treatments, drying temperatures of 35/40 °C, 35/45 °C and 35/50 °C resulted in the highest values of EC, LK and Germination, indicating a higher compromise of coffee quality, compared to drying at temperatures 50/35 °C, 45/35 °C and 40/35 °C. This increase in the electrical conductivity of coffees processed through drought and drought, when using higher temperatures after half-drought, compared to using the same temperatures before half-drought, can be explained by the longer exposure time and greater disruption of cell membranes when using high temperatures at the time the product was at a lower moisture content. Similar phenomena were observed by Saath et al. (2012), who analyzed the

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damage caused by the drying temperature in the cellular structures of coffee beans, found that they occur more intensively between moisture contents ranging from 30% (w.b.) to 20% (w.b.), when using the temperature of 60 °C in drying.

As in the electrical conductivity test, the highest values of potassium leaching were found in coffees processed dry, again indicating that the exposure time of these coffees to drying, both in terrarium and in dryers, may have been one of the causers of this phenomenon. The same phenomenon was observed by Taveira et al. (2012), studying temperature changes during the drying process.

The high temperature at the beginning of drying, before the half-dry, at the treatment of 50 / 35 °C, and at the end of drying, after the half-dry in the treatment 35/50 °C, could have been harmful to the physiological integrity of the beans, indicated by high potassium leaching values, compared to drying on ground. The highest potassium leaching values were found in natural and pulped coffees dried at temperatures of 35/45 °C and 35/50 °C, indicating the higher sensitivity of the membranes to low moisture contents. At this moment, there was a very large energy accumulation inside the bean, which can, depending on the temperature used in drying, compromise the cellular structures with consequent solute leaching.

There is agreement that degeneration of cell membranes and subsequent loss of permeability control is one of the early events that characterize deterioration. According to Malta et al. (2005), any factor that changes the structure of the membrane,

such as the attack of insects and microorganisms, physiological changes, mechanical and thermal damage, cause a rapid deterioration of coffee beans. These changes cause chemical reactions that modify the original chemical composition of coffee beans and, consequently, their sensory and physiological properties.

# 3.4 Grease acidity

Table 6 presents the results of the effect of the drying treatment for each type of bean processing on the acidity of the grease.

Significant differences were observed in the acidity content of the grease between drying and processing treatments. These results are related to membrane stabilization and cell wall integrity, indicating that a greater cell membrane degradation will give rise to a greater amount of free fatty acids (MARQUES, 2008). According to Coradi and Lemes (2018), the reduction in lipid content and the increase in free fatty acid contents during storage are directly correlated with the speed and intensity of the bean deterioration process, and it is possible to use the free fatty acid content as an indicator of bean deterioration.

In relation to the processing of coffee beans, Table 6 reveals that the highest values were found in coffees processed dry. It is assumed that the higher exposure of these coffees to high temperatures caused the rupture of cell membrane structures, extravasating oils and compromising coffee quality with oxidation processes, demonstrating the higher sensitivity of these coffees at high temperatures (OLIVEIRA et al., 2012).

**TABLE 6-** Average grease acidity values for the interaction between drying and processing treatments Lavras - 2013.

	Grease Acid	dity
Drying treatment	Natural	Pulped
	(mL KOH.100 g <sup>-1</sup> DM)	(mL KOH.100 g <sup>-1</sup> DM)
On Ground	3.39 aD	3.13 bB
35/40 °C	3.49 aC	3.04 bB
35/45 °C	3.71 aB	3.23 bB
35/50 °C	3.91 aA	3.40 bB
40/35 °C	3.32 aD	3.16 bB
45/35 °C	3.53 aC	3.35 bB
50/35 °C	3.66 aB	3.48bB

Means followed by same lowercase letters in the rows and capitals in the columns belong to the same cluster by Scott-Knott's test at 1% probability.

It can be observed that the results for pulped coffee with mechanical drying were satisfactory, since the value of its grease acidity was statistically equal to that of drying on ground, a phenomenon that indicates a greater maintenance potential of the quality of these coffees when stored, suggesting that husk removal in pulped coffees decreased the amount of free fatty acids. The drying treatments 35/50 °C and 50/35 °C were those that yielded the highest grease acidity values, suggesting that this drying treatment damaged the cellular structures of the coffee beans, giving rise to a greater number of free fatty acids.

# **4 CONCLUSIONS**

Drying on ground for the climatic conditions during the experiment provided the best physiological sensory quality of coffee beans, when compared with drying with heated air.

Pulped coffee has better physiological quality.

The temperatures of 35/50°C and 50/35 °C were the ones that yielded the worst germination results.

The use of higher temperatures after half-dry was more damaging than when used before half-dry in terms of electrical conductivity.

The use of lower temperatures (35/40 °C and 40/35 °C) showed similar results to drying on ground.

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# ESTIMATION OF REFERENCE EVAPOTRANSPIRATION FOR COFFEE IRRIGATION MANAGEMENT IN A PRODUCUTIVE REGION OF MINAS GERAIS CERRADO

André Luís Teixeira Fernandes<sup>1</sup>, Rafaella Esthefania Cardoso Gomes Mengual<sup>2</sup>, Giovani Luiz de Melo<sup>3</sup>, Leonardo Campos de Assis<sup>4</sup>

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ABSTRACT: Evapotranspiration (evaporation and transpiration) represents vegetated soil water loss to the atmosphere and can be estimated by various empirical methods. The aim of this study was to evaluate the performance of methods of Blaney-Criddle, Jensen-Haise, Linacre, Solar Radiation, Hargreaves-Samani, Makkink, Thornthwaite, Camargo, Priestley-Taylor and Penman in the estimation of potential evapotranspiration comparing to the standard method Penman-Monteith (FAO56) regarding the climatic conditions of the city of Araxá, MG. A set of 35 years of monthly data (1976 to 2010) was used, working with the climatic elements: temperature, relative humidity, wind speed and insolation. The empirical methods to estimate reference evapotranspiration were compared with the standard method using linear regression, simple statistical analysis, Willmott agreement index (d) and performance index (c). The method of Makkink showed the best performance according to the set of parameters evaluated and it is recommended to calculate ETo in Cerrado of Minas Gerais, for coffee irrigation management.

Index terms: Climatic elements, empirical equations, FAO Penman-Monteith, irrigation, Cerrado.

# ESTIMATIVA DA EVAPOTRANSPIRAÇÃO DE REFERÊNCIA PARA O MANEJO DA IRRIGAÇÃO DO CAFEEIRO EM UMA REGIÃO PRODUTIVA DO CERRADO DE MINEIRO

RESUMO: A evapotranspiração representa a perda de água do solo com vegetação para a atmosfera, podendo ser medida por métodos diretos ou estimada empiricamente. O objetivo deste estudo foi avaliar o desempenho dos seguintes métodos empíricos: Blaney-Criddle, Jensen-Haise, Linacre, Radiação Solar, Hargreaves-Samani, Makkink, Thornthwaite, Camargo, Priestley-Taylor e Penman na estimativa da evapotranspiração potencial, comparando-os com o método padrão Penman-Monteith (FAO56) quanto às condições climáticas da cidade de Araxá, MG. Utilizaram-se 35 anos de dados mensais (1976 a 2010), das seguintes variáveis climáticas: temperatura, umidade relativa do ar, velocidade do vento e insolação. Os métodos empíricos utilizados para estimar a evapotranspiração de referência foram comparados com o método padrão por meio de regressão linear, análise estatística simples, índice de concordância de Willmott (d) e índice de desempenho (c). Dentre os métodos alternativos, o de Makkink apresentou melhor desempenho, portanto, pode ser utilizado para estimar a ETo para fins de manejo da irrigação do cafeeiro cultivado no cerrado mineiro.

Termos para indexação: Dados meteorológicos, métodos empíricos, FAO Penman-Monteith, Irrigação, Cerrado.

# 1 INTRODUÇÃO

The hydrologic cycle is a planetary phenomenon caused by water movement, mainly affected by gravity action and solar energy. Rainfall, infiltration, surface runoff, evaporation and transpiration are the main processes which dynamically interact with each other and are influenced by human actions caused by industry and agricultural activities development (BORGES; MENDIONDO, 2007). The actual evapotranspiration is a key process to hydrological cycle and the element that associates land surface water and energy balance (ZHAO et al., 2013).

The water balance relates data on deficiency, surplus, withdrawal and water

replacement throughout the year. The analysis of these components is crucial to agricultural activities planning and implantation (CASTRO et al., 2010). Certainly, one of the great interests in understanding the water balance of an agricultural area is to infer about water volume availability, mainly due to the possibility of increasing the crop production activity through irrigation practices and consequently, the regional economic income. The most important variables to water balance calculation are evapotranspiration and rainfall (BORGES; MENDIONDO, 2007). Crop evapotranspiration is necessary to design and management irrigation systems (ESTEVES et al., 2010; SOUZA et al., 2010).

 $<sup>^{1,4}</sup>$ Universidade de Uberaba/UNIUBE - Campus Aeroporto - 38.055-500 - Uberaba - MG - andre.fernandes@uniube.br, leonardo.assis@uniube.br

<sup>&</sup>lt;sup>2</sup>Avenida Imbiara, 1455 – Centro - 38.183.244 - Araxá - MG - rsthefany@hotmail.com

<sup>&</sup>lt;sup>3</sup>Vale Fertilizantes - Estrada da Cana, Km 11 - 38.001-970 - Uberaba-MG -giovanicaseca@gmail.com

Evapotranspiration is the combination of two distinct process, the loss of water of land surface from evaporation and the vegetables transpiration through the plant stomata (ALLEN et al., 1998). Reference evapotranspiration (ETo) is defined as the evapotranspiration of a hypothetic crop with a fixed high of 0.12 m, albedo equal to 0.23 and surface resistance to water transport equal to 70 s.m<sup>-1</sup>.

According to Alves Sobrinho et al. (2011), evapotranspiration can be directly measured by specific equipment, as the weighing lysimeter, however the high cost restricts its use to research institutes and for regional calibration of indirect methods (MELO; FERNANDES, 2012), hence several authors develop and test empirical methods to estimate evapotranspiration indirectly. Serrat-Capdevila et al. (2011) presented an approach to quantify reference evapotranspiration under climate changes, employing field observations, theoretical models and meteorological predictions of global climate models using a watershed from southern Arizona - USA. Sun et al. (2009) developed a simple evapotranspiration model based only on remote sensing data. The authors adjusted a model that showed a good performance for instantaneous estimations.

The climatic features of the Brazilian Cerrado region have induced agricultural producers (i.e., coffee, soybean, corn and others) to use irrigation systems in order to reduce the loss risk associated to water deficit in dryer periods (FERNANDES et al., 2012). The region is a hot spot zone to agriculture production of crops as coffee, grains and fruits, covering about 200 million hectares on the states of Minas Gerais, Mato Grosso, Mato Grosso do Sul, Tocantins, Bahia, Piauí, Maranhão e Distrito Federal.

Cerrado region requires irrigation to supply water during important crop phenological phases on the dry months of the year (FERNANDES et al., 2012). However, there are conflicts for water availability in some locations, which makes difficult the implementation of large-scale irrigation techniques. Efficiency on water use allows greater economic viability and environmental sustainability of irrigated agriculture (LEVIDOW et al., 2014). Hence, one way of increasing the efficiency on water use is through accurate evapotranspiration estimates.

Accurate estimation of reference evapotranspiration is required to rational water resource use (SHIRI, 2017). The Food and Agriculture Organization (FAO) recommends using the Penman-Monteith method (FAO56-

PM) as standard to evapotranspiration estimation (ALLEN et al., 1998). This method, beside is used to compute evapotranspiration as well as calibration of other models (BEZERRA et al., 2010a; TABARI et al., 2013). However, this method depends on several meteorological data, frequently unavailable, what makes difficult the large-scale usage, especially to smaller farmers. This implies the need for simpler approaches to estimate evapotranspiration, through developing methods that require fewer parameters but able to provide reliable on ETo (PEREIRA et al., 2009; SHIRI, 2017).

The assessment of simpler approaches applied to ETo estimation has been receiving considerable attention in developing countries, where the data required by the FAO56-PM method are often incomplete and/or unavailable (TABARI; GRISMER; TRAJKOVIC, 2013). Therefore, empirical methods to ETo estimation, founded on physic properties, empirical equations or the combination of both, have become alternative approaches in the last decades (ALENCAR et al., 2011a). Additionally, considering that: i) ETo is necessary to design and manage irrigation systems (ESTEVES et al., 2010; SOUZA et al., 2010); ii) the role played by irrigation to improve agricultural production in semi-arid environments; iii) the representability of Brazilian Cerrado of Minas Gerais state agricultural production to the national total production; iv) the need for irrigation on scarcity periods of the year (i.e., from April to September); We aimed to assess the performance of different methodologies to estimate the ETo in relation to the FAO56-PM method (ALLEN et al., 1998), to the Cerrado region from Minas Gerais state.

# 2 MATERIALS AND METHODS

Meteorological data used to ETo estimation were obtained from a conventional land surface meteorological station, OMN code 83579, located at the Cerrado biome, specifically in Araxá municipal district, Minas Gerais state at coordinates 19° 36' 36" S, 46° 57' 00" W and 1023.61m above the sea level (Figure 1).

According with the Köopen climatic classification (ALVARES et al., 2013), the climate of region is of the type Cwa, associated to humid temperate climate of rainy summer and dry winter (SÁ JÚNIOR, 2009) with annual mean rainfall about 1500 mm. The dry period varies from four to seven months and the rainfalls occur from October to March (Figure 2).

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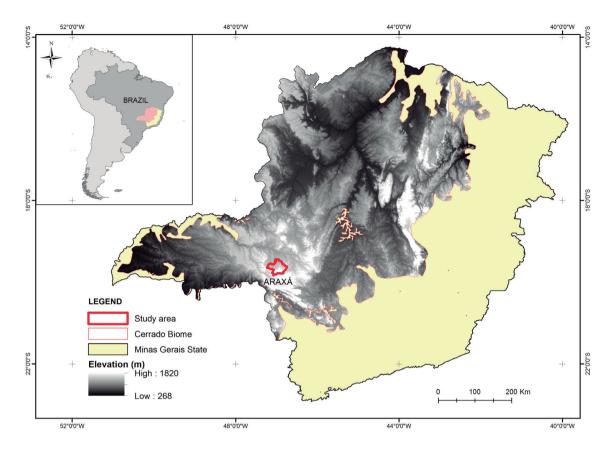
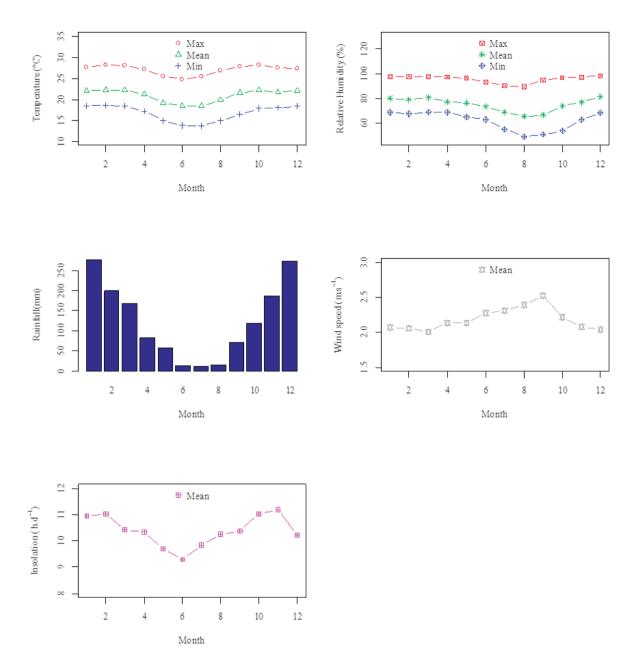


FIGURE 1 – Location of study area, which highlight to Araxá municipality (red boundary) at the Cerrado biome.

Figure 2 also shows mean monthly variation of the parameters used by the methods to compute evapotranspiration: maximum, minimum and mean temperature and relative humidity, wind speed measured at 2 m height and insolation, of 35 years of records, from January 1976 to December 2010. Note that maximum temperature is in February; the driest month is August; in September there is the highest wind speed and in November the greatest solar radiation. The Eto methods were categorized according to their premises, as: i) energy-based; ii) temperature-based, and iii) mass transfer-based (ZHAO et al., 2013).

Eventual missing data were discarded to avoid compromising the comparison procedure. Hence, all methods were analyzed with the same dataset. The mean monthly values of ETo

estimation were calculated according to the following empirical methods: Blaney-Criddle (BLANEY; CRIDDLE, 1950), Jensen-Haise (JENSEN; HAISE, 1963), Linacre (LINACRE, 1977), Solar Radiation (DOORENBOS; PRUITT, Hargreaves-Samani (HARGREAVES; SAMANI, 1985), Makkink (MAKKINK, 1957), Thornthwaite (THORNTHWAITE, Camargo (CAMARGO, 1971), Priestley-Taylor (PRIESTLEY; TAYLOR, 1972) and Original Penman (PENMAN, 1948). Then, the results were evaluated by level of agreement to Penman-Monteith standard method. The empirical methods were selected because of their applicability features to the study area. The next sections present the methods by type (i.e., based on temperature, energy or mass-transfer).



**FIGURE 2** - Mean monthly values of climatic parameters from 1976 to 2010; Maximum, minimum and mean temperature; Maximum, minimum and mean relative humidity; Rainfall; Wind speed; and insolation.

# Temperature-based methods

The temperature-based methods are some of the earliest methods (TABARI; GRISMER; TRAJKOVIC, 2013); they are recommended under limited climate data conditions (ZHAO et al., 2013).

Blaney and Criddle (1950): initially, the method was developed for semi-arid region from New Mexico and Texas in the West of United States. Doorenbos and Pruitt (1977) proposed a correction factor to the original equation, using humid variables as wind speed and insolation, enabling the application to several climatic conditions. To easy calculus and avoid interpolations, (FREVERT; HILL; BRAATEN, 1984) proposed the following change on Blaney-Criddle FAO-24 method (Equation 1):

ETo 
$$_{BC} = a + b p (0.46T_{m} + 8.13)$$
 (1)

Where a and b are the coefficient values, obtained from:

$$a = 0.0043 RH_{min} - \frac{n}{N} - 1.41$$

$$b = a_{_0} + a_{_1}RH_{_{min}} + a_{_2}\frac{n}{N} + a_{_3}U_{_2} + a_{_4}RH_{_{min}}\frac{n}{N} + a_{_5}RH_{_{min}}U_{_2}$$

Were RH<sub>min</sub> is the monthly relative humidity minimum (%); p is the percent of the total mean monthly photoperiod (°C) relative to the total annual photoperiod, according to Table 1; n is the total insolation (h); N is the photoperiod (h); U<sub>2</sub> is the wind speed at 2m above ground (m s<sup>-1</sup>). The coefficients are:  $a_0 = 0.81917$ ,  $a_1 = -0.0040922$ ,  $a_2 = 1.0705$ ,  $a_3 = 0.065649$ ,  $a_4 = -0.0059684$  e  $a_5 = -0.0005967$ .

Thornthwaite (1948): the method was based on the water balance of watersheds and lysismeter measures, using only air temperature as independent variable. The mean monthly potential evapotranspiration for grass surface is obtained through the Equation 2:

$$ETP = 16 \left( 10 \frac{T_m}{I} \right)^a \tag{2}$$

$$T_m > 0^{\circ}C$$

$$a = 6.75 \cdot 10^{-7} I^3 - 7.71 \cdot 10^{-5} I^2 + 0.01791 \cdot I + 0.49239$$

$$I = \sum_{i=1}^{12} (0.2T_{\rm m})^{1.514}$$

Where I is the regional heat index; Tm is the mean temperature (°C).

The Thornthwaite method estimates evapotranspiration to a standard condition of 12 hours of sun shining and a month of 30 days and can be calculated through the Equation 3:

ETo <sub>Th</sub> = ETP 
$$\frac{N}{12} \frac{ND}{30}$$
 (3)

Where ND is the number of days in a month. Hargreaves-Samani (1985): the method was developed using lysimeter data in Davis-CA under grass surface and semi-arid climatic condition. The Equation 4 is as follows:

ETO 
$$_{HS} = 0.0023$$
 Qo (Tmax – Tmin )<sup>0.5</sup> (T + 17.8 ) (4)

Where Qo is the total daily solar radiation incident in horizontal surface (mm d<sup>-1</sup>)

Linacre (1977): the method was originally proposed to the Australian climatic conditions, based on a Penman-Monteith simplification. It uses only temperature and relativity humid data, associated to the location in terms of latitude and elevation. Evapotranspiration values are obtained from the Equation 5:

$$ETo_{Lin} = \frac{J(T_{m} + 0.006h)}{100 - |\Phi|} + 15(T_{m} - T_{0})$$

$$80 - T_{m}$$
(5)

$$(T_m - T_0) = 0.023h + 0.37T_m + 0.53 (T_{max} - T_{min}) + 0.35R - 10.9$$

Where J is a constant equal 500 in case of vegetation (albedo = 0.25);  $\Phi$  is the latitude in degrees; Tmax is the monthly maximum temperature (°C); Tmin is the monthly minimum temperature (°C); T0 is the monthly mean temperature of the dew point (°C); R is the difference among the mean temperatures of the hottest and coldest months (°C); h is the local height (m).

Camargo (1971): from the results of Thornthwaite, Camargo propose a simpler method, however, as efficient as the one mentioned. The method uses only mean air temperature and extraterrestrial solar radiation data and the evapotranspiration is given by the Equation 6:

$$ETo_{Cam} = F Qo T_m ND$$
 (6)

Where F is an adjustment factor dependent on annual mean temperature ( $T_a < 23.5$  °C, F = 0.01).

Month		South Latitude	
Month	15°	19,73°	20°
January	0.29	0.299	0.30
February	0.28	0.289	0.29
March	0.28	0.280	0.28
April	0.27	0.261	0.26
May	0.26	0.251	0.25
June	0.25	0.250	0.25
July	0.26	0.251	0.25
August	0.26	0.260	0.26
September	0.27	0.270	0.27
October	0.28	0.280	0.28
November	0.29	0.290	0.29
December	0.29	0.299	0.30

TABLE 1 - P factor as function of Latitude and month according to Doorenbos & Pruitt (1977).

# **Energy-based methods**

The energy-based method applies the energy balance concept to estimate potential evapotranspiration (ZHAO et al., 2013).

Jensen-Haise (1963): this method was defined to calculate evapotranspiration from temperature and solar radiation data, applied to arid and semi-arid regions, through the Equation 7:

$$ETo_{JH} = Sr(0.0252T_{m} + 0.078)$$
 (7)

Where Sr is the solar radiation at ground level (mm d<sup>-1</sup>).

Makkink (1957): based on Penman method, Makkink uses solar radiation at ground level to estimate evapotranspiration. Developed in the Netherlands, it is well known at European western region and can be calculated through the Equation 8:

ETo 
$$_{\text{Mak}} = 0.61$$
 W Sr  $- 0.12$  (8)
$$W = \frac{\Delta}{\Delta + \gamma}$$

Where W is a weighting factor representing proportion of Sr (solar radiation) used by evapotranspiration, adjusted to different height and temperature values;  $\Delta$  is the slope of the pressure curve of water vapor in the air atmosphere (kPa °C<sup>-1</sup>);  $\gamma$  is the psychometric constant (kPa °C<sup>-1</sup>).

Solar Radiation: The method is a Doorenbos & Pruitt (1977) adaptation of FAO-24 method

of solar radiation proposed by Makkink (1957), previously developed to humid conditions of the Netherlands, according to the Equation 9:

$$ETo_{RS} = c_0 + c_1 W Sr$$
 (9)

The value of c1 is determined as follows:

$$c_1 = a_0 + a_1 RH_0 + a_2 U_2 + a_3 RH_0 U_2 + a_4 RH_0^2 + a_5 U_2^2$$

Where  $C_0$  is a constant equal to -0,3 (mm d<sup>-1</sup>); and the coefficients:  $a_0 = 1.0656$ ,  $a_1 = -0.0012795$ ,  $a_2 = 0.044953$ ,  $a_3 = -0.00020033$ ,  $a_4 = -0.000031508$  and  $a_4 = -0.0011026$ 

-0.000031508 and  $a_5 = -0.0011026$ .

Priestley & Taylor (1972): the method approximates Penmans' method through simplification, maintaining only the solar radiation balance corrected by an empirical coefficient known as the Priestley and Taylor parameter, what incorporates the additional energy to the process of evapotranspiration as function of the aerodynamic term. The method follows the Equation 10:

$$ETo_{PT} = \frac{\alpha W(Rn - G)}{\lambda}$$
 (10)

Where  $\alpha$  is the Priestley and Taylor parameter equals to 1.26;  $\lambda$  is the evaporation latent heat equals to = 2.45 MJ Kg<sup>-1</sup>; Rn is the daily amount of liquid solar radiation – or radiation balance (MJ m<sup>-2</sup> d<sup>-1</sup>); G is the soil heat flux (MJ m<sup>-2</sup> d<sup>-1</sup>).

# Mass transfer-based method

Known as one of the oldest evapotranspiration methods, the mass transfer-based method estimates the free water surface potential evaporation mainly considering the effect of air pressure deficit and wind speed (ZHAO et al., 2013).

Penman original (1948): is one of the most used methods applied to evapotranspiration estimative; the equation is derived assuming proportionality among water evaporation and subirrigated grass evapotranspiration. The method is obtained by the Equation 11:

$$ETo_{PE} = \frac{WRn}{\lambda} + (1 - W) E_a$$
 (11)

Where  $E_a$  is the power of air evaporation (MJ  $m^{-2} d^{-1}$ ).

# **Reference Method (Penman-Monteith)**

The Penman-Monteith method assume evapotranspiration as an outcome of the energy and aerodynamic terms which are governed by the resistance transport of water vapor to the atmosphere (FERNANDES; FRAGA JÚNIOR; TAKAY, 2011). The Penman-Monteith method (FAO56-PM) is parametrized by Allen et al. (1998) in the Equation 12:

$$ETo_{PM} = \frac{0.408 \Delta (Rn - G) + \left[ \frac{\gamma 900 U_2 (e_s - e_a)}{T_m + 273} \right]}{\Delta + \gamma (1 + 0.34 U_2)}$$
(12)

Where  ${\rm ETo_{PM}}$  is the reference evapotranspiration (mm.d-¹);  $\Delta$  is the slope of the pression curve of water vapor in the air atmosphere (kPa °C-¹); Rn is the daily amount of liquid solar radiation – or radiation balance (MJ m-² d-¹); G is the soil heat flux (MJ m-² d-¹);  $\gamma$  is the psychometric constant (kPa °C-¹);  $U_2$  is the wind speed at 2m above ground (m s-¹);  $e_s$  is the water vapor saturation pressure (kPa);  $e_s$  is the current water vapor pression (kPa);  $e_s$  is the mean air temperature (°C);  $e_s$  is the grass surface reflection coefficient, assumed equal 0.25.

# **Evaluation of methodologies**

The ETo data obtained through the different methodologies were compared with the data obtained by the FAO56-PM method. The performance of the methods in relation to the

FAO56-PM method was verified by comparing the linear regression parameters, mean bias (MB), root of mean squared error (RMSE), mean absolute error (MAE), standard error (SE) and, coefficient of performance (c). Table 2 was used to interpret the confidence coefficient.

The mean bias (MB) of each method was calculated from the Equation 13:

$$MB = N^{-1} \sum_{i=1}^{N} (P_i - O_i)$$
 (13)

Where O<sub>i</sub> is the ETo estimated FAO56-PM method (mm d<sup>-1</sup>); P<sub>i</sub> is the ETo estimated by the empirical methods (mm d<sup>-1</sup>); N is the number of observation.

The discrepancy was calculated by the root of mean squared error (RMSE) and the mean absolute error (MAE) by the Equations 14 and 15:

$$RMSE = \sqrt{\left(N^{-1} \sum_{i=1}^{N} (P_i - O_i)^2\right)}$$
 (14)

$$MAE = N^{-1} \sum_{i=1}^{N} |P_i - O_i|$$
 (15)

The standard error (SE) was calculated by the Equation 16:

$$SE = \sqrt{\frac{\sum_{i=1}^{N} (O_i - P_i)^2}{N - 1}}$$
 (16)

The agreement was given by the determination coefficient (R<sup>2</sup>), the correlation coefficient (r) and the agreement index (d) as proposed by Willmott et al. (1985), by the Equation 17:

$$d = 1 - \left[ \frac{\sum_{i=1}^{N} (P_i - O_i)^2}{\sum_{i=1}^{N} (|P_i - O| - |O_i - O|)^2} \right]$$
(17)

The agreement index is an efficiency measure of the analyzed methods that takes into account the dispersion over the straight line 1:1. Reliability was determined by the confidence index (c) proposed by Camargo & Sentelhas (1997) as the product of r and c, i.e., c = r.d. The confidence results are classified according the c values as Table 2.

c values	Classification
> 0.85	Excelent
0.76 a 0.85	Very good
0.66 a 0.75	Good
0.61 a 0.65	Moderate
0.51 a 0.60	Inadequate
0.41 a 0.50	Bad
< 0.41	Very bad

**TABLE 2** - Confidence index classification (c) according to Camargo & Sentelhas (1997).

# **3 RESULTS AND DISCUSSION**

The evaluation criteria of the empirical methods analyzed are in Table 3. Note that considering the global mean of the entire period. Thornthwaite and Camargo methods underestimate evapotranspiration according to the Mean Bias (MB) criterion; whereas Hargreaves-Samani and Blaney-Criddle are close to the standard reference method. In general, the methods that overestimate evapotranspiration show greater discrepancy as well, as exhibited by the RMSE, MAE and SE criteria results relative to Jensen-Haise. Solar Radiation and Penman Original methods. Similar results were observed by Melo & Fernandes (2012) evaluating these methods to estimate evapotranspiration of a neighboring region (i.e., at Uberaba-MG) in the period from 1990 to 2010.

Alencar et al. (2011a) used Blaney-Criddle, Solar Radiation and Hargreaves-Samani methods to estimate evapotranspiration at Uberaba-MG region in the period from 1996 to 2005 and verified that, these methods overestimate the ETo based on VM criteria.

A graphical analysis was performed where the regression graphs of the empirical methods against the standard reference method (FAO56-PM) for the long-term period (35 years) are exhibited in Figure 3 (first column - left margin).

Hence, each blue line in the Figure 3 (left column graphs) shows the graphical analysis of each one of the specific empirical methods according to regression line, residuals and timeseries of monthly ETo estimates.

Visual analysis of the graphs in Figure 3 based in time-series of monthly evapotranspiration estimation, residuals and regression line shows a better performance of Jensen-Haise, Solar Radiation, Makkink and Camargo methods, highlighting the Solar Radiation method relative to the residuals. This is confirmed by the residual standard error (RSE) and determination coefficient (R²) of Table 4.

However, Jensen-Haise and Solar Radiation were not as good as Makkink and Hargreaves-Samani considering the agreement index (d) and performance index (c), which represents respectively bad and inadequate classification for the first two, and, very good and good for the last two ones. Solar Radiation showed the better performance according to the coefficient of determination but was categorized as inadequate by the Confidence index classification. Among Hargreaves-Samani and Makkink, the former was not categorized as very good by the confidence index classification as the later did. In the light of evidence, the best set of results highlights Makkink as the most adequate empirical method to estimate potential evapotranspiration to the Cerrado region of Minas Gerais state. Additionally, Makkink method uses information about air temperature and solar radiation only, which are easy obtainable parameters.

Ørum et al., 2010 point alternatives to save water by using new irrigation systems as well as technology of water saving devices. Public policies must support these alternatives as proposals to reduce the water demand and expand irrigated area over the Cerrado region based on the perspective of water rational use. The crops water requirement on the drier months at Cerrado are not supplied by rainfalls, they need to be fulfilled by efficient irrigation management practices. Therefore, effective evapotranspiration estimation methods play an important role. Makkink and Hargreaves-Samani methods have exhibited good performance on evapotranspiration estimation to the study area and have a low cost in terms of parameters required, such as temperature and solar radiation, which make them accessible to the Cerrado farmers in Minas Gerais state.

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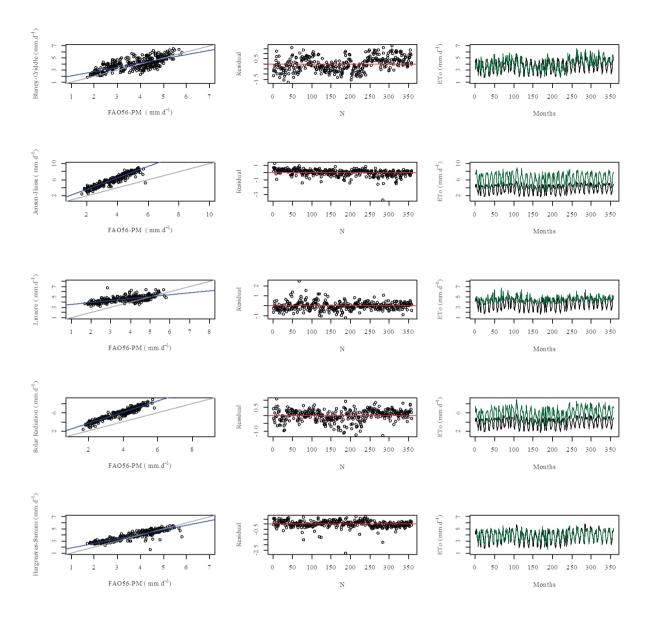
**TABLE 3** - Evaluation criteria of the empirical methods.

Method	Evaluation criteria					
Method	MB (mm d <sup>-1</sup> )	RMSE (mm d <sup>-1</sup> )	MAE (mm d <sup>-1</sup> )	SE (mm) d <sup>-1</sup> )		
Blaney-Criddle	0.22	0.79	0.65	0.79		
Jensen-Haise	2.36	2.44	2.36	2.44		
Linacre	0.78	1.08	0.86	1.08		
Solar radiation	2.06	2.10	2.06	2.11		
Hargreaves-Samani	0.17	0.46	0.34	0.46		
Makkink	0.50	0.61	0.55	0.61		
Thornthwaite	-1.09	1.22	1.10	1.22		
Camargo	-0.85	0.94	0.85	0.94		
Priestley-Taylor	0.42	0.96	0.81	0.96		
Penman Original	2.11	2.28	2.11	2.28		

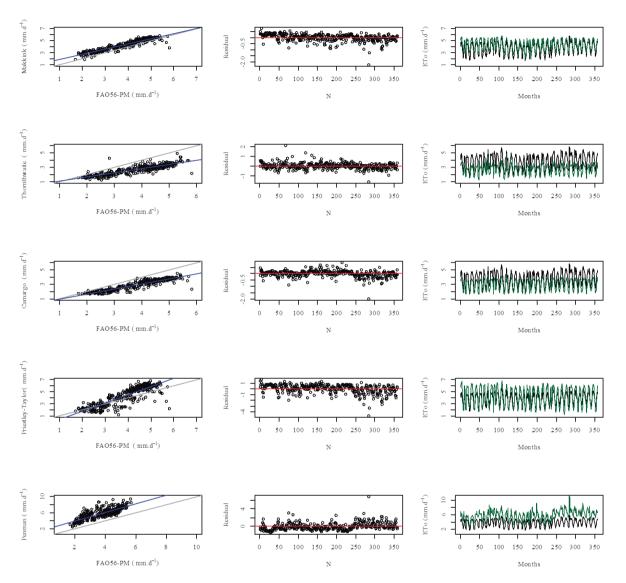
**TABLE 4** - Evaluation of the empirical methods performance.

Methods	Intercept p-value	Coefficient p-value	RSE	$\mathbb{R}^2$	d	c	Classification
Blaney-Criddle	1.414 2e-16'	0.687 2e-16'	0.69	0.49	0.83	0.58	Inadequate
Jensen-Haise	0.776 9.57e-15'	1.413 2e-16'	0.45	0.9	0.53	0.5	Bad
Linacre	3.16 2e-16'	0.375 2e-16'	0.42	0.44	0.65	0.44	Bad
Solar radiation	1.215 2e-16'	1.218 2e-16'	0.36	0.92	0.57	0.55	Inadequate
Hargreaves-Samani	1.144 2e-16'	0.744 2e-16'	0.34	0.82	0.94	0.84	Very good
Makkink	1.048 2e-16'	0.854 2e-16'	0.31	0.88	0.9	0.85	Very good
Thornthwaite	0.508 9.29e-13'	1.577 2e-16'	0.32	0.75	0.69	0.6	Inadequate
Camargo	0.3 1.94e-7'	0.697 2e-16'	0.27	0.86	0.78	0.73	Good
Priestley-Taylor	-0.894 1.31e-7'	1.347 2e-16'	0.79	0.74	0.86	0.74	Good
Penman	1.861 2e-16'	1.066 2e-16'	0.87	0.59	0.53	0.41	Very bad

Significance level: #1; \*0.1; \*\*\*0.01; \*\*\*0.001;  $^{\circ}$ . < 0.0001; RSE residual standard error at 356 degrees of freedom; Adjusted R<sup>2</sup>; d – index of agreement and c – index of performance.



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**FIGURE 3** - Monthly evapotranspiration values (FAO56 Penman-Monteith versus empirical methods) compared by linear regression. The grey line shows the 1:1 relation. Residuals are in the middle column and time-series of evapotranspiration estimation values in the right column.

# **4 CONCLUSION**

Among the alternative methods, the Makkink method presented better performance, therefore can be used to estimate the reference evapotranspiration for irrigation management purposes of coffee crop in the Cerrado region from Minas Gerais. As a second option, the Hargreaves-Samani and Camargo temperature-based methods also can be used to reference evapotranspiration estimation.

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# FUZZY LOGIC APPLICATION AND CLUSTER ANALYSIS IN THE QUALITY OF THE BEVERAGE FROM CONILON COFFEE

Julião Soares de Souza Lima<sup>1</sup>, Samuel de Assis Silva<sup>2</sup>

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ABSTRACT: The quality of coffee beverages has been under study due to the demand of the consumer market for both arabica and conilon coffee. The aim of this work was to study beverage quality from different clones by means of sensory analysis, in 13 clones of the variety Victoria INCAPER 8142 produced at average altitudes of 100.0 m and 528.0 m and with the cherry fruits processed by natural drying or depulping. Fuzzy classification was adopted for the global scores obtained in the sensory analysis, on a scale of 70.0 to 100.0 points, with the Euclidean distance from the cluster analysis being used to define the dissimilarity between the global fuzzified scores for the different clones at the two altitudes and for the two methods of processing the fruit. Clones C4 and C10, at the intermediate maturation stage, presented a mean global score (GS) of 85.0 points for the coffee produced at the altitude of 528.0 m and for the depulped fruit, corresponding to a degree of fuzzy pertinence (FI) of 0.50, and being classified as fine coffee. These same clones presented dissimilarities in the beverage produced by the depulped fruit, with better quality for the coffee at the higher altitude. The fuzzy classification taken together with the cluster analysis to interpret the mean global scores (GS) in the sensory analysis of the beverage for the different treatments under study identified variation in beverage quality.

Index terms: Sensory analysis, Coffea canephora, different altitudes.

# APLICAÇÃO DA LOGICA FUZZY E ANALISE DE AGRUPAMENTO NA QUALIDADE DA BEBIDA DO CAFÉ CONILON

RESUMO: A qualidade da bebida do café vem sendo estudada em decorrência da exigência do mercado consumidor, tanto para o café arábica como para o café conilon. Neste trabalho objetivou-se estudar, para os 13 clones da variedade Vitória INCAPER 8142 produzidos nas altitudes médias de 100,0 m e 528,0 m e com processamento dos frutos cereja secos em coco e desmucilados, a qualidade da bebida dos diferentes clones por meio da análise sensorial. Adotou-se a classificação fuzzy para as notas globais obtidas na análise sensorial, na escala de 70,0 a 100,0 pontos, e utilizou-se a distância euclidiana oriunda da análise de agrupamento para definir a dissimilaridade entre as notas globais fuzzificadas para os diferentes clones, nas duas altitudes e nos dois processamentos dos frutos. Os clones C4 e C10, do estádio de maturação intermediário, apresentaram nota global média (NG) de 85,0 pontos para o café produzido na altitude de 528,0 m e para os frutos desmucilados, correspondendo a um grau de pertinência fuzzy (IF) de 0,50, classificados como café fino. Estes mesmos clones apresentaram dissimilaridades na bebida produzida pelos frutos desmucilados, com melhor qualidade para o café na maior altitude. A classificação fuzzy associada à análise de agrupamento na interpretação das notas globais médias (NG) da análise sensorial da bebida para os diferentes tratamentos estudados identificou dissimilaridade na qualidade da bebida.

Termos para indexação: Análise sensorial, Coffea canephora, diferentes altitudes.

# 1 INTRODUCTION

Analysis of the beverage quality of conilon coffee has been the subject of research due to the demands of the domestic and international markets, resulting in value being added to the product based on sensory analysis. Several studies have shown improvements in the sensory quality of the blends without compromising the quality desired by the consumer, as well as in the direct consumption of conilon coffee.

The quality of coffee can be defined as being the sum of all attributes that meet the needs of the consumer (THOMAZINI Eet al. 2011). For conilon coffee, its quality is related to the criteria demanded or even imposed by the market, such

as social and environmental criteria, and those of sustainability, origin, food security, and physical characteristics, in addition to the characteristics of the beverage (PALACIN et al, 2009).

Sensory methods in evaluating food and beverage quality are based on responses to the stimuli that produce sensations, such as: intensity, extension, duration, quality, pleasure and dissatisfaction. While stimuli can be measured by physical and chemical methods, sensations are measured through psychological processes (LANZILLOTTI; LANZILLOTTI, 1999). Borjes, Cavalli and Proença (2010) affirmed that sensory quality of a food relates, at the same time, with the food and with the physiological, psychological and sociological of the appraiser.

<sup>&</sup>lt;sup>1,2</sup> Universidade Federal do Espirito Santo / UFES - Departamento de Engenharia Rural - Cx P. 16 - 29.500-000 - Alegre-ES - limajss@yahoo.com.br, samuel.assilva@gmail.com

One of the requirements of sensory analysis is a decision about product quality for acceptance in the consumer market, grading the product on a hedonic scale according to a determined protocol that in the case of conilon coffee includes the Coffee Quality Institute (CQI) and the cupping tests carried out by qualified tasters (R-Grader).

Since human perception of quality is possibly undefinable, fuzzy sets that are not limited to a deterministic value have been used as an alternative to the traditional methods of evaluating the results of sensory analysis. As a result, fuzzy logic has been used as a tool in decisionmaking (LANZILLOTTI; LANZILLOTTI, 1999; LAZIM; SURIANI, 2009; CAVALCANTI et al., 2013; CHOROBURA; CASTANHO; TEIXEIRA, 2016). This method aims to solve problems where information is not well defined, and is based on modelling such problems, translating into mathematical terms the imprecise information contained in the natural language expressed by linguistic variables that can be transformed (LIMA et al., 2016; SENTÜRK, 2017).

Fuzzy logic aims to treat the uncertainties contained in information; in cases where there are a large number of data, multivariate statistics such as cluster analysis have been used. This explains the variance structure of the information by the construction of linear combinations of that information and the dimensional reduction of the phenomenon under study, where different attributes are grouped together according to the dissimilarities that exist, based on the Euclidean distance between them.

Considering that human sensitivity in defining sensory characteristics by palate and smell is a subjective evaluation, it is necessary, even when following a determined protocol, to include other methods in the analysis, such as fuzzy logic and clustering, with the aim of explicit results and interactions between different tasting arrangements, so as to define quality standards, highlighting the strengths and weaknesses responsible for acceptance or non-acceptance by the market.

With this in mind, the aim of this study was to apply fuzzy classification and clustering to interpretation of the mean global scores of the results of the sensory analysis of the beverage from coffee conilon produced by 13 clones at two different altitudes, and in the post-harvest processing of the cherry fruit by natural drying (d) or depulping (p).

# 2 MATERIAL AND METHODS

The experiment was carried out in two plantations of conilon coffee, known as environments 1 (E1) and 2 (E2), the first with a mean elevation of 528.0 m, located at 20°37'31" S and 41°05'23" W in the district of São Vicente. and the other with a mean elevation of 100.0 m, located at 20°45'21" S and 41°17'05" W in the district of Pacotuba, both in the town of Cachoeiro de Itapemirim in the State of Espirito Santo, Brasil. The conilon coffee under study came from a set of clones of the Victoria INCAPER 8142 variety, comprising 13 clones classified according to the characteristic of fruit maturation: early clones: 01V (C1e), 06V (C6e), 08V (C8e), 11V (C11e) and 12V (C12e); intermediate clones: 02V (C2i), 03V (C3i), 04V (C4i), 07V (C7i), 09V (C9i) and 10V (C10i); and late clones: 05V (C5l) and 13V (C131).

The coffee fruit at the cherry stage were harvested manually and selectively by sieve. Harvesting took place from May to August of 2014, when 10 litres of fruit from each clone were harvested in each area, based on the maturation characteristic of the clones.

The harvested cherry fruit were washed and divided into two batches for the two forms of processing: a) depulped fruit (p): wet processing, with mechanical removal of the shell (epicarp) and mucilage, submerging the fruit in clean water in a plastic bucket for 24 h at room temperature to induce natural biological fermentation, the samples were then washed with clean water and placed for pre-drying for 24 h on raised beds to obtain the depulped fruit; and b) natural-dried fruit (d): the fruit with the shell (epicarp) were submerged in clean water, washed, and left for 24 h on raised beds for pre-drying. After both treatments, the fruits were dried in an air circulation and renewal oven at  $45^{\circ}$ C ( $\pm 2^{\circ}$ C) to a water content of 11.5%bu ( $\pm 1\%$ ).

The dried samples were placed in paper bags and stored for 45 days in a sealed polystyrene box protected against variations in the environment (RIBEIRO, 2013, SILVA et al., 2015). The samples were then peeled by mechanical peeler and 0.30 kg of raw grain from each sample was packed in a silver metallised stand-up pouch.

The sensory analysis was performed by a team of three specialised R-Graders certified by the Coffee Quality Institute (CQI) (2011). The analysis was prepared, and evaluated by cupping test, as per the methodology of the tasting protocol for *Coffea canephora* Pierre ex A.Froehner (ICO, 2016).

The associative function of fuzzy classification was applied to the mean global score results (GS) of the coffee beverages for the 13 clones, at two altitudes and in two post-harvest processes (drying and depulping), a total of 52 treatments, to determine the degree of linear pertinence (DP) (fuzzy index = FI), as used by Silva and Lima (2009), Souza et al. (2009) and Lima et al. (2018) with soil attributes, as follows:

$$MF_A(Z) = 0$$
 se  $z < p$  (1)

$$MF_A(Z) = \left(\frac{1}{\alpha}\right) * (z-p) \quad se \ p \le z < q$$
 (2)

$$MF_{A}(Z) = 1$$
  $se \ z \ge q$  (3)

where:  $MF_A$  = degree of pertinence (DP = FI) of element Z (beverage) to the fuzzy set;  $\alpha = q-p$ ; q and p = highest and lowest value of set A (clones, altitude and processes) respectively.

The values of p and q were defined according to the classification of beverage quality as per the tasting protocol for C. canephora of the CQI (2011), UCDA (2010) and ICO (2016) (Table 1). The mean global scores (GS) in the cupping test considered in the set of the total universe were from 70.0 (minimum) to 100.0 (maximum) points. After fuzzification (standardisation) of the results of the sensory analysis, the intervals fell between zero (0.00) and 1.00 (crisp value) respectively, defining the GS (FI) for each beverage value, as shown in the graphical representation in Figure 1.

For defuzzification of the data, the following terms were used for the linguistic variable beverage quality (BQ) of conilon coffee: very good coffee (VGC), fine coffee (FC) and exceptional coffee (EC), as shown in Table 1.

The rules of inference used for defuzzification were IF and THEN. IF the degree of pertinence (FI) for each clone (C), at each altitude (A), and for each type of fruit processing (d and p) is between 0.00 to < 0.33, THEN = VGC; When the FI is between  $\geq$  0.33 to < 0.67, THEN = FC, and when the FI is between  $\geq$  0.67 to  $\leq$  1.00, THEN = EC.

In order to verify the dissimilarity between beverage results obtained by sensory analysis for the different treatments, the multivariate analysis method of hierarchical clustering (Joining) was applied, adopting the following procedures:

- i) data standardisation: in the data matrix, the value of the ith column (beverage) and jth line (clones, altitudes and processing), called Xij. The corresponding value was considered standardised in the data matrix of fuzzified values, with the data represented in degrees of pertinence (DP = FI). Standardisation was adopted so that the attributes would contribute with the same weight in calculating the coefficient of dissimilarity between them.
- ii) choice of the coefficient of similarity: the measure of dissimilarity was adopted using the Euclidean distance  $(d_{AB})$ , since the lower the value, the closer the beverage results between the different treatments. The Euclidean distance  $(d_{AB})$  was determined by equation 4, between the accessions for the attribute (beverage) set under study:

$$d_{AB} = \sqrt{(X_{1A} - X_{1B})^2 + (X_{2A} - X_{2B})^2 + \dots + (X_{mA} - X_{mB})^2}$$
 (4)

where:  $X_{1A}$  = value of the attribute (beverage) of clone 1 for condition one (1,0) A;  $X_{1B}$  = value of the attribute (beverage) of clone 1 for condition two (2) B;  $X_{mA}$  and  $X_{mB}$  = value of the attribute (beverage) of clone m for condition A and B.

iii) Grouping strategy: Ward's method was used, which forms groups by seeking to minimise the sum of the differences between the elements of each group and the mean value of the group, minimising the standard deviation between the data of each formed group.

# **3 RESULTS AND DISCUSSION**

The mean global scores (GS) obtained from the sensory analysis (SA), the cupping test, are shown in Table 2, together with their respective degrees of pertinence (DP = FI) after applying the fuzzy classification.

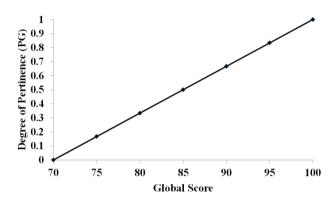
In environment 1 (E1), for the clonal conilon coffee in the depulping process (p), the maximum GS was 85.0 for C4i and C10i, both clones with an intermediate stage of maturation. For the natural-dried fruit (d), C4i again had the highest GS, with 84.0 points.

In environment 2 (E2), the highest GS for the depulped fruits (p) was 80.0 points, obtained by C2i, C4i and C5l. For the natural-dried fruits (d), clones C12e, C3i and C5l had a GS of 80.0 points and C4i a score of 82 points. In general, clone C4i presented the highest GS irrespective of the environment (altitude) or process. As this is an intermediate clone (i), it can be inferred that this fruit maturation stage indicates suitable plant physiology for favouring a better beverage.

TABLE 1 - Classification of beverage quality for Coffea canephora

Mean global score	Quality description	
$90.0 \le EC \le 100.0$	Exceptional coffee (EC)	
$80.0 \le FC < 90.0$	Fine coffee (FC)	
$70.0 \le VGC < 80.0$	Very good coffee (VGC)	

Source: Adapted from CQI (2011), UCDA (2010) e ICO (2016)



**FIGURE 1** - Graphical representation of the pertinence function for the scale of quality scores of conilon coffee.

**TABLE 2** - GS results for each clone at the two altitudes in the processes of depulping (p) and natural drying (d), with their respective degrees of pertinence (DP = FI) (fuzzy) for beverage potential.

Cle	C6e	C8e	C11e	C12e	C2i	C31	i C4	i (	C7i (	C9i	C10i	C51	C131
Mean global score (GS)													
80.	0 80.	0 77	'.0 82	.0 80	.0 8	1.0	82.0	85.0	79.0	81.0	85.0	83.0	80.0
79.	0 79.	0 78	3.0 76	.0 77	.0 8	0.0	78.0	80.0	78.0	79.0	79.0	80.0	74.0
80.	0 78.	0 81	.0 79	.0 80	.0 8	1.0	83.0	84.0	79.0	80.0	81.0	82.0	79.0
79.	0 77.	0 79	0.0 77	.0 80	.0 7	9.0	80.0	82.0	77.0	79.0	78.0	80.0	71.0
				Degree	e of per	tinenc	e (DP=	FI)					
0.3	3 0.3	3 0.2	23 0.4	10 0.3	33 0	.37	0.40	0.50	0.30	0.37	0.50	0.43	0.33
0.3	0 0.3	0 0	27 0.2	20 0.2	23 0	.33	0.27	0.33	0.27	0.30	0.30	0.33	0.13
0.3	3 0.2	7 0	37 0.3	30 0.3	33 0	.37	0.43	0.47	0.30	0.33	0.37	0.40	0.30
0.3	0 0.2	3 0	30 0.2	23 0.3	33 0	.30	0.33	0.40	0.23	0.30	0.27	0.33	0.03
	80. 79. 80. 79. 0.3 0.3	80.0 80. 79.0 79. 80.0 78. 79.0 77. 0.33 0.3 0.30 0.3 0.33 0.2	80.0 80.0 77 79.0 79.0 78 80.0 78.0 81 79.0 77.0 79  0.33 0.33 0.3 0.30 0.30 0.3 0.33 0.27 0.3	80.0 80.0 77.0 82 79.0 79.0 78.0 76 80.0 78.0 81.0 79 79.0 77.0 79.0 77 0.33 0.33 0.23 0.4 0.30 0.30 0.27 0.2 0.33 0.27 0.37 0.3	Me 80.0 80.0 77.0 82.0 80 79.0 79.0 78.0 76.0 77 80.0 78.0 81.0 79.0 80 79.0 77.0 79.0 77.0 80  Degree  0.33 0.33 0.23 0.40 0.3 0.30 0.30 0.27 0.20 0.3 0.33 0.27 0.30 0.30	Mean glob  80.0 80.0 77.0 82.0 80.0 8  79.0 79.0 78.0 76.0 77.0 8  80.0 78.0 81.0 79.0 80.0 8  79.0 77.0 79.0 77.0 80.0 7  Degree of per  0.33 0.33 0.23 0.40 0.33 0  0.30 0.30 0.27 0.20 0.23 0  0.33 0.27 0.37 0.30 0.33 0	Mean global sco  80.0 80.0 77.0 82.0 80.0 81.0  79.0 79.0 78.0 76.0 77.0 80.0  80.0 78.0 81.0 79.0 80.0 81.0  79.0 77.0 79.0 77.0 80.0 79.0  Degree of pertinence  0.33 0.33 0.23 0.40 0.33 0.37  0.30 0.30 0.27 0.20 0.23 0.33  0.33 0.27 0.37 0.30 0.33 0.37	Mean global score (GS)  80.0 80.0 77.0 82.0 80.0 81.0 82.0  79.0 79.0 78.0 76.0 77.0 80.0 78.0  80.0 78.0 81.0 79.0 80.0 81.0 83.0  79.0 77.0 79.0 77.0 80.0 79.0 80.0  Degree of pertinence (DP=  0.33 0.33 0.23 0.40 0.33 0.37 0.40  0.30 0.30 0.27 0.20 0.23 0.33 0.27  0.33 0.27 0.37 0.30 0.33 0.37 0.43	Mean global score (GS)           80.0         80.0         77.0         82.0         80.0         81.0         82.0         85.0           79.0         79.0         78.0         76.0         77.0         80.0         78.0         80.0           80.0         78.0         81.0         79.0         80.0         81.0         83.0         84.0           79.0         77.0         77.0         80.0         79.0         80.0         82.0           Degree of pertinence (DP=FI)           0.33         0.33         0.23         0.40         0.33         0.37         0.40         0.50           0.30         0.30         0.27         0.20         0.23         0.33         0.27         0.33           0.33         0.27         0.37         0.30         0.33         0.37         0.43         0.47	Mean global score (GS)           80.0         80.0         77.0         82.0         80.0         81.0         82.0         85.0         79.0           79.0         79.0         78.0         76.0         77.0         80.0         78.0         80.0         78.0           80.0         78.0         81.0         79.0         80.0         81.0         83.0         84.0         79.0           79.0         77.0         77.0         80.0         79.0         80.0         82.0         77.0           Degree of pertinence (DP=FI)           0.33         0.33         0.23         0.40         0.33         0.37         0.40         0.50         0.30           0.30         0.30         0.27         0.20         0.23         0.33         0.27         0.33         0.27         0.30           0.33         0.27         0.37         0.30         0.33         0.37         0.43         0.47         0.30	Mean global score (GS)           80.0         80.0         77.0         82.0         80.0         81.0         82.0         85.0         79.0         81.0           79.0         79.0         78.0         76.0         77.0         80.0         78.0         80.0         78.0         79.0           80.0         78.0         81.0         79.0         80.0         81.0         83.0         84.0         79.0         80.0           79.0         77.0         79.0         79.0         80.0         82.0         77.0         79.0           Degree of pertinence (DP=FI)           0.33         0.33         0.23         0.40         0.33         0.37         0.40         0.50         0.30         0.37           0.30         0.30         0.27         0.20         0.23         0.33         0.27         0.33         0.47         0.30         0.33           0.33         0.27         0.37         0.30         0.33         0.37         0.43         0.47         0.30         0.33	Mean global score (GS)         80.0       80.0       77.0       82.0       80.0       81.0       82.0       85.0       79.0       81.0       85.0         79.0       79.0       78.0       76.0       77.0       80.0       78.0       80.0       78.0       79.0       79.0       79.0       80.0       81.0       83.0       84.0       79.0       80.0       81.0         79.0       77.0       79.0       79.0       80.0       82.0       77.0       79.0       78.0         Degree of pertinence (DP=FI)         0.33       0.33       0.23       0.40       0.33       0.37       0.40       0.50       0.30       0.37       0.50         0.30       0.30       0.27       0.20       0.23       0.33       0.27       0.33       0.27       0.30       0.33       0.37       0.43       0.47       0.30       0.33       0.37	Mean global score (GS)           80.0         80.0         77.0         82.0         80.0         81.0         82.0         85.0         79.0         81.0         85.0         83.0           79.0         79.0         78.0         76.0         77.0         80.0         78.0         80.0         79.0         79.0         79.0         80.0         81.0         82.0         79.0         80.0         81.0         82.0         79.0         80.0         81.0         82.0         79.0         80.0         81.0         82.0         79.0         79.0         78.0         80.0         80.0         82.0         77.0         79.0         78.0         80.0         80.0         82.0         77.0         79.0         78.0         80.0         80.0         80.0         82.0         77.0         79.0         78.0         80.0         80.0         80.0         82.0         77.0         79.0         78.0         80.0         80.0         80.0         82.0         77.0         79.0         78.0         80.0         80.0         80.0         80.0         80.0         80.0         80.0         80.0         80.0         80.0         80.0         80.0         80.0         80.0         <

C: clone; e: early; i: intermediate; l: late; A1: area at an average altitude of 528.0 m; A2: area at an average altitude of 100.0 m; p: depulped coffee fruit and d: natural-dried coffee fruit

From the results obtained with the fuzzy inference (Table 2), it can be seen that the highest values for the fuzzy index (FI) are in the clones that obtained the highest GS due to adoption of the linear model.

At altitude A1, according to the inference adopted for 'very good coffee' (VGC) (FI between 0.00 and < 0.33) in the depulped condition (p), THEN includes clones C8e (FI = 0.23) and C7i (FI = 0.30), while the remainder are THEN grouped

in the fine coffee class (FC), with FI between  $\geq$  0.33 and < 0.67. For the natural-dried fruit (d), clones C6e (FI = 0.27), C11e (FI = 0.30), C7i (FI = 0.30) and C13l (FI = 0.30) are THEN classed as VGC with FI between 0.00 and < 0.33. The other clones are THEN in the FC class, with FI between  $\geq$  0.33 and < 0.67. This is related to the natural fruit-drying process (d) that contributed to a fall in beverage quality, which increased the number of clones in the VGC class.

At altitude 2 (A2), clones C2i, C4i and C5l for the depulped fruits (p) are THEN grouped in the FC class, with FI equal to 0.33. For the natural-dried fruits (d), clones C12e (FI = 0.33), C3i (FI = 0.33), C4i (FI = 0.40) and C5l (FI = 0.33) are THEN grouped in the FC class. These results indicate to the producer that, a priori, the work of depulping the fruit is not viable when the aim is to improve beverage quality.

Depulping the fruit of the clonal coffee produced at the higher altitude provided 11 of the 13 clones in the fine coffee class (FC). It should be noted that under the conditions of the study, none of the clones were classified as 'exceptional coffee' (EC).

In Figure 2 it is shown the distribution of the fuzzy index (FI) for the 13 clones, where it can be seen that the two internal curves refer to the results obtained at altitude 2 (A2), for both the depulped (p) and the natural-dried coffee (d). The product obtained from the sensory analysis (SA) highlighted the C4i and C10i clones as having an FI of 0.50 when the fruits were depulped (p), showing them to be half the value of an exceptional coffee (EC).

Despite the greatest values for FI at altitude 1 having been seen for all samples in the depulped clones, the difference for the same clones when natural-dried was small, showing that the post-harvest processing to be adopted by the producer should be well thought out, taking into account the market price of quality coffees. Lima Filho et al. (2013) evaluated the physico-chemical and sensory quality of conilon coffee processed by different methods and concluded it was possible to obtain superior beverages for natural-dried coffees.

More important than the post-harvest processing of conilon coffee appears to be the maturation stage, since greater values for FI were seen in the intermediate clones. Early clones had the smallest overall scores, showing that the reddish colour of the shell did not ensure the physiological maturation of the fruit and thus provide better beverage quality, seeing that the harvest was selective and the sensory analysis considered only the cherry fruit.

Similar behaviour to that discussed for the early clones (e) was seen in the late (l) clones. For the C131 clone in A2, the GS and the FI were lower in the dried samples, also showing little effect from depulping on improving beverage quality.

For the combinations of clonal coffee produced in A1 when the fruits were depulped (p) and natural-dried (d), 84.6% and 69.2% respectively were in the FC class. Whereas for the

coffee produced in A2 under both methods of fruit processing, 23.1% were in the FC class. These results reinforce the idea that beverage quality, as measured in the sensory analysis, shows a gain for clonal conilon coffee in area A1 when depulped (p). According to Silva et al. (2014), the environment has a great effect on the quality of coffee beverages, contributing to a qualitative improvement in the roasted beans, regardless of the variety grown.

Crops grown at higher altitudes tend to produce sweeter coffees with higher quality scores, and can be discriminated from those cultivated at lower altitudes (SILVA et al., 2016). Ricci, Cocheto Junior and Almeida (2013) explained that, among other factors, those of climate are the most important for the expression of the qualitative standards of coffees grown in environments at higher altitudes. According to those authors, maturation occurs more slowly due to the lower temperatures, which favours the physiological development of the fruit.

In the cluster analysis, a cut-off distance of 0.40 was defined for each treatment under study, forming three groups of clone accessions. For any one clone, its presence in different groups defined it as being dissimilar in relation to beverage for the treatment under consideration. The dissimilarities that exist (Table 3) for the depulped fruit (p) when produced at different altitudes were determined from the Euclidean distance  $\geq 0.14$ . That is, clones C11e, C12e, C3i, C4i, C10i, C5l and C13l have higher GS when produced at altitude 1 (A1) and with the fruit depulped (p). When considering the maturation stage, clones C11e and C13l showed a greater reduction (greater  $d_{AB}$ ) for beverage in relation to the change in altitude. This greater  $d_{AB}$  is reflected in the highest and lowest values for FI between the two processes employed: the clones C4ip (i = intermediate, p = depulped) and C10ip, compared to C13ld (l = late, d = dried).

Silva et al. (2014), delimiting terroirs in plantations of arabica coffee, verified that at higher altitudes there are microclimates that are more propitious to the production of special coffees. The quality scores tend to be more homogeneous, a similar behaviour to that seen in this study for conilon coffee. Broadly speaking, conilon coffee plants are grown in areas of lower altitude and higher temperature, which has traditionally meant lower sensory quality. It is evident from the results of the above work, that cultivation at higher altitudes, as in arabica coffee, also favours an increase in the beverage quality of conilon coffee.

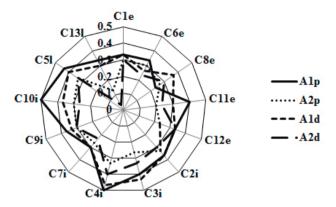


FIGURE 2 - Radar chart of the fuzzy classification (FI) of GS obtained in the sensory analysis of the beverage from clonal conilon coffee.

**TABLE 3** - Euclidean distance  $(d_{AB})$  for the GS for beverage (fuzzified) of clones in the depulping process (p) at different altitudes (A1 and A2)

	C1A2	C6A2	C8A2	C11A2	C12A2	C2A2	C3A2	C4A2	C7A2	C9A2	C10A2	C5A2	C13A2
C1A1	0.05	0.05	0.09	0.19*	0.14*	0.00	0.09	0.00	0.09	0.05	0.05	0.00	0.28*
C6A1	0.05	0.05	0.09	0.19*	0.14*	0.00	0.09	0.00	0.09	0.05	0.05	0.00	0.28*
C8A1	0.09	0.09	0.05	0.05	0.00	0.14*	0.05	0.14*	0.05	0.09	0.09	0.14*	0.14*
C11A1	0.14*	0.14	0.19*	0.28*	0.24*	0.09	0.19*	0.09	0.19*	0.14*	0.14*	0.09	0.38*
C12A1	0.05	0.05	0.09	0.19*	0.14*	0.00	0.09	0.00	0.09	0.05	0.05	0.00	0.28*
C2A1	0.09	0.09	0.14*	0.24*	0.19*	0.05	0.14	0.05	0.14*	0.09	0.09	0.05	0.33*
C3A1	0.14*	0.14	0.19*	0.28*	0.24*	0.09	0.19*	0.09	0.19*	0.14*	0.14*	0.09	0.38*
C4A1	0.28*	0.28	0.33*	0.42*	0.38*	0.24*	0.33*	0.24*	0.33*	0.28*	0.28*	0.24*	0.52*
C7A1	0.00	0.00	0.05	0.14*	0.09	0.05	0.05	0.05	0.05	0.00	0.00	0.05	0.24*
C9A1	0.09	0.09	0.14*	0.24*	0.19*	0.05	0.14*	0.05	0.14*	0.09	0.09	0.05	0.33*
C10A1	0.28*	0.28	0.33*	0.42*	0.38*	0.24*	0.33*	0.24*	0.33*	0.28*	0.28*	0.24*	0.52*
C5A1	0.19*	0.19	0.24*	0.33*	0.28*	0.14*	0.24*	0.14*	0.24*	0.19*	0.19*	0.14*	0.42*
<u>C13A1</u>	0.05	0.05	0.09	0.19*	0.14*	0.00	0.09	0.00	0.09	0.05	0.05	0.00	0.28*

C: clone; A1: production area at the higher altitude and A2: production area at the lower altitude; \*: Euclidean distance that shows dissimilarity between clones

In the environment at the higher altitude (A1), considering the two methods of fruit processing, the dissimilarities that exist (Table 4) for the depulped (p) and natural-dried fruit (d) at altitude 1 (A1) were determined from the Euclidean distance  $\geq 0.14$ . It can be seen that only clones C8e, C11e and C10i showed dissimilarity for beverage. The C8e clone presented a beverage gain in the drying process, and the C4i clone showed no dissimilarity between processes, maintaining the highest score in both, thereby forming part of a single group. The absence of dissimilarity for the two processes reinforces the above comment concerning the small qualitative gain obtained when depulping the fruit of conilon coffee. Because it is a less laborious process and requires less care, and provided that only cherry fruits are harvested, the results of this research point to the recommendation of natural processes (natural-dried coffee), taking into consideration the maturation stage, unless the financial gain for

depulped coffee is far greater for a small variation in GS.

Table 5 shows the Euclidean distances ( $d_{AB}$ ) for the fuzzified quality of the natural-dried (d) coffee fruit from clones produced at altitude 1 (A1) and altitude 2 (A2) respectively.

In these treatments, distances ≥ 0.09 characterised a variation in the final beverage value, representing dissimilarities for clones C8e, C11e, C2i, C3i, C4i, C7i, C10i, C5l and C13l, the greatest distance being obtained for clone C13t, with a value of 0.38. This characterises a similar behaviour when the fruits were depulped and compared between environments, i.e. the beverage potential for this clone is higher when produced at the higher altitude; in A1 and depulped the clone was classified as FC, and when dried, as VGC. The classes that describe coffee quality in the classification key (Table 1) represent financial gain to the producer, an important point to be kept in mind.

Table 6 shows the results for Euclidean distance  $\geq 0.09$ , which presented dissimilarity for the fuzzified beverage in clones of conilon coffee at altitude 2, for the depulped dried fruit (p) and the natural-dried fruit (d).

Among all the clones under study, C12e and C4i at altitude 2 (A2) had a Euclidean distance of 0.14 and 0.09 respectively, indicating

dissimilarity between the natural-drying method of fruit processing in relation to depulping, i.e. with an increase in GS for the natural-dried coffee. A shorter Euclidean distance can be seen in environment 2 for any one clone in the different processes, indicating that depulping is not a practice to be used in processing the fruit.

**TABLE 4** - Euclidean distance between the GS for beverage (fuzzified) of clones at altitude 1 (A1) in the processes of depulping (p) and natural drying (d).

	Cld	C61d	C81d	C11d	C12d	C2d	C3d	C4d	C7d	C9d	C10d	C5d	C13d
Clp	0.00	0.09	0.05	0.05	0.00	0.05	0.14*	0.19*	0.05	0.00	0.05	0.09	0.05
C6p	0.00	0.09	0.05	0.05	0.00	0.05	0.14*	0.19*	0.05	0.00	0.05	0.09	0.05
C8p	0.14*	0.05	0.19*	0.09	0.14*	0.19*	0.28*	0.33*	0.09	0.14*	0.19*	0.24*	0.09
Cllp	0.09	0.19*	0.05	0.14*	0.09	0.05	0.05	0.09	0.14*	0.09	0.05	0.00	0.14*
C12p	0.00	0.09	0.05	0.05	0.00	0.05	0.14*	0.19*	0.05	0.00	0.05	0.09	0.05
$C2p^{1}$	0.05	0.14*	0.00	0.09	0.05	0.00	0.09	0.14*	0.09	0.05	0.00	0.05	0.09
C3p C4p	0.09	0.19*	0.05	0.14*	0.09	0.05	0.05	0.09	0.14*	0.09	0.05	0.00	0.14*
C4p	0.24*	0.33*	0.19*	0.28*	0.24*	0.19*	0.09	0.05	0.28*	0.24*	0.19*	0.14*	0.28*
C7p	0.05	0.05	0.09	0.00	0.05	0.09	0.19*	0.24*	0.00	0.05	0.09	0.14*	0.00
C9p	0.05	0.14*	0.00	0.09	0.05	0.00	0.09	0.14*	0.09	0.05	0.00	0.05	0.09
C10p	0.24*	0.33*	0.19*	0.28*	0.24*	0.19*	0.09	0.05	0.28*	0.24*	0.19*	0.14*	0.28*
C5p <sup>1</sup>	0.14*	0.24*	0.09	0.19*	0.14*	0.09	0.00	0.05	0.19*	0.14*	0.09	0.05	0.19*
C13p	0.00	0.09	0.05	0.05	0.00	0.05	0.14	0.19*	0.05	0.00	0.05	0.09	0.05

C: clones; p: dried depulped fruit and d: natural-dried fruit; \*: Euclidean distance that shows dissimilarity between clones

**TABLE 5** - Euclidean distance between the GS for beverage (fuzzified) of natural-dried (d) clones at altitude 1 (A1) and altitude 2 (A2)

	C1A2	C6A2	C8A2	C11A2	C12A2	C2A2	C3A2	C4A2	C7A2	C9A2	C10A2	C5A2	C13A2
C1A1	0.05	0.14*	0.05	0.14*	0.00	0.05	0.00	0.09*	0.14*	0.05	0.09*	0.00	0.42*
C6A1	0.05	0.05	0.05	0.05	0.09	0.05	0.09*	0.19*	0.05	0.05	0.00	0.09*	0.33*
C8A1	0.09*	0.19*	0.09*	0.19*	0.05	0.09*	0.05	0.05	0.19*	0.09*	0.14*	0.05	0.47*
C11A1	0.00	0.09*	0.00	0.09*	0.05	0.00	0.05	0.14*	0.09	0.00	0.05	0.05	0.38*
C12A1	0.05	0.14*	0.05	0.14*	0.00	0.05	0.00	0.09*	0.14*	0.05	0.09*	0.00	0.42*
C2A1	0.09*	0.19*	0.09*	0.19*	0.05	0.09*	0.05	0.05	0.19*	0.09*	0.14*	0.05	0.47*
C3A1	0.19*	0.28*	0.19*	0.28*	0.14*	0.19*	0.14*	0.05	0.28*	0.19*	0.24*	0.14*	0.57*
C4A1	0.24*	0.33*	0.24*	0.33*	0.19*	0.24*	0.19*	0.09*	0.33*	0.24*	0.28*	0.19*	0.61*
C7A1	0.00	0.09*	0.00	0.09*	0.05	0.00	0.05	0.14*	0.09*	0.00	0.05	0.05	0.38*
C9A1	0.05	0.14*	0.05	0.14*	0.00	0.05	0.00	0.09*	0.14*	0.05	0.09*	0.00	0.42*
C10A1	0.09*	0.19*	0.09*	0.19*	0.05	0.09*	0.05	0.05	0.19*	0.09*	0.14*	0.05	0.47*
C5A1	0.14*	0.24*	0.14*	0.24*	0.09*	0.14*	0.09*	0.00	0.24*	0.14*	0.19*	0.09*	0.52*
C13A1	0.00	0.09*	0.00	0.09*	0.05	0.00	0.05	0.14*	0.09*	0.00	0.05	0.05	0.38*

C: clone; A1: production area at the higher altitude and A2: production area at the lower altitude; \*: Euclidean distance that shows dissimilarity between clones

**TABLE 6** - Euclidean distance between the GS for beverage (fuzzified) of clones grown at altitude 2 (A2) for depulped (p) and natural-dried (d) fruit

	C1d	C6d	C8d	C11d	C12d	C2d	C3d	C4d	C7d	C9d	C10d	C5d	C13d
Clp	0.00	0.09*	0.00	0.09*	0.05	0.00	0.05	0.14*	0.09*	0.00	0.05	0.05	0.38*
C6p	0.00	0.09*	0.00	0.09*	0.05	0.00	0.05	0.14*	0.09*	0.00	0.05	0.05	0.38*
C8p	0.05	0.05	0.05	0.05	0.09*	0.05	0.09*	0.19*	0.05	0.05	0.00	0.09*	0.33*
Cllp	0.14*	0.05	0.14*	0.05	0.19*	0.14*	0.19*	0.28*	0.05	0.14*	0.09*	0.19*	0.24*
C12p	0.09*	0.00	0.09*	0.00	0.14*	0.09*	0.14*	0.24*	0.00	0.09*	0.05	0.14*	0.28*
C2p <sup>1</sup>	0.05	0.14*	0.05	0.14*	0.00	0.05	0.00	0.09*	0.14*	0.05	0.09*	0.00	0.42*
C3p	0.05	0.05	0.05	0.05	0.09*	0.05	0.09*	0.19*	0.05	0.05	0.00	0.09*	0.33*
C4p	0.05	0.14*	0.05	0.14*	0.00	0.05	0.00	0.09*	0.14*	0.05	0.09*	0.00	0.42*
C7p	0.05	0.05	0.05	0.05	0.09*	0.05	0.09*	0.19*	0.05	0.05	0.00	0.09*	0.33*
C9p	0.00	0.09	0.00	0.09*	0.05	0.00	0.05	0.14*	0.09*	0.00	0.05	0.05	0.38*
C10p	0.00	0.09	0.00	0.09*	0.05	0.00	0.05	0.14*	0.09*	0.00	0.05	0.05	0.38*
C5p <sup>2</sup>	0.05	0.14*	0.05	0.14*	0.00	0.05	0.00	0.09*	0.14*	0.05	0.09*	0.00	0.42*
<u>C13p</u>	0.24*	0.14*	0.24*	0.14*	0.28*	0.24*	0.28*	0.38*	0.14*	0.24*	0.19*	0.28*	0.14*

C: clone; p: depulped fruit; c: natural-dried fruit; \*: Euclidean distance that shows dissimilarity between clones

# **4 CONCLUSIONS**

In the sensory analysis, the clones of conilon coffee (Vitoria) present different global scores for beverage depending on altitude and post-harvest fruit processing. Clones 4 (C4i) and 10 (C10i), of intermediate maturation, had a score of 85.0 for the depulped fruit, cultivated at an altitude of 528.0 m.

In the fuzzy classification, 84.6% and 69.2% of the different clones produced in environment 1 (altitude of 528.0 m) were classified as 'fine coffee' (FC) for the depulped (p) and natural-dried (d) fruit respectively. In the A2 environment (altitude of 100.0 m), 23.1% were obtained in the two processes with the clones classified as 'fine coffee' (FC).

The fuzzy classification taken together with the cluster analysis to interpret the global scores in the sensory analysis of the beverage for the different treatments under study identified dissimilarity in beverage quality between the different clones.

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# OCCUPATIONAL NOISE LEVEL IN MECHANIZED AND SEMIMECANIZED HARVEST OF COFFEE FRUITS

José Antonio Ramos da Silva<sup>1</sup>, Geraldo Gomes de Oliveira Júnior<sup>2</sup>, Carlos Emanuel de Melo Costa<sup>3</sup>, Adriano Bortolotti da Silva<sup>4</sup>, Camila Pires Cremasco Gabriel<sup>5</sup>, Fernando Ferrari Putti<sup>6</sup>

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ABSTRACT: Coffee cultivation has undergone significant changes, especially with regard to the mechanization process of the various existing operations that were previously carried out manually by the workers. It is observed that the intensification of mechanized activities can expose workers to noise levels capable of compromising their hearing health. In this sense, the objective in the present study was to determine the level of occupational noise in the activities of mechanized and semi-mechanized harvesting of coffee fruits and compare them with the limits of tolerance of the current legislation. The occupational noise level was determined considering the exposure of homogeneous groups, using an integrative meter for personal use, noise dosimeter, electromechanically calibrated and with field calibration. The results demonstrate that the noise levels found are above the limits allowed for an 8-hour working day. The highest observed level was 100.7 dB (A) in the sweeping operation with the blower equipment and the lowest level 89.0 dB (A) in the auxiliary activity of the selected collection equipment Vicon H3000. Harvesting activity with a portable mechanical stripper equipment showed a noise level 4.2% higher compared to harvesting with automotive harvester equipment.

Index terms: Coffee cultivation, mechanization, occupational risk.

# NÍVEL DE RUÍDO OCUPACIONAL NA COLHEITA MECANIZADA E SEMIMECANIZADA DOS FRUTOS DO CAFEEIRO

RESUMO: A cafeicultura tem passado por mudanças significativas, em especial no que se refere ao processo de mecanização das diversas operações existentes que anteriormente eram realizadas manualmente pelos trabalhadores. Observa-se que a intensificação das atividades mecanizadas pode expor trabalhadores a níveis de ruído capazes de comprometer sua saúde auditiva. Nesse sentido o objetivo do presente estudo foi determinar o nível de ruído ocupacional nas atividades de colheita mecanizada e semimecanizada dos frutos do cafeeiro e compara-los com os limites de tolerância da legislação vigente. O nível de ruído ocupacional foi determinado, considerando-se a exposição de grupos homogêneos, utilizando-se medidor integrador de uso pessoal, dosímetro de ruído, calibrados eletromecanicamente e com aferição de campo. Os resultados demonstram que os níveis de ruído encontrados estão acima dos limites permitidos para uma jornada de 8 horas de trabalho. O maior nível observado foi de 100,7 dB(A) na operação de varrição com o equipamento soprador e o menor nível 89,0 dB(A) na atividade de auxiliar do equipamento recolhedora selecta H3000 Vicon. A atividade de colheita com o equipamento derriçadeira portátil apresentou nível de ruído 4,07% mais elevado em relação a colheita com o equipamento colhedora automotriz.

Termos para indexação: Cafeicultura, mecanização, risco ocupacional.

# 1 INTRODUCTION

Coffee production is an important activity in Brazilian agribusiness (APARECIDO et al., 2017), responsible for a large financial movement in the country, as well as being a financial base for several municipalities and regions (FERREIRA JÚNIOR et al., 2015).

It has been verified that the activities of the coffee crop have undergone significant changes over time, especially with regard to the mechanization process of the several existing operations that were previously performed manually by the workers (CUNHA et al., 2016).

The harvesting process is one of the main coffee operations that influence the production activity, and due to the need for changes, these activities are carried out through mechanized systems, being able to be harvesters and / or mechanical stripper (SANTINATO et al., 2015; SILVA et al. 2013).

Studies have shown that the harvest yield of semi-mechanized fruits is eight times higher than the manual harvest (MATIELLO et al., 2013) and that manual harvesting is 50% to 60% more expensive than mechanized harvesting (SANTINATO et al., 2014).

<sup>&</sup>lt;sup>1,2</sup>Intituto Federal de Educação, Ciência e Tecnologia do Sul de Minas - Campus Muzambinho - Estrada de Muzambinho, Km 35, Cx. P. 02 - 37.890-000 - Muzambinho -MG - jaramos@gmail.com, geraldo.junior@muz.ifsuldeminas.edu.br

<sup>&</sup>lt;sup>3,4</sup>Universidade José do Rosário Vellano/UNIFENAS - Rodovia MG 179 Km 0 - Cx. P. 23 - 37.130-000 - Alfenas - MG - adriano.silva@unifenas.br

<sup>&</sup>lt;sup>5,6</sup> Universidade Estadual Paulista/UNESP - Campus Tupã - Av. Domingos da Costa Lopes, 780 - 17.602-496 - Tupã - SP - camila@tupa.unesp.br, fernandoputti@tupa.unesp.br

However, even contributing to the optimization of agricultural operations, the intensification of the use of mechanized equipment in the coffee harvesting process can expose workers to high levels of noise that, depending on the intensity and time of exposure, can compromise their occupational health (LIMA JÚNIOR et al., 2014; VALLONE et al., 2016).

In this way, exposure to noise over a long period can lead to hearing loss, psychological damage, sleep disturbances, cardiovascular alterations, immune system dysfunction, fatigue, irritability, also increase the risk of work-related accidents and decrease the performance of workers (SILVA et al., 2014).

Noise is a type of sound or mixture of these that has the capacity to cause harm to the health of the people who perceive it, characterized by being a set of unpleasant sounds in the ear of individuals (COSTA et al., 2015).

It is worth noting that hearing loss, in addition to the physiological and mental damages caused to the worker, may also progress to more severe stages over the years (SANTOS; ALMEIDA, 2016).

Therefore, the quantitative evaluation of the physical noise risk allows verifying that the limits of tolerance are within those allowed in the current legislation, subsidizing the selection of occupational protection measures to reduce the health damages of workers who are connected to these operations (SANT'ANA; ZANNIN, 2016).

In this sense, the objective in the present study was to determine the level of occupational noise in the mechanized and semi-mechanized harvesting activities of coffee fruits and compare them with the tolerance limits of the current legislation, since the tolerance limit is 85 dB (A) for an 8-hour workday and 82 dB (A) for the action level, according to Regulatory Standard NR 15 and Occupational Hygiene Standard NHO 01.

# 2 MATERIAL AND METHODS

The present case study was carried out at São Manoel Farm, located in the municipality of Muzambinho, in the south of Minas Gerais, with a total area of 98 ha, of which 60 ha are occupied by coffee cultivation, whose average annual production is 2,400 bags per year.

Data collection was carried out during coffee harvesting activities from July to August 2017, in the so-called "Café do Curral", red catuaí 144 variety, spacing 3.5 x 0.8 m; "Café da Casa", red catuaí 144, spacing 3.5 x 0.8 m; "Café da Paineira", red catuaí 144, spacing 3.6 x 0.8 m and "Café do Coqueiro", acaiá variety, spacing 2.5 x 1.0 m.

The evaluation was performed by means of the quantification of the occupational noise level for the activities developed in the mechanized and semi-mechanized harvesting stage, using the automotive harvester Electron Auto TDI, MWM D229-4 model, with booth, 2012 manufacturing, 4 engine cylinders, power of 67 hp, working with a rotation of 1800 rpm; the portable mechanical stripper Shindaiwa, C230 model, with 22.5/1.4 displacement (cc/Pol<sup>3</sup>), with power/rpm of 1.2 hp/7500 rpm; homemade blower, with SWZ blower turbine, with working rotation of 2200 rpm; the SWZ picker, B-900 model, 2016 manufacturing, work rotation of 1450 rpm and the Vicon picker, model Selecta H3000 Bag Bag. manufacturing 2011, work rotation 2200 rpm and an auxiliary worker positioned at the coffee exit in the Vicon picker (Figure 1).

The implements, blower and pickers SWZ and Vicon were dragged by the Yanmar tractor, 1155-4 SR model, year of manufacture 2010, power of 40.5 kw (55 hp), unmanned.

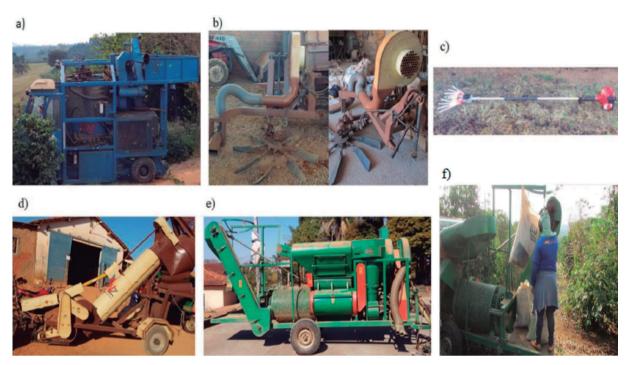
The data collection was done in a way to represent the working day, respecting the meal schedule, totaling 8 hours of work a day.

The noise quantification was developed in order to characterize the exposure of workers through homogeneous groups, considering the following treatments: Harvester, Blower; SWZ picker; Vicon picker, Vicon auxiliary picker and Shindaiwa C230 picker.

For the data collection, 21 (twenty-one) dosimetry repetitions were performed for the picker, 5 (five) for the harvester, blower, pickers and Vicon auxiliary picker.

For this study, the noises were classified as continuous and for their evaluation was used personal integrator meter, noise dosimeter DOS-100 model of SKILL TEC brand, which were calibrated electromechanically with certificate of the Brazilian Calibration Network (RBC) and calibration before and after the evaluation, using the CAL-4000 calibrator, INSTRUTHERM, IEC 942/CLASSE 2, with output sound pressure levels of 94 and 114 dB.

The dosimeter was set up according to the following parameters: reference criterion of 85 dB (A), which corresponds to the 100% dose for an 8 hours exposure, integration threshold level of 80 dB(A), increased dose doubling of 3 dB (q-3) and indication of occurrence levels above 115 dB (A) (GIAMPAOLI et al., 2001).



**FIGURE 1** - Equipment used in mechanized and semi-mechanized harvesting coffee fruits: a) automotive harvester Electron Auto TDI, b) homemade blower, c) portable mechanical stripper, d) SWZ picker, e) Vicon picker and f) auxiliary worker position in the Vicon picker.

Throughout the data collection process, a wind shield was used in the dosimeter microphone to avoid possible air velocity interference and to protect the microphone against dust, as recommended by Giampaoli et al. (2001).

The noise dosimeter was installed with the microphone positioned in the auditory zone, near the ear of the worker. After the evaluation and data collection, the SEL- Standard Exposure Level in dB (A) was determined, using the following expression:

$$SEL = EL + 10 \log \left(\frac{T_E}{480}\right) [dB(A)] \tag{1}$$

On what,

EL - is the exposure level: it is the average representative level of daily occupational exposure; and

 $T_{\rm E}$  - is the duration time, in minutes, of the daily workday.

The Exposure Level (EL) - was calculated by the following formula:

$$EL = 10 \log \left[ \left( \frac{480}{T_E} \right) x \left( \frac{D}{100} \right) \right] + 85 \tag{2}$$

On what,

EL - is the level of exposure;

 $T_{\scriptscriptstyle\rm E}$  - is the duration time, in minutes, of the daily workday.

D - Daily dose of noise in percentage;

For this criterion it is considered as action level the SEL value equal to 82 dB (A).

Note that the working day established was 8 hours daily (480 minutes) to which workers were exposed to the physical agent noise, therefore  $T_{\rm E}$  will be 480 minutes daily.

After the evaluation, the mean results obtained were compared with the tolerance limits established by the Occupational Hygiene Standard NHO 01 (GIAMPAOLI et al., 2001). From the mean data found, the maximum time of exposure to occupational noise in which the operators and auxiliary workers could be exposed without adequate ear protection was evaluated, and for the intermediate values found, the maximum allowable daily exposure was considered relative to the immediately higher level according to Occupational Hygiene Standard NHO 01 (CUNHA; TEODORO, 2006).

Data on the occupational noise levels of the mechanized and semi-mechanized harvesting activities of the coffee crop were then submitted to analysis of variance using the F test. In cases in which the F test value was significant, tests were performed comparing Tukey's mean, at the 5% level of significance. For this, the computational statistical software "SISVAR" was used.

#### **3 RESULTS AND DISCUSSION**

In the coffee mechanized harvesting, the stripping stage is carried out with automotive equipment and the sweeping and harvesting stages are performed with sweeping implements and pickers coupled to the tractor and may or may not have the presence of helpers. The results demonstrate that the continuous / intermittent noise level at all stages of the mechanized operations of the coffee harvest exceeded the reference criterion that limits the daily exposure limits, which correspond to a dose of 100% for the exposure of 8 hours at the level of 85 dB (A) (Table 1).

It was observed that the lowest noise level was found in the activity performed by the equipment's auxiliary of the Vicon picker, with 89.0 dB(A) and the highest level, in the sweeping activity, using the blower equipment, with 100.7 dB(A) (Table 1). It is worth mentioning that exposure to noise above tolerance limits can generate occupational health problems, such as hearing loss, as well as negatively influencing, reducing the efficiency of the operations (MASSA et al., 2012).

It should also be noted that the activities using the equipments: the harvester TDI MWM D229-4 and the picker Vicon Selecta H3000, as well as the activities with the picker SWZ and the operation's auxiliary of the Vicon Selecta H3000 did not present statistical differences between them (Table 1), however, all noise levels are above the tolerance limits.

It was also verified that, although the Yanmar 1155-4 SR tractor, carried out the sweeping activities with the blowing equipment and the picking activity with the selecta H3000 Vicon equipment, at the same rotation, the noise levels found presented a difference of 7.8 dB (A), demonstrating the influence of the coupled implement for the generation of the noise level.

For the stripping and harvesting of the coffee tree fruits, in addition to the use of automotive equipment, portable mechanical Stripper have been used in semi-mechanized operations, replacing the activity that was previously performed manually.

In the present study, the equipment, mechanical stripper and automotive harvester, presented noise levels above 85 dB (A) allowed for an 8-hour work day (Table 2).

The average level of occupational noise obtained in the harvesting activity with the evaluated portable mechanical stripper equipment, whose value is 97.8 dB(A), is below the values found by Cunha and Teodoro (2006) 104,6, 100,7 e 102,2 dB(A) respectively, when evaluating different types of mechanical stripper. Also, according to the same authors, when the mechanical stripper worked at idle, the average level of occupational noise reached high values approaching the acceptable maximum limit. Possibly the difference in noise levels found between the present study 2017 and the values pointed out by Cunha and Teodoro (2006) stems from the evolution in the equipment of the breakers over time, considering that the work of Cunha and Teodoro was carried out 11 years ago.

It is worth noting that the harvesting activity with the mechanical stripper equipment presented a higher level of occupational noise compared to that found in the harvesting activity using automotive harvester equipment, that is, the automotive harvester emitted 4.2% less occupational noise when compared to the mechanical stripper. According to Sales et al. (2015) the activity with the operation of the collapsible equipment can present a high potential of health risk of the workers in function of the level of noise to which they are exposed during the activities of semi-mechanized harvesting.

Therefore, comparing the exposure time and the occupational noise level found, it is verified that the workers involved in all activities of mechanized and semi-mechanized coffee harvesting cannot carry out their activities without the use of adequate ear protection, as seen that according to the Regulatory Standad NR 15 and Occupational Hygiene Standard NHO 01, the exposure of workers to occupational noise levels exceeding 85 dB (A) exceeds the tolerance limit for an 8-hour working day (BRAISL, 1978; GIAMPAOLI et al., 2001).

Also, according to Regulatory Standard NR 15, the exposure of workers to occupational noise levels of more than 85 dB(A) for an 8-hour workday, without due preventive control, requires the payment of an additional health insurance of 20% (BRASIL, 1978).

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From the analysis of Figure 2, it is possible to observe the behavior of the data in the mechanized and semi-mechanized coffee harvest stages, since these present good behavior, that is, they do not present great variation, being able to say that the data of the noise levels generated by the mechanized and semi-mechanized harvesting operations of coffee trees did not suffer significant variations (Figure 2).

Table 3 shows the maximum allowable daily times for noise exposure by workers, considering whether the levels found in each work situation

evaluated, as recommended by Occupational Hygiene Standard NHO 01 (GIAMPAOLI et al., 2001).

It is also worth noting that the exposure limit, also known as the tolerance limit, does not constitute the protection of all workers involved in the activity, since Occupational Hygiene Standard NHO 01 defines as the exposure limit the parameter that represents conditions on which it is believed that most workers may be exposed repeatedly without adverse effects on their ability to hear and understand normal conversation (GIAMPAOLI et al., 2001).

**TABLE 1** - Average noise level in mechanized coffee harvesting.

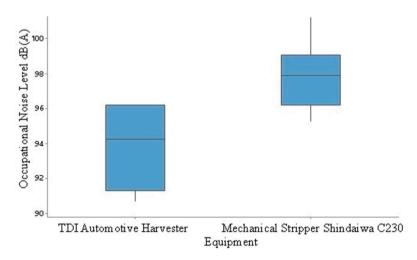
Tractor / Harvester	RPM	Implement / activity	Model	Noise dB(A	)*	CV (%) <sup>1</sup>
Yanmar 1155-4 SR	2200	Blower	Home made	100.7±0.51	a	
Automotive harvester	1800	-	TDI MWM D229-4	93.8±2.49	b	
Yanmar 1155-4 SR	2200	Picker	Vicon Selecta H3000	92.9±0.64	b	1,3
Yanmar 1155-4 SR	1450	Picker	SWZ	89.1±0.20	c	
-	-	Auxiliary picker	Vicon Selecta H3000	89.0±0.10	c	

Means followed by the same letter in the column do not differ by Tukey's test. \* They are not significant (P > 0.05) and are significant (P < 0.05). CV - Coefficient of variation.

**TABLE 2** - Comparison of mean noise level in mechanized and semi-mechanized harvesting of coffee.

Harvest type	Equipment	Noise dB(A)*	CV (%) <sup>1</sup>
Semi-mechanized	Mechanical stripper Shindaiwa	97.8±1.78 a	2.0
Mechanized	TDI Automotive Harvester	93.8±2.49 b	2,0

Means followed by the same letter in the column do not differ by Tukey's test. \* They are not significant (P > 0.05) and are significant (P < 0.05). <sup>1</sup> CV - Coefficient of variation.



**FIGURE 2** - Box-plot of noise dB (A) generated by the TDI automotive harvester and Mechanical Stripper Shindaiwa C230.

<b>TABLE 3</b> - Maximum permissible daily exposure time in the use of mechanized and semi-mechanized equip	oment
in the coffee harvesting activity according to Occupational Hygiene Standard NHO 01.	

Equipment / Activity	Noise dB(A)	Exposure time*	CV (%) <sup>1</sup>
Blower	100.7±0.51	11.90 min	
Mechanical Stripper Shindaiwa	97.8±1.78	23.81 min	
Automotive harvester	93.8±2.49	60.00 min	1.6
Picker	92.9±0.64	75.59 min	1,6
Picker	89.1±0.20	190.48 min	
Auxilary	89.0±0.10	190.48 min	

<sup>\*</sup> Approximate exposure time allowed. <sup>1</sup> CV - Coefficient of variation.

#### 4 CONCLUSIONS

Under the conditions in which the study was conducted, it was concluded that the noise levels in mechanized and semi-mechanized harvesting activities are above the limits allowed for an 8-hour workday, and individual protection devices should be used to attenuate to the tolerance limit in order to improve working conditions for the workers.

The highest noise level was found in the sweeping operation with the use of the blower equipment with 100.7 dB(A) and the lowest value in the sweeping activity with the auxiliary of the picker equipment selecta H3000 Vicon with 89.0 dB(A).

Harvesting activity with portable mechanical stripper equipment showed a 4.2% higher noise level compared to harvesting with automotive harvester equipment.

#### **5 ACKNOWLEDGEMENTS**

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#### SENSORY ANALYSIS OF COFFEE DRIED WITH AND WITHOUT STIRRING

Bruno Batista Ribeiro<sup>1</sup>, Francisco Mickael de Medeiros Câmara<sup>2</sup>, Antônio Nazareno Guimarães Mendes<sup>3</sup>, Virgílio Anastácio da Silva<sup>4</sup>, Fernanda Faria Montanari<sup>5</sup>

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ABSTRACT: The production of quality coffees, with different sensory characteristics, is strongly related to drying techniques. Experiments were carried out on coffee fruits, with the presence and absence of the fruit turnover process during drying, using Catuaí Vermelho 144 coffee fruits, from the Cerrado Mineiro, processed dry and wet. The treatments consisted of natural coffee, natural green coffee, pulped coffee and semi-washed coffee fruits. They were carried out in a completely randomized design, with 4 post-harvest processes, 2 types of drying (with or without Stirring) and 3 replicates, totaling 24 plots. Natural green coffee and natural coffees were more responsive in the final scores, when not stirred during the drying process, unlike the coffees obtained by wet processing. Peeled coffees obtained the highest scores for the attributes, regardless of the adoption or not of stirring during the drying process. It was possible to obtain scores above 80 points by the SCAA protocol, without stirring the coffee.

**Index terms:** Sensory analysis, *Coffea arabica* L., drying, SCAA, quality.

#### ANALISE SENSORIAL DE CAFÉS SECADOS COM E SEM REVOLVIMENTO

RESUMO: A produção de cafés de qualidade, com características sensoriais diferenciadas, está fortemente relacionada com técnicas de secagem. Foram realizados experimentos de secagem de frutos de cafés, com a presença e ausência do processo de revolvimentos de frutos durante a secagem, utilizando frutos de café da cultivar 'Catuaí Vermelho 144', proveniente da região do Cerrado Mineiro, processados por via seca e via úmida. Os tratamentos contaram com frutos de cereja natural, verde natural, cereja descascado e cereja desmucilado os quais passaram por secagem conduzida em terreiro de concreto. Utilizou-se o delineamento inteiramente casualizado, com 4 processos pós-colheita, 2 tipos de secagem (com ou sem revolvimento) e 3 repetições, totalizando 24 parcelas. Os cafés, verde natural e cereja natural foram mais responsivos nas notas finais, quando não revolvidos durante o processo de secagem, diferentemente dos cafés obtidos pelo processamento via úmida. Os cafés descascados obtiveram as maiores notas para os atributos, independentemente da adoção ou não do revolvimento durante o processo de secagem. Foi possível obter notas acima de 80 pontos pelo protocolo SCAA, sem o procedimento de revolvimento do café.

Termos para indexação: Análise sensorial, Coffea arabica L., secagem, SCAA, qualidade.

#### 1 INTRODUCTION

Coffee farming is crucial to the economy of many countries as it generates millions of jobs in various parts of the world (HUGHES et al., 2014). The increasing demand for coffees with different qualitative characteristics has presented a scenario of innovations in technologies and production techniques for coffee cultivation.

Drying is the oldest technique for preserving food and agricultural products (MAISNAM et al., 2017), and it is the process wherein moisture is removed from the food material as a result of concurrent heat and mass transfer (MARQUES et al., 2008; SONTAKKE; SALVE, 2015). Before drying, coffee fruits can be processed by two methods: dry or dry (RATTAN et al., 2015). During the drying of fruits, the resulting decrease in water potential induces several metabolic responses (KLEINWÄCHTER; SELMAR, 2009) and consequently, changes in the composition of the substances present, besides coffee sensory quality (JOËT, et al., 2010).

According to Palacin et al. (2009), the encouragement for the development of new concepts to improve the unit operations of preprocessing and processing of fruits and grains is evident, in which hygiene and care to reduce, or even reduce mechanical and thermal damage, have become essential.

Thus, drying is a key stage in coffee post-harvest and should be started soon after harvest to rapidly reduce the high water content of the husk, pulp and mucilage and avoid fermentation, which may impair coffee quality; in addition, the operation is considered critical for providing thermal stress, development of fungi, addition of smoke odors, and other contaminants in fruits or grains (ABRAÃO et al., 2010; PALACIN et al., 2009; RESENDE et al., 2009). However, the best drying method is the one that meets the characteristics of each region, producer and desired quality standard, aiming at profitability and consumer satisfaction (SAATH, 2010).

<sup>1.2.3.4.5</sup> Universidade Federal de Lavras/UFLA - Departamento de Agricultura/DAG - Cx. P. 3037 - 37.200-000 - Lavras - MG-brunoberibeiro@hotmail.com, mickaelmedeiros@hotmail.com, naza@dag.ufla.br, virgílio@dag.ufla.br, fernanda\_montanari@hotmail.com

Sensory analysis has the ability to give a scientific evaluation and a quantitative measure to the appearance, flavor and texture of a food product, and it is essential to define performance standards and evaluate the progress of the implemented process (IANNARIO et al., 2012).

The stirring rate in coffee drying process is not treated in literature. There is a lack of knowledge about its benefits or damage in coffee sensory characteristics. It is important to emphasize that the absence of stirring reduces costs, once you diminish labor to do this activity.

In this context, the objective in this research was to identify the sensory attributes by the protocol of the Specialty Coffee Association of America (SCAA) for coffees obtained by the wet and dry process, dried with and without stirring.

#### 2 MATERIAL AND METHODS

The experiment was carried out in Minas Gerais in 2016, in the region of the Cerrado Mineiro, municipality of Coromandel, in a private coffee property. 'Catuaí Vermelho 144' coffee fruits were processed dry and wet. Treatments, as well as the control of natural coffee fruits, natural green coffees, pulped coffee, semi-washed coffee underwent drying process.

Initially it was delimited in the border space in the field, respecting three rows started after the "carrier way" and the three plants in the row to start harvest. The plants were demolished by an automotive harvester until a total of 1500 liters of coffee fruits, and a selection was made, removing remaining green fruits, which composed the natural green plot, besides hydraulic separation of drier fruits in a washer. Of the remaining coffee from the washer, part passed by the parchment peeler and part by the peeler and by the demister. It is worth mentioning that the tests were performed on days of full sun.

The experiment was carried out in a completely randomized experimental design (DIC) with 4 processes (natural coffee, natural green coffee, peeled parchment, semi-washed coffee, 2 types of drying (with and without stirring) and 3 replicates, totaling 24 plots with 30 liters each.

The drying procedures for each treatment are described below:

#### Coffee beans dried with stirring process Natural coffee and Natural Green Coffee

The coffees were scattered in a layer 3 cm high, stirred 8 times a day and, after being "half-

dry", they were surrounded and covered with cloths + plastic canvas. The next day, the cloths along with the tarpaulins were removed after the haze disappeared, and the coffee was again spread and stirred 8 times, until the samples reached 11.5% moisture. The drying time of the samples in cement yard in the region and conditions under study was between 16 and 18 days.

## Pulped coffee and Semi-washed Coffee

The coffees were scattered in a layer 3 cm high, tilted 12 times a day and, after being "half-dry", they were layered and covered with cloths + plastic canvas. The next day, the cloths along with the tarpaulins were removed after the haze disappeared, and the coffee was again spread and stirred 12 times, until the samples reached 11.5% moisture. The drying time of the samples in cement yard in the region and conditions under study was between 9 and 12 days.

## Coffee beans dried without stirring process

### Natural coffee

In the first days of drying in a cement yard, the coffees were scattered in thin layers (fruit to fruit) allowing the rapid removal of surface water and dehydration of the remaining mucilage. during 4 days. After the "browning of the bark", the drying layer was passed to 50% of the yard space. After drying the remaining water of the bark (as the fruits stop melting in the hand), the drying layer went to 25% of the vard space and was kept for 1 day in full sun. Subsequently, a sombrite screen with 50% brightness was placed on the coffee samples, and cloths were placed together with the plastic canvas on the sombrite, remaining all day long. By the next day, the cloths along with the tarpaulins were removed after the haze had disappeared, and the sombrite remained. This procedure was repeated until the samples reached 11.5% moisture. The drying time of the samples in cement yard, in the region and conditions under study was between 15 and 17 days.

## **Natural Green Coffee**

In the first 2 days of drying of the green coffees the fruits were scattered in the cement yard in thin layers "fruit to fruit", allowing the appearance of dark spots on the bark. After these darkening points, the drying layer was passed to 50% of the yard space and maintained, for 2 days in full sun. After the 2 days, the layer went to 25% of the space of the yard and was maintained,

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for 2 days in full sun. Subsequently, a sombrite screen with 50% brightness was placed on the coffee samples, and cloths were placed together with the plastic canvas on the sombrite, remaining all day long. By the next day, the cloths along with the tarps were removed after the fog had disappeared, but the sombrite was kept. This procedure was repeated until the samples reached 11.5% moisture. The drying time of the samples in cement yard in the region and conditions under study was between 17 and 20 days.

#### Pulped coffee

In the early days of drying, the coffees were scattered in thin layers, "seed to seed" during drying in cement yard, allowing the rapid removal of surface water and dehydration of the remaining mucilage, during 2 days. After drying the mucilage, the drying layer was passed to 50% of the yard space and maintained for 2 days in full sun. After the 2 days, the layer went to 25% of the vard space, for 1 day in full sun. Subsequently, a sombrite screen with 50% brightness was placed on the coffee samples, and cloths were placed together with the plastic canvas on the sombrite, remaining all day long. By the next day, the cloths along with the tarps were removed after the fog had disappeared, and the sombrite was kept. This procedure was followed until the samples reached 11.5% moisture. The drying time of the samples in cement yard in the region and conditions under study was between 9 and 10 days.

#### **Semi-washed Coffee**

On the first day, the coffees were scattered in thin layers, "seed to seed" during drying in cement yard, allowing the rapid removal of surface water and dehydration of the remaining mucilage. After removal of the surface water and dewatering of the mucilage, during 1 day, the drying layer was passed to 50% of the yard space and kept for 1 day in full sun. After 1 day, the layer went to 25% of the yard space, for 1 day in full sunlight. Subsequently, a sombrite screen with 50% brightness was placed on the coffee samples in the hottest hours of the day, remaining all day long. By the next day, the cloths along with the tarpaulins were removed after the haze had disappeared, and the sombrite remained. This procedure was repeated until the samples reached 11.5% moisture. The drying time of the samples in cement yard in the region and conditions under study was between 9 and 10 days.

In the drying treatments of coffees without stirring, sombrites with graduation of 50% were used in order to minimize the direct incidence of the sunlight, which would damage the appearance of the grains, causing stains. This sombrite was added on the coffee, at different times according to the processes adopted.

#### Sensory analysis protocol

The sensory analyses were performed by two qualified and accredited panelists for the evaluation of special coffees (Q-Graders), using the methodology proposed by the Special Coffees Association of America - SCAA (LINGLE, 2011).

100 grams of each sample were roasted in a Probat TP2-Leogap roaster, in a 24 hour interval, before sensory analysis, with a coloring profile between #58 for roasted grain and #63 for roasted and ground grain, as verified by the equipment Mbasic Agtron. The roasting time was monitored, respecting the interval of 8 to 12 minutes, with the average roasting time between samples of 9 minutes and 15 seconds. Milling was standardized in a Guatemala - Mahlkönig mill, with an average texture of 70% of the powder passed through a 20 mesh sieve. The mineral water, used for infusion, was boiled at a temperature of 93°C  $\pm$  1. For each sample, five cups of 5.5% m/v (roasted and ground coffee/water) were prepared for the sensory analysis procedure. All testers underwent calibrations prior to sensory analysis. The environment and location of the tastings were prepared for noise reduction, odor elimination and careful standardization for the evaluation of the treatments with quality levels with intervals of 0.25 points.

The following sensory attributes were evaluated: Aroma; Flavor; Finalization; Body; Acidity; Balance (acidity and body); Uniformity between Cups; Absence of defects; Sweetness and Global Impression. The sum of the scores obtained in the attributes resulted in the final scores.

## Histochemical analysis

The bean samples were submerged in distilled water for four days and kept at room temperature. The sections for histochemical reactions, obtained by hand-free cutting using a stainless-steel blade, were treated for 3 minutes with the reagent Sudan IV in 80% ethanolic solution for the visualization of lipids, (JENSEN, 1962) with modifications (GOULART et al., 2007). The plank assembly was performed in the Corel Draw software.

#### **Experimental Analysis**

Statistical analyses were performed using the Sisvar software (FERREIRA, 2011), using the Tukey test at 5% probability, the PCA analysis were performed using the PAST3 software (HAMMER et al., 2001).

#### **3 RESULTS AND DISCUSSION**

Through analysis of variance it was verified that there was an interaction between processes and treatments used for the analyzed variables, except for uniformity and clean cup. There was an isolated effect of process type only for sweetness.

For aroma and flavor, it was verified that the best treatments were for pulped coffee, with averages of 7.58 and 7.54 (with stirring) and 7.75 and 7.42 (without stirring), respectively. It is noteworthy that there was no difference between the two processes (with or without stirring) for pulped coffee. Semi-washed coffee had lower mean values when compared to the pulped coffee type, but only differed significantly in aroma, when the process without stirring was adopted (Table 1). For natural coffee and natural green coffee processing, the absence of stirring resulted in higher averages when compared to the stirring process.

These results might be related with a thinner layer of coffee beans arranged for drying, since drying without stirring needs a lesser mass of coffee to be dried by sunlight temperature and air, promoting an evenly moisture removal. Once the drying procedure goes on, it is necessary fold the layers and using the sombrite, to avoid excessive moisture removal and luminosity, preventing stains on the coffee beans.

This procedure initially has a high moisture removal rate, however, with the folds of layers and sun protection (sombrite), the rate decreases, preserving the lipid fraction. Lipids in coffee serve as carriers for flavors and for fat-soluble vitamins and contribute to texture and mouthfeel (RIBEIRO et al., 2009)

According to Manzocco and Lagazio (2009), any changes in composition of lipids may contribute to a loss of sensory quality of coffee beverages. Lipids might be hydrolyzed enzymatically or chemically and the rate at which these reactions occur depends on environmental and technological aspects, as well as availability of oxygen, moisture, temperature, and packaging material.

Aroma and flavor arise from quality, intensity and complexity of all the attributes found in the coffee beverage, consequently, it strongly influences the panelist's note, and it is one of the most important requirements for beverage quality (KNYSAK, 2017).

According to Tables 2 and 3, it is verified that pulped coffee had the highest mean values for the variables finalization, acidity, body and balance, with values of 7.33, 7.54, 7.42 and 7.42 (with stirring) and 7.29, 7.33, 7.42 and 7.33 (without stirring), respectively. Although once again the semi-washed coffee had smaller averages than pulped coffee, they only differ statistically in the variables body, with stirring, and in balance, both with and without stirring. For both natural coffees, the absence of stirring resulted in higher averages when compared to the process with stirring.

Body is recognized to be one of the most important sensory characteristics determining the quality of coffee beverages, high scores correspond to the richness of aroma and increased viscosity and density of beverages (NAVARINI et al., 2004), it is often related to the total solids and occasionally also linked to fat or fatty acid content (GLOESS et al., 2013) and results to body may be correlated with better aroma and flavor results, once pulped and semi-washed coffees had the higher averages.

Acidity contributes to the liveliness of a coffee, with the perception of sweetness (SCAA, 2008). Furthermore, for sustained sensation at the end of a sip, the finishing should be well balanced between the aroma, acidity, bitterness and astringency (GLOESS et al., 2013). Overall, semi-washed and pulped coffees obtained great results for these variables.

It is important to note that natural coffee without stirring had higher means compared to stirring, this could be associated to drying kinetics, where thick layers where the coffee beans are disposed in drying with stirring, reduces water removal and homogeneity. The increase in temperature reduces the viscosity of the water, directly influencing the resistance of the fluid to the flow; the decrease of the viscosity facilitates the diffusion of the water molecules in the capillaries of the product (CORRÊA et al., 2010). In other words, thick layers and holding the peel and pulp in the coffee beans, gives conditions to fermentation and/or enhance in loss of quality.

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**TABLE 1** - Averages of aroma and flavor of coffee drinks according to treatments and drying processes\*.

	DRYING PROCESSES					
TREATMENTS	WITH STIRRING	WITHOUT STIRRING	WITH STIRRING	WITHOUT STIRRING		
	AF	ROMA	FLAVOR			
NAT. GREEN COFFEE	6.04 Cb	6.45 Ca	6.04 Cb	6.50 Ca		
NAT. COFFEE	7.13 Bb	7.70 Aa	7.04 Bb	7.20 Ba		
PULPED COFFEE	7.58 Aa	7.75 Aa	7.54 Aa	7.42 Aa		
SEMI-WASHED COFFEE	7.42 Aa	7.38 Ba	7.29 Aa	7.50 Aa		

<sup>\*</sup> Averages followed by the same letter, upper case in the column and lowercase in the row, do not differ by Tukey's test (p < 0.05).

**TABLE 2** - Averages of finishing and acidity of coffee drinks according to treatments and drying processes\*.

	DRYING PROCESSES				
TREATMENTS	WITH STIRRING	WITHOUT STIRRING	WITH STIRRING	WITHOUT STIRRING	
	F	INISHING	ACIDITY		
NAT. GREEN COFFEE	6.00 Cb	6.25 Ca	6.04 Cb	6.37 Ca	
NATURAL COFFEE	6.91 Bb	7.29 Aa	7.04 Bb	7.45 Aa	
PULPED COFFEE	7.33 Aa	7.29 Aa	7.54 Aa	7.33 Aa	
SEMI-WASHED COFFEE	7.16 Aa	7.12 Aa	7.29 Aa	7.25 Aa	

<sup>\*</sup> Averages followed by the same letter, upper case in the column and lowercase in the row, do not differ by Tukey's test (p < 0.05).

**TABLE 3** - Averages of body and balance of coffee beverages according to treatments and drying processes\*.

	DRYING PROCESSES					
TREATMENTS	WITH STIRRING	WITHOUT STIRRING	WITH STIRRING	WITHOUT STIRRING		
		BODY	BA	LANCE		
NAT. GREEN COFFEE	6.00 Cb	6.29 Ca	6.00 Db	6.25 Ca		
NATURAL COFFEE	6.92 Bb	7.50 Aa	6.91 Cb	7.45 Aa		
PULPED COFFEE	7.42 Aa	7.42 Aa	7.42 Aa	7.33 Ab		
SEMI-WASHED COFFEE	7.13 Ba	7.25 Aa	7.16 Ba	7.12 Ba		

<sup>\*</sup> Averages followed by the same letter, upper case in the column and lowercase in the row, do not differ by Tukey's test (p < 0.05).

Guimarães et al. (2002) reported that during drying, some sugars may accumulate, and it is one of the possible defense mechanisms against stress caused by desiccation. The authors point out that these sugars can be enhanced or reduced according to the seed drying rate and/or environmental conditions. Sugars are precursors of sweetness and contribute to the body of the coffee beverage.

Except for general score with stirring not differing from semi-washed coffee, general and final scores of the beverages with pulped coffee were higher to the other treatments, with averages of 7.50 and 82.29 (with stirring) and 7.46 and 82 (without stirring), respectively (Table 4). For natural coffee, the average of the final scores was higher, due to the absence of stirring in the drying process, when compared to the adoption of stirring 8 times a day.

These results reveal no necessities of stirring coffee, which can assist producers in making decisions in post-harvest processes, reviewing the need for the requirement to constantly stir up lots of coffees.

It should be emphasized that the quality of specialty coffees is related to the intrinsic characteristics of the beans, showing chemical compounds which, after roasting, will provide aroma, flavor, acidity, sweetness and bitterness to the beverage, in addition to the synthesis, accumulation and degradation of chemical compounds of the raw coffee bean, considered flavor precursors of the beverage (TAVEIRA et al., 2014).

Researches with specialty coffees is still scarce due to the small production of quality coffee and the lack of researchers working in this specific field. However, some studies have been carried out to verify the influence of pre and post-harvest management on the quality of specialty coffees (ALVES et al., 2013; KREUML et al., 2013). Among the post-harvest stages, drying has been one of the main targets of studies (ALVES et al., 2017; SIQUEIRA et al., 2016), since it is known that this technique has a great influence on the final quality of agricultural products.

For sweetness stirring effect was not verified, there was statistical difference only among treatments. Semi-washed coffee, pulped coffee and natural coffee had better grade means 9.92, 9.88 and 9.87, respectively, where green coffee was inferior to the other treatments.

Figure 1 shows the two principal components, accounting for 99.48% of data variability and the principal component 1 holds the major variance, (PC1=98.964% and PC2=0.516%), it may perfectly be used to represent the set of the variables measured for the tested treatments, since it incorporates over 80% of the variance.

In this type of data analysis, it is possible to group the treatments that reveal similarity among variables. To obtain coffee beverage with better aroma, flavor, body and balance the best treatments are pulped coffee with or without stirring and natural coffee with stirring. Great acidity and finishing can be reached in the beverages when semi-washed coffees are treated or not with stirring and in natural coffee stirring is made. Natural green coffee had the worst results to all variables, independent of the usage of stirring.

These results lead to a new possibility of making different beverages, depending on market trends and costumer's acceptability. Overall, it is possible to obtain high-quality coffee beverages without the stirring process, thus decreasing the manpower required for this operation.

It is worth to emphasize that semi-washed and pulped coffees were better than natural green and natural coffees. Green coffee was expected to present these lower results, once the fruits did not reach the physiological maturity. For natural coffee, the drying process may be linked to the lower results. According to Santos et al. (2009), natural coffee beans are exposed to higher initial water contents, which may favor the occurrence of undesirable fermentation, depreciating the product.

Wet processing could bring more benefits to beverage quality, Joët et al. (2010), noted that significant metabolism occurs during wet processing which could lead to increase of chemical substances such lipids. In this sense, an increase in lipid content plus effects from no using stirring procedure may lead to a better coffee beans quality.

TABLE 4 - Averages of the general and final scores of coffee beverages according to treatments and drying processes\*.

	DRYING PROCESSES					
TREATMENTS	WITH STIRRING	WITHOUT STIRRING	WITH STIRRING	WITHOUT STIRRING		
	GENER	AL SCORE	FINAL SCORE			
NAT. GREEN COFFEE	6.13 Ca	6.38 Ca	62.25 Db	64.50 Ca		
NATURAL COFFEE	6.96 Cb	7.54 Aa	78.87 Cb	82.45 Aa		
PULPED COFFEE	7.50 Aa	7.46 Aa	82.29 Aa	82.00 Aa		
SEMI-WASHED COFFEE	7.25 Aa	7.13 Ba	80.75 Ba	80.45 Ba		

<sup>\*</sup>Averages followed by the same letter, upper case in the column and lowercase in the row, do not differ by Tukey's test (p < 0.05).

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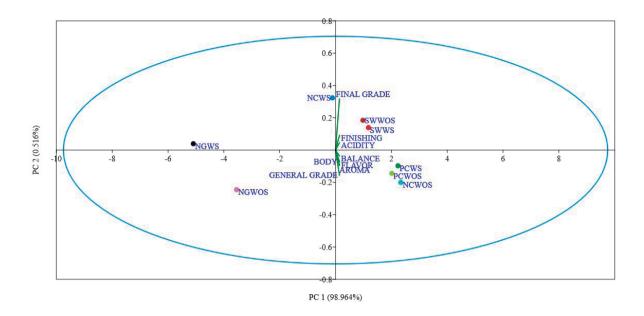


FIGURE 1 - Relationship between the two principal components of the treatments and drying processes variables. NGWS= natural green coffee with stirring, NGWS= natural green coffee without stirring, NCWS= natural coffee with stirring, NCWOS= natural coffee without stirring, PCWS= pulped coffee with stirring, PCWOS= pulped coffee without stirring, SWWS= semi-washed coffee with stirring.

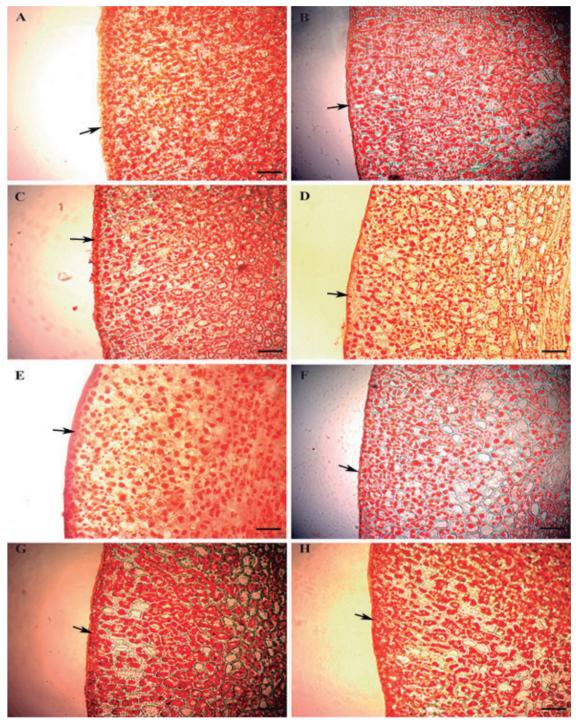
Figure 2 presents the histochemical sections made in the coffee beans submitted to the different post-harvest processes, dried in the presence and absence of stirring. Coffees without the stirring procedure during drying had visually higher lipid concentrations in the peripheral regions of the beans as can be verified by arrows. Probably drying coffee without stirring promotes a better lipid content integrity, which implies better aroma and flavor on beverage.

The agronomical management made during the fruit production must preserve the integrity of the cell wall of the coffee beans and, consequently, collaborates with lipid organization (GOULART et al., 2007).

According to Goulart et al. (2007), better quality coffee shows a higher lipid concentration in the peripheral region of the beans, with well defined globular lipid bodies within the

protoplasts. With the loss of beverage quality, the lipids are homogeneously distributed in the coffee beans tissues. The preservation of the integrity of these structures is important, since the oils besides the retainers of aromatic components act together with other compounds in the formation and perception of the beverage body (SCAA, 2008).

It is worth mentioning that, given the influence of the drying process in the post-harvest phase of coffee, and considering that the price paid for this product is directly proportional to its quality, this step is of great importance and must be performed in a technical way and to maintain the desirable characteristics of the product. In the case of special coffees drying, special attention must to be given, since the value paid for this product is well above that paid by the commodity coffee, thus, small flaws can lead to great losses.



 $BAR = 100 \mu m$ 

**FIGURE 2** - Sections of coffee beans: A) Natural coffee without stirring; B) Natural coffee with stirring; C) Pulped coffee without stirring; D) Pulped coffee with stirring; E) Semi-washed coffee without stirring; F) Semi-washed coffee with stirring; G) Natural green coffee without stirring; H) Natural green coffee with stirring

#### **4 CONCLUSIONS**

Semi-washed and pulped coffees obtained higher scores between attributes and final scores. Coffees dried without stirring had scores above 80 points by the SCAA methodology and no differences compared to those stirred. Production of coffee without stirring can be adopted without loss in quality reducing cost in post-harvest coffee management.

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# PERFORMANCE OF DIFFERENT FERMENTATION METHODS AND THE EFFECT ON COFFEE QUALITY (Coffee arabica L.)

Aida Esther Peñuela-Martínez<sup>1</sup>, Arley David Zapata-Zapata<sup>2</sup>, Diego Luis Durango-Restrepo<sup>3</sup>

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ABSTRACT: Fermentation of coffee mucilage is a spontaneous process caused by microorganisms growing in the environment, which is influenced by factors such as the coffee variety, climate and fruit maturity. These external factors play an important role in fermentation evolution because they have effect on the microorganism activity and the time for substrate transformation. The aim of this research was to asses different fermentation wet process and evaluate their effect on coffee quality (*C. arabica*), as well as on organic acid concentrations and volatile organic compounds content, in the green coffee beans. The study was divided in two phases, one in which the pulping time was delayed and the fermentation methods were modified, and the second phase in which a bioreactor was used to control the pH and temperature of the coffee mass during fermentation. Two control treatments were used: without fermentation (mechanical removal of mucilage) and the traditional fermentation done in the farm. Significant differences in coffee quality were observed. The best quality was obtained from the treatments that used short process times and low temperatures. The concentrations of acetic, lactic and citric acids between the treatments and the control treatments were different. Higher contents of esters and ketones were found in the coffee that obtained the highest quality. The assessed processes lead to the conclusions that it is possible to improve coffee quality throughout introducing changes in the fermentation process, as well as modulating the acidity and fragrance of the final product.

**Index terms:** Fermentation processes, special attributes, organic acids, volatile organic compounds.

# DESEMPENHO DE DIFERENTES MÉTODOS DE FERMENTAÇÃO E O EFEITO NA QUALIDADE DO CAFÉ (Coffea arabica L.)

RESUMO: A fermentação da mucilagem de café é um processo espontâneo causado por microrganismos que crescem no meio ambiente, influenciado por fatores tais como a variedade, o clima e a maturação dos frutos. Estes fatores externos desempenham um papel importante na evolução da fermentação porque determinam atividade dos microrganismos e o tempo da transformação do substrato. O objetivo do presente trabalho foi avaliar o efeito de diferentes modificações no processo de fermentação e avaliar o seu efeito na qualidade do café (*C. arabica*), assim como, nas concentrações de ácidos orgânicos e no teor de compostos orgânicos voláteis, no café verde. O estudo foi dividido em duas fases, uma em que o tempo de despolpamento foi atrasado e os métodos de fermentação modificados, e a segunda fase em que foi utilizado um biorreator para controlar o pH e a temperatura da massa de café durante a fermentação. Utilizaram-se dois tratamentos de controle: sem fermentação (remoção mecânica de mucilagem) e a fermentação tradicional feito na exploração agrícola. Foram observadas diferenças significativas na qualidade do café. A melhor qualidade foi obtida a partir dos tratamentos que utilizaram os tempos de processo curtos e baixas temperaturas. A concentração de ácidos acético, láctico e cítrico entre os tratamentos que usaram a fermentação e os controles foi diferente. Maiores teores de ésteres e cetonas foram encontrados no café que obteve maior qualidade. Os processos avaliados permitiram concluir que é possível melhorar a qualidade do café através da introdução de alterações no processo de fermentação, bem como modular a acidez e a fragrância do produto final.

Termos para indexação: Processos de fermentação de café, qualidade do café, ácidos orgânicos, compostos orgânicos voláteis.

#### 1 INTRODUCTION

Growing specialty coffee market is a response of customers desiring to pay more for unique attributes in the beverage. In that regard, the fermentation stage plays one of the most important roles due the wide variety of modifications that have effect on the flavors and tastes (FOLMER 2014, LEE et al., 2015; POLTRONIERI; ROSSI, 2016). Coffee quality is an outcome not only of the intrinsic grain characteristics but for a combination of the environmental characteristics of the place it is grown (altitude, climate, and soils, among others), the ripeness at which it is harvested, and the post-harvest process carried out (JOËT et al.,

2010; TOLEDO et al., 2016). The wet method, which is a post-harvest process used to maintain the mildness of the Arabica coffee, involves the mechanical removal of the fruit skin (exocarp or pulp) to expose the jelly layer (mesocarp or mucilage) adhered to the bean's surface. Then pulped coffee is put into tanks to perform mucilage degradation by spontaneous fermentation, to facilitate mucilage removal by washing. In the mucilage degradation process, microorganisms in the environment use mucilage as a substrate for their metabolic processes to generate organic acids among others. The wet process of coffee is associated with a higher acidity, which leads to a

<sup>1.2.3</sup> Universidad Nacional de Colombia - Sede Medellín - Escuela de Química - Carrera 65 # 59A-110 - Medellín - Código Postal 050034 - Colombia - aepenuelam@unal.edu.co, adzapata@unal.edu.co, dldurango@unal.edu.co

better cup qualification. This process is widely used in different coffee-growing countries to enhance coffee quality, because it plays a significant role in modulating the quality since the variation of each factor contributes to the composition of coffee, the formation of the aroma and the flavor precursor compounds that can produce different sensory experiences (JOËT et al., 2010; SUNARHARUM; WILLIAMS; SMYTH, 2014). Since acidity and fragrance are two important attributes of the sensory analysis, correlating the fermentation practices with the organic acids and volatile organic compounds (VOCs) in green coffee beans is crucial because these compounds are precursors of other compounds formed through roasting, which are important for the development of the taste and aroma of coffee (TOLEDO et al., 2016).

Studies on coffee fermentation have been performed with the aim of improving the wet process. For example, some research works have reduced the fermentation time (SILVA et al., 2013; PEÑUELA et al., 2010; TAI et al., 2014), inoculated microorganisms to improve quality (LIN, 2016; PEREIRA et al., 2015, PEREIRA et al., 2016; VELMOUROUGANE et al., 2008), or controlled the process to avoid deterioration of the product quality (JACKELS; JACKELS, 2005; PEÑUELA et al., 2012). However, the effect of different coffee fermentation methodologies on both quality and chemical composition is scarce. Coffee growers sometimes use traditional practices to conduct fermentation, which affect the development of the process and causes differences coffee quality. These practices include maintaining the coffee fruits for longer times before pulping, adding up freshly pulped coffee to coffee with a fermentation process in progress and adding water during processing. Temperature and pH are critical variables in the evolution of fermentation. When spontaneous fermentation occurs, the mass temperature of coffee increases between 4 to 8 °C, and pH decreases down to values between 5.5 to 4.0, or even lower (AVALLONE et al., 2001; CORREA et al., 2014; CORDOBA-CASTRO et al., 2016; JACKELS; JACKELS, 2005; VELMOUROUGANE, 2013). However, the effect of controlling these parameters during fermentation on the coffee quality is unknown.

The coffee quality could be improved changing the fermentation stage, which would produce better prices in the market. The aim of the present paper was to determine the effect of some common practices carried out by coffee growers in both the beverage quality and the chemical composition of the beans. Furthermore, the effect of maintaining the pH and temperature constant during the fermentation process using a bioreactor was evaluated.

#### 2 MATERIALS AND METHODS

There were used coffee fruits of C. arabica. (Caturra variety) from a plantation located at an altitude of 1.940 m in Ciudad Bolívar, Antioquia-Colombia. A vertical cylinder pulper was used to remove the coffee fruit outermost skin, with no use of water for the process itself, as well as for the transportation of the pulp and pulped beans. An after-pulping size classification was performed in a cylindrical screen. The research work was divided into two phases. The first phase consisted in assessing the different fermentation strategies, to obtain a higher coffee quality and choose the best strategy to be used in the second phase. In the second phase, a bioreactor was used to control pH and temperature within the mass of coffee during fermentation. The pH and temperature of coffee mass were monitored during the fermentations.

In order to run the first phase and determine the fermentation strategy that results in better coffee quality, two factors were evaluated; pulping delaying time and the fermentation method. The two delaying times were t1 (6h±1h) and t2 (16h± 1h), meaning the pulping at the same day and pulping a day after harvesting, respectively. The fermentation methods were: fermentation for 24 h (F24), fermentation for 48 h with an addition of freshly pulped coffee at hour 24 (MF), and fermentation for 72h with addition of freshly pulped coffee at hour 24 and addition of water at hour 48 (WF). These fermentations were evaluated to allow changes in the microbial activity conditions. The experimental unit randomly assigned to each treatment was 60 kg of pulped coffee. Two control treatments were used: a negative control (without Fermentation), which is the mechanical removal of mucilage of pulped coffee, and a standard control (traditional fermentation), which consists in performing a spontaneous fermentation during 16 h. Once the treatment with the best results was selected, the first phase was concluded.

To conduct the second phase in the bioreactor, it was selected the treatment of the first phase that generated the highest final score in the sensory analysis. The stainless-steel bioreactor was a 35 L stirred fermenter tank with a water jacket to heat-up or cool-down the coffee mass, according to the desired temperature (Designed by Centricol®, Colombia). With the characteristics and physical properties of the coffee mass under a fermentation process, it was selected a helical belt propeller because it causes low shear stresses to viscous fluids with suspended particles.

The agitation was performed by one minute every two hours. A randomized experimental design, with two temperature levels (17 °C and 23 °C) and three pH levels (without control, 5,0 and 4,0) was followed. The experimental unit consisted of 30 kg of pulped coffee. One standard control, fermentation for 24 h was adopted. During the first phase, the environment temperature varied between 17,0 and 22 °C, while it varied between 15,2 and 28,6 °C during the second phase.

For the sensory analysis, the coffee samples from the fermentation treatments were washed to remove the degraded mucilage. Then, they were dried with forced air at 50 °C down to a moisture of 11 %  $\pm$ 1,0% w.b. Finally, an expert panel with five trained coffee cuppers with O-Grader certification evaluated the samples. The coffee cuppers considered all the important flavor attributes of coffee using the Specialty Coffee Association of America protocol for roasting, preparation and analysis (SCA, 2015). Each of the five cuppers evaluated five cups of each coffee sample. The attributes for the sensory profile were fragrance/ aroma, uniformity, cleanliness, sweetness, flavor, acidity, body, aftertaste, balance and overall impression. Each attribute was evaluated in the 10-point scale and the final qualification is given in a 100-point scale. The response variable was the final score, which was obtained by adding the individual scores given to each attribute to represent the overall coffee quality. The highest final score indicates the best classification of coffee quality. The median of the attributes and the final score was chosen as a robust measure of central tendency for this type of analysis.

In order to perform the chemical analysis, washed coffee samples of 50 g were taken and then stored at  $-20\,^{\circ}$ C, until analysis. To keep the coffee composition unaltered, the samples of coffee beans were washed and frozen at -80 °C overnight and then lyophilized for 48 h until moisture went down to a range between 9% and 13% w.b. Subsequently, the samples were subjected to cryogenic grinding (<100  $\mu$ m). Ground samples were kept frozen at -20 °C in plastic containers until the analysis. The moisture content of the ground coffee was estimated after drying 0,2 g completely in an oven at 105 °C for 24 h (International Organization for Standardization ISO:6673, 2003).

For the organic acid extraction, the procedure described by Evangelista et al., (2014) was used, including the following modifications:

0,5 g of the sample was combined with 5,0 mL of Milli-O water. Then, the mixture was set to vortex for 2 min and allowed to reach steady state for 10 min before undergoing a double centrifugation at 10000g, at 4 °C during 10 min. To clarify the samples, the Carrez reagent (RODRIGUES; BRAGAGNOLO, 2013) was added and the samples were centrifuged again for 5 min at 1585g. The supernatant was micro-filtered through a 0,45 µm nylon membrane and stored at -20 °C until analysis. The samples were prepared in triplicates. The contents of the malic, citric, lactic, acetic, succinic and quinic acids were determined by HPLC using a Shimadzu Prominence 20 A chromatograph, which was equipped with a diode array detector, using a column Synergi 4U Hydro-RP 80 A (250 mm  $\times$  4,6 mm) operated at 25 °C-29 °C. Acids were eluted at a flow rate of 0,7 mL/ min, with the mobile phase buffer phosphate at 20 mM and pH level of 2,90 for 45 min. The injection volume was 20 µL. The organic acids were monitored at a wavelength of 205 nm. The identification was performed by co-elution with analytical standards of each acid (purity > 99 %, Sigma-Aldrich, Pennsylvania, USA). Quantification was conducted using calibration curves (peak area vs. compound concentration) for concentrations of 5-300 mg/L. All concentration curves presented with high linearity (correlation coefficient greater than 0.99).

In order to determine differences in volatile organic compounds, the samples corresponding to the control treatments and the highest and lowest final score in sensory analysis were chosen. For extraction of volatile compounds, 0.5 g of ground coffee were placed in Solid Phase Microextraction (SPME) vials. The samples were placed for 30 min in an oven at 35 °C to reach sample headspace equilibrium. The compounds were trapped by means of Carboxen/poly(dimethylsiloxane) (CAR/PDMS) type 75 µm SPME fibre (Supelco Co.). Desorption process was done in the GC injection port for 3 min at 280 °C in splitless mode. The separation process for the chromatography analysis was performed with a DB-WAX capillary column measuring 50 m x 320 µm in an Agilent HP 6890 GC chromatograph, which coupled to Agilent HP 5973 Mass Spectrometer where detection was done. The carrier gas was helium with a flow was 1,2 mL/min. The column temperature was programmed from 40 to 110 °C at 7°C/min and then at 10°C/min until 280 °C. The mass were scanned from 40 to 550 amu at a scanning rate of 2,89 scans/s. Ionization method used was electronic impact with an ionization energy of 70 eV. The compound identification was performed in accordance with the mass spectrum comparison with the databases (Wiley 275 Mass Spectral Data and NIST 98 Mass spectral Library). The probability of this relationship in all cases was greater than 80%. Furthermore, the concentration was related to the area under the curve of each chromatographic peak.

Data analysis was conducted using the Statistical Analysis System program (Cary, NC, USA) for ANOVA ( $\alpha$ =0,05). The Tukey and Dunnett tests, with a 5 % significance level were applied to compare the treatments and controls, which were evaluated separately in order to obtain the treatment with the best quality. Multidimensional scaling (MDS) analyses were applied to the means of the organic acids and volatile organic compounds using the R program (version 3.3.1.).

#### **3 RESULTS AND DISCUSSION**

According to the sensory analysis (Table 1), the final score obtained in all treatments was higher than 80 points; the coffee obtained in this study was classified as Premium according to the SCA scale. The control treatments assessed in this research showed that quality was associated with the intrinsic characteristics of the Arabica species, wet process and growth conditions, such as soil and climate (JOËT et al., 2010). ANOVA showed that the interaction of factors, such as pulping delaying time and fermentation methods, affected the final coffee quality (p=0,0383). The treatments applied indicated a higher quality than the standard and negative controls.

In general, the temperature of the mass of coffee in the fermentation processes was between 18 and 22 °C and the pH values decreased from 5,89 to 3,77. The lowest final score in quality was obtained by the treatment with the longer time (t2-WF); this fermentation had a final pH value below 4,00. Other treatments with high qualifications showed final pH values above 4,10. According to Jackels and Jackels (2005), the final coffee quality is better when the pH value is higher than 4,00 at the end of the fermentation.

Uniformity, cleanliness and sweetness obtained the maximum score (10 points) in all samples (*Data not shown*). Therefore, neither astringency or 'green' flavors nor negative impressions were found. Moreover, this indicate

the consistency of flavor in the five cups of each several coffee sample considered by the coffee cuppers.

Differences in the other attributes scores were detected between the samples with high final scores and the standard and negative controls, mainly in acidity, fragrance, body and flavor (Table 1). Treatments with t2 tend to obtain the highest score in body and fragrance attributes. The main organoleptic descriptors were fruity, with citric and red berries notes, although the cuppers were not consistent. This indicates the variations of the perceptions of the panelists, which was also noticed by Pereira et al., (2017). Acidity and fragrance can be related with microbial activity because of organic acids and aromatic compounds production. The processes occurring naturally within the fruit affect the coffee beverage. Therefore, the longer time between harvest and pulping process could be influencing these results. For example, the differences in characteristics. such as sweetness and body, have been identified to depend on the type of process. When the fruit remains whole in the drying process, the coffee beverage has a high sugar content and full body because of the metabolism of the beans (KNOPP et al., 2006). Moreover, the different pulping delaying times have been assessed and found to have no negative effects on coffee quality, in the specific varieties and environmental conditions (CAIXETA et al., 2013).

The treatment t2-F24 presented significant differences in quality with the control treatments and was classified as Excellent specialty coffee according to SCA, as it obtained a higher final score of 86 points. Therefore, this treatment was selected to perform the fermentation in the bioreactor with pH and temperature control.

Fermentations with pH and temperature control was conducted for 24 h on coffee pulped the day after harvest (16h±1h) (Table 2). The ANOVA showed a significant effect of the interactions of pH and temperature on coffee quality (p=0.0347). The final score tends to be higher when the fermentation was conducted at low temperature. However, neither pH control nor temperature control produce significant effect on coffee quality separately (p=0.2983 and p=0.1587 respectively). The fermentation processes in the bioreactor showed that the final score was significantly high in the coffee obtained at 17 °C and pH 5,00, having the highest qualifications in acidity, fragrance and body.

Time before pulping	Fermentation method	Final score	Acidity	Fragrance/ aroma	Body	Flavor
	F24	84,25±0,71	7,75±0,21	7,88±0,30	7,75±0,35	7,81±0,41
t1	MF	84,94±2,21*,**	$7,94\pm0,34$	$7,88\pm0,13$	$7,69\pm0,36$	$8,00\pm0,21$
	WF	85,50±0,35**	$8,00\pm0,09$	$7,94\pm0,26$	$7,88\pm0,27$	$8,19\pm0,19$
	F24	86,31±0,09*,**	7,94±0,30	8,13±0,22	8,00±0,35	8,13±0,22
t2	MF	$84,13\pm0,35$	$7,81\pm0,13$	$8,00\pm0,28$	$7,94\pm0,19$	$7,94\pm0,13$
	WF	$82,81\pm0,44$	$7,62\pm0,40$	$7,69\pm0,25$	$7,38\pm0,33$	$7,81\pm0,37$
Standar	rd Control	81,00±0,38*	7,25±0,13	7,75±0,20	7,00±0,15	7,50±0,13
Negativ	e Control	80,50±1,23**	$7,25\pm0,24$	$7,31\pm0,24$	$7,25\pm0,20$	$7,50\pm0,13$

**TABLE 1 -** Average values in sensory quality, final score and acidity, fragrance/aroma, body and flavor in coffee samples from the fermentations with different conditions of coffee processes.

Values are expressed as mean  $\pm$  standard deviation. \*: treatments with significant differences (Dunnett p=0,05) on the standard control. \*\*: treatments with significant differences (Dunnett p=0,05) on the negative control

**TABLE 2** - Average values in sensory quality, final score and acidity, fragrance/aroma, body and flavor in coffee samples from the fermentations for 24h on coffee pulped in t2 level delaying time and with different levels of pH and temperature.

Temperature	рН	Final score	Acidity	Fragrance /aroma	Body	Flavor
	5,0	86,63±0,88*,a	8,13±0,18	8,19±0,18	8,13±0,18	8,13±0,18
17	4,0	$84,25\pm0,71^{ab}$	7,75±0,00	7,88±0,18	7,88±0,18	$7,75\pm0,00$
	WC	83,25±1,06ab	7,63±0,18	8,00±0,00	7,63±0,18	7,63±0,18
	5,0	$82,75\pm2,47^{ab}$	7,63±0,18	7,75±0,00	7,50±0,35	$7,50\pm0,35$
23	4,0	$86,00\pm1,41^{ab}$	7,88±0,18	8,13±0,18	$8,00\pm0,00$	8,25±0,35
	WC	80,88±1,59 <sup>b</sup>	7,13±0,18	7,75±0,00	7,25±0,35	7,38±0,35
Standard C	ontrol	81,75±0,35*	$7,50\pm0,00$	$7,75\pm0,35$	$7,25\pm0,00$	$7,50\pm0,00$

Values are expressed as mean  $\pm$  standard deviation. Means followed by the same letter are not significantly different (p=0,05), according to Tukey's test. \*: treatments with significant differences (Dunnett p=0,05) on the standard control

The other treatments did not present statistical differences in quality probably because of the tight variation range of the temperature and pH control in the process. Additionally, it is important to consider that, due to the exothermic behavior of the fermentation process, the temperature inside the coffee fermentation tank can increases 4,6 °C (CORREA et al., 2014) until 8,0 °C (PEÑUELA et al., 2010; VELMOUROUGANE, 2013) during a fermentation process for 16 h. The changes in this variable along the process can be difficult to control. Generally, the pH and temperature conditions controlled by the bioreactor generate coffee with quality comparable with that obtained by the effect of fermentation strategies of the first phase.

#### Organic acid composition

Most of the acids analyzed in this research are part of the chemical composition of a coffee bean given by fruit metabolic processes (FLAMENT 2002). Quinic, citric, malic and acetic acids were identified as the result of the development and maturity process of coffee fruits (ROGERS et al., 1999). Conversely, lactic acid in coffee has been related with metabolic activity of microorganisms (LÓPEZ et al., 1989, AVALLONE et al., 2001; EVANGELISTA et al., 2015; JACKELS; JACKELS, 2005; PEREIRA et al., 2016). However, all acids identified can also can be substrates or precursors related to the

different fermentation processes assessed. The concentrations of quinic, citric and malic acids determined in this research were in the average values reported from the green beans of Arabica coffee (5,5, 7,6 and 4,1 mg/g d.b., respectively) (JHAM et al., 2002; ROGERS et al., 1999).

Tables 3 and 4 present the acid concentration values corresponding to the average obtained by the extraction of the samples at the end of each treatment. The final concentrations of quinic, malic and succinic acids did not present variations that can be attributed to the fermentation strategies evaluated. The interaction among the factors, pulping delaying time and fermentation method

significantly affected the concentrations of lactic, acetic and citric acids (p<0,0001, 0,0031 and 0,0357, respectively) (Table 3). Treatments with longer times (t1-MF, t1-WF, t2-MF and t2-WF) showed the highest concentration of acetic and citric acids, meanwhile the greatest accumulation of lactic acid was associated to the longest pulping delaying time used in the evaluation.

In the same way, the fermentations conducted under combination of temperature and pH control in the bioreactor (Table 4), ANOVA showed a significant effect on the malic, quinic, lactic and acetic acid concentrations (p=0.0007, 0.0006, 0.0002 and <0.0001, respectively).

TABLE 3 - Organic acid concentrations (mg/g of coffee d.b.) in coffee samples from different fermentation strategies.

Time before pulping	Fermentation method	Acetic acid	Lactic acid	Citric acid	Malic acid	Succinic acid	Quinic acid
	F24	(12,00±0,78)b*,**	(2,93±0,10)c**	(4,43±2,18) <sup>b</sup>	1,53±0,80	1,31±0,20	5,17±1.53
t1	MF	$(25,48\pm1,01)^{a^*,**}$	$(4,36\pm0,26)^{c}$	$(6,45\pm0,27)^a$	$2,01\pm0,26$	$1,62\pm0,27$	$5,83\pm0,22$
	WF	(24,89±2,76)a*,**	$(7,45\pm1.39)^{b*}$	$(4,82\pm0,23)^{ab}$	1,63±0,09	0,43±0,13	5,51±0,56
	F24	(16.86±6.62)ab*,**	(8,40±0,99)b*,**	(3,12±1,38)b	1,13±0,78	1,46±0,89	4,28±1.79
t2	MF	(22,92±2,40)a*,**	(9,93±0,54)a*,**	$(4,03\pm1,68)^b$	1,16±0,62	N.D.	4,29±0,50
	WF	(20,16±2.87)ab*,**	(10,93±0,21)	(4,43±1,29) <sup>b</sup>	1,41±0,70	N.D.	4,26±0,99
Stand	ard Control	4,69±0,41	6,01±0,27	5,23±0,98	1,37±0,49	1,48±0,28	5,24±0,07
Negat	tive Control	2,31±0,25	4,88±0,25	N.D.	N.D.	N.D.	5,54±0,00

Values are expressed as mean  $\pm$  standard deviation. Means followed by the same letter are not significantly different (p=0,05), according to Tukey's test. \*: treatments with significant differences (Dunnett p=0,05) on the standard control. \*\*: treatments with significant differences (Dunnett p=0,05) on the negative control. N.D: Not detected

**TABLE 4** - Organic acid concentrations (mg/g of coffee d.b.) in coffee samples from fermentation processes for 24 h on coffee pulped in t2 level delaying time at distinct levels of pH and temperature

Temperature	рН	Acetic acid	Lactic acid	Quinic acid	Malic acid	Citric acid S	Succinic acid
	5,0	(12,38±0,47)bc	(0,95±0,19)b	(3,48±0,16)bc	0,38±0,05 <sup>b,*</sup>	1,82±0,56	0,27±0,01
17 °C	4,0	$(12,74\pm0,56)^{abc,*}$	$(3,76\pm0,24)^a$	$(3,63\pm0,44)^{abc}$	$0,43\pm0,06^{b,*}$	$2,28\pm0,03$	$1,18\pm0,11$
	W.C.	$(11,15\pm0,40)^{c}$	$(1,23\pm0,19)^b$	$(2,99\pm0,17)^{c}$	1,07±0,01a	1,69±0,10	$0,33\pm0,18$
	5,0	(14,33±0,67)abc,*	(3,58±1,29)a,*	(3,95±0,13)ab,*	0,28±0,10 <sup>b,*</sup>	1,70±0,42	N.D.
23 °C	4,0	$(15,05\pm1,73)^{a,*}$	$(3,94\pm1,02)^{a,*}$	$(4,51\pm0,36)^{a,*}$	$0,38\pm0,21^{b,*}$	2,45±1,12	$0,09\pm0,01$
	W.C.	$(14,36\pm1,03)^{ab}$	$(4,10\pm0,30)^{a,*}$	$(3,89\pm0,93)^{abc}$	0,43±0,05 <sup>b,*</sup>	$1,71\pm0,61$	N.D.
Standard Co	ontrol	12,51±2,05	1,35±0,68	3,15±0,40	2,06±1,06	4,07±0,55	0,56±0,19

Values are expressed as mean  $\pm$  standard deviation. Means followed by the same letter are not significantly different at p=0,05, according to Tukey's test. \*: Treatments with significant differences on the standard control (SC), according to Dunnett test at p=0,05. N.D: Not detected

The highest values of acetic, lactic and quinic acid seemed to be associated with a high fermentation temperature. The treatment performed at 17 °C and pH 5,0 obtained the most representative difference given by the low concentration of lactic acid. Furthermore, the main differences with the standard control were obtained with the coffee from fermentations conduced to 23 °C.

The previous results are explained as follows. Quinic acid is part of the natural chemical composition of green coffee beans, as it forms the molecular structure of chlorogenic acids along with caffeic acid. The quinic acid concentration was not altered by the fermentation processes applied in the first part of this study. Consequently, the microorganisms that contributed in the treatments did not use quinic acid as a carbon source. Nevertheless, under the conditions of the bioreactor, treatments at 17 °C showed a slight decrease in quinic acid compared with the other fermentation processes. This result suggests that microbial population could be stimulated under these conditions and use this compound as a substrate, reducing its concentration. Conversely, the increase in quinic acid concentration in coffee is directly related to the degradation of chlorogenic acids during roasting; this compound is also related to the astringency and body of coffee (SCA 2015).

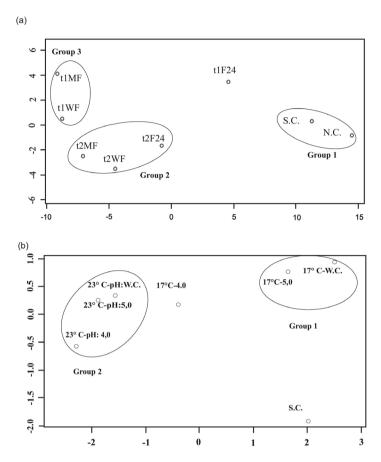
Citric and malic acids produce a beverage with pronounced acidity. However, they are not desirable in high concentrations because they can create an acidic and astringent taste (FLAMENT, 2002). These acids presented higher values in t1-MF. The concentration of succinic acid seemed to be associated with the low process time or low fermentation temperature, as this compound was not detected during longer fermentation and when the process temperature was 23 °C.

Conversely, the variation in the fermentation strategy directly affected the acetic acid production, as demonstrated by the difference in concentration between the treatments and the controls (Dunnett test). The higher levels of acetic acid were related to higher process times (t1-MF, t1-WF, t2-WF and t2-MF) or with a higher fermentation temperature (23 °C). Other studies reported that the accumulation of this compound in coffee occurs in the environmental temperature of 22 °C or when the fermentation time is greater than 20 h (BERTRAND et al., 2012). High acidity is known to be produced during fruit development and in the fermentation of coffee grown at high altitudes (low temperature) (BERTRAND et al., 2006). This condition has been found to influence the environmental conditions of some compounds in coffee, such as chlorogenic acids, lipids and sugars (JOËT et al., 2010). The concentrations of some organic acids are presumed to have the same behavior. The presence of acetic acid in coffee is related to the vinegar taste in the beverage; therefore, its excess production during fermentation is not recommended. During the fermentation process carried out for over 44 h, overfermented coffee beans and a vinegar taste were present along with an acetic acid concentration of 0,471 mg/mL (LÓPEZ et al., 1989). This value is higher than that reported in the current research (25,48 mg/g coffee d.b. or 0,172 mg/mL). Furthermore, none of the coffee samples analyzed in the sensory analysis reported a vinegar taste or a defect in cup.

In lactic acid production, the fermentation conditions at the highest temperature (23 °C) or the longest processing time, including the addition of water at the end of the process, generated a high concentration of this compound. Lactic acid is as a product of fermentation that increases during the process time, and its accumulation has been related to the decline of the pH value (AVALLONE et al., 2001; JACKELS; JACKELS, 2005; PEREIRA et al., 2016). Lactic acid bacteria are included in the microbial community involved in coffee fermentation (AVALLONE et al., 2001; EVANGELISTA et al., 2015; LÓPEZ et al., 1989; PEREIRA et al., 2016). The highest concentration of lactic acid coincided with the pH value of 3,77 in coffee fermentation mass.

The processes assessed showed that changing the development of fermentation is possible. Changes in the organic acid concentrations support this claim. In addition, CHAVERRA, (2016) found that microbial composition associated with these coffee fermentations changed according to the different conditions applied.

The spatial map generated by the application of Multidimensional Scaling (MDS) in the six organic acid concentrations facilitated the understanding of the relationship among the different coffee fermentation processes (Figure 1). Points that are close together represent similar treatments. Points far apart represent dissimilar treatments. For the fermentation strategies, (Figure 1a) coffee was classified into three groups, and one treatment (t1-F24) that coincided with the low concentration of lactic acid. Group 1 corresponded to the standard and negative controls, these treatments had the lowest means of acetic acid and the lowest final score.



**FIGURE 1 -** Similarity map of coffee samples according to organic acid concentrations obtained from (a) different fermentation strategies and (b) fermentations with different levels of pH and temperature.

The groups 2 and 3 was composed of treatments with different level of pulping delaying time thus indicating the effect of this condition on the composition of organic acids in green coffee beans, furthermore, were associated with coffee quality. The groups coincided with a high lactic acid concentration (Group 2) and the highest mean concentrations of acetic and malic acids (Group 3); the coffee from Group 3 was classified as specialty. Figure 1b shows two groups and two individual treatments. These groups reflected the effect of temperature on organic acid concentrations; because of the groups were conformed by treatments with the same level of temperature and similar concentrations of acetic and lactic acids mainly. Moreover, the graphical representation shows the dissimilarities between the control treatments and the coffee obtained by different methods to conduct the fermentation processes. The standard control and the treatment 17 °C and pH 4.0 were different from the groups: because both had intermediate values of acetic acid and high lactic acid concentrations. Finally, the

application of MDS correctly classified the coffee treatments according to the different fermentation processes, and it coincided with the results of coffee quality, especially the dissimilarities found in the control treatments.

#### **Volatile Organic Compounds**

Twenty-two volatile compounds were identified by the HS-SPME/GC-MS, which were grouped in eight alcohols, seven esters, three aldehydes, two acids and two ketones (Table 5). The compounds associated with thermal origin in green coffee such as furans and pyrroles, are produce in the drying stage, because they were not found in the freeze-dried samples (lyophilisation). Most of the compounds have been identified in green coffee beans and linked with the postharvest process (FLAMENT, 2002;), especially with the fermentation stage (GONZALEZ-RIOS et al., 2007). Alcohols and esters were the most abundant in the HS of green coffee, mainly ethanol and ethyl acetate, respectively.

<b>TABLE 5 -</b> Average values of Volatile Organic Compounds (VOCs) contents (Arbitrary Units x 10 <sup>5</sup> ) identified by
HS-SPME/GC-MS in samples of green coffee obtained by different fermentation processes

Compound	t2-F24	t2-WF	17°C pH 5,0	23°C pH 4,0	23°C pH SC	Standard Control	Negative Control
Acetic acid	1637,0	490,1	508,1	941,2	1438,3	1246,7	121,4
Pentanoic acid	45,4	17,9	76,4	60,9	46,0	150,4	27,2
Methanol	201,7	183,3	344,2	99,9	206,0	206,9	84,0
Ethanol	6050,2	6376,6	4078,3	4517,4	3600,6	1759,6	256,8
1-Propanol	13,8	8,8	13,4	26,9	16,4	12,5	0,3
1-Butanol	11,1	7,8	18,4	13,3	10,4	58,7	0,1
1-Penten-3-ol	16,0	6,4	7,4	12,6	9,3	0,1	2,3
2-Methyl-1-propanol	43,2	14,1	24,3	61,4	21,1	33,0	0,4
3-Methyl-1-butanol	25,4	10,6	28,4	38,2	10,4	39,9	2,0
3-Methyl-2-buten-1-ol	6,4	1,3	12,2	4,5	7,4	27,5	3,0
Ethyl-formate	5,0	1,1	1,4	3,7	3,1	4,4	0,3
Methyl-acetate	123,3	13,8	104,5	58,1	59,3	145,9	0,2
Ethyl-acetate	5068,3	593,3	1482,0	3086,5	583,2	820,1	1,8
Methyl-lactate	9,1	5,4	1,7	1,0	7,8	4,0	0,0
Ethyl-lactate	103,8	44,6	9,4	14,4	26,6	1,5	0,3
3-Methyl-1-Butanol acetate	22,3	13,0	4,4	22,0	3,9	11,4	7,6
Ethyl-isovalerate	30,3	23,7	69,5	106,9	22,3	45,1	0,4
Acetaldehyde	58,9	106,3	177,9	116,4	41,7	183,1	28,8
2-Methyl-propanal	3,7	5,1	42,2	5,3	2,5	68,5	0,7
n-Hexanal	9,8	4,0	19,8	4,9	3,8	18,8	3,1
2,3-Butanodione	33,6	7,4	13,7	51,3	7,6	31,7	7,8
3-Hydroxy-2-butanone	267,3	47,0	52,3	613,1	92,4	271,8	54,8

Furthermore, these chemical classes are important because of their aroma prolife is related with pleasant, fruity and floral notes. The acids, alcohols and esters were formed mainly because of microbial activity. Therefore, the lowest values of these chemical classes were for the negative control (without fermentation), which also had the lowest value in the fragrance attribute.

Composition of green coffee beans from the fermentation processes was different and produced samples richer in volatile compounds. Even though the alcohols were the main chemical class, different concentrations in the other classes were found. For example, the main compounds of the samples corresponding to highest quality qualification (t2-F24, 17°C-pH5,0 and 23°C-pH4,0) were esters mainly ethyl acetate, ethyl isovalerate and 3-methyl-1-butanol acetate (isoamyl acetate) which fruity odor. On the other

hand, ketones (3-hydroxy-2-butanone and 2,3 butanodione) in the sample corresponding to 23 °C-pH 4,0, can be considerate important given their characteristics of aroma. These treatments as part of Group 1, agree with the MDS analyzes (Figure 2).

On the other hand, samples corresponding to lowest quality score showed a higher concentration of aldehydes and acids, such as acetic acid, and acetaldehyde and hexanal, the latter related with insect damaged beans, which can cause undesirable flavors and odors (TOCI; FARAH, 2008). The similarity map shows the Group 2, composed by the samples corresponding to control treatments, exhibited low concentrations of alcohols, esters and ketones. In addition, the similarity map also shows two treatments with dissimilarity; t2-WF and 23 °C-pH WC, that differ mainly in the alcohols and acids content.

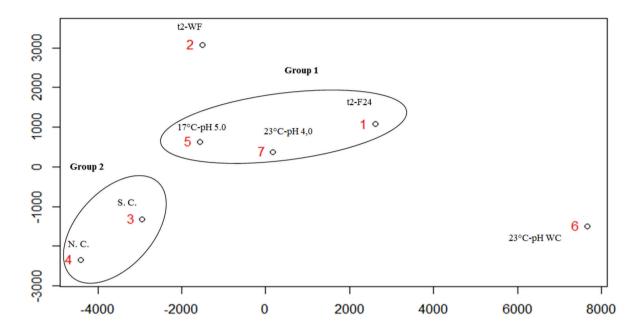


FIGURE 2 - Similarity map of coffee samples according to Volatile Organic Compounds (VOCs) contents.

Different contents of compounds in the samples suggest the possibility of modifying the composition through fermentation processes conducted under different conditions. These results suggest changes in microbial activity in the processes, given the variations to perform the fermentations. A similar behavior was observed in the composition of volatile organic compound from green coffee beans inoculated with yeast in dry method (EVANGELISTA et al., 2014) and in different regions of coffee production in Brazil with different conditions of air temperature (EVANGELISTA et al., 2015).

It is known that the aroma and taste of coffee develop during roasting (RIBEIRO et al., 2009). However, the final quality depends on the quality of the green bean, which in turn shows the characteristics given by the species and process type, in which fermentation has an important part in the formation of compounds as result of the metabolism of the microorganisms in the process. The utility of the results shown in this research work demonstrate the possibility of improving the coffee quality by using different fermentation techniques.

### **4 CONCLUSIONS**

The fermentation processes assessed increased the coffee quality regarding the control treatments. Therefore, this study is the first to show that variations in the coffee fermentation process and variables control, such as pH and

temperature, increase the coffee quality under the environmental conditions, variety and methods applied in this research.

The fermentations applied facilitated the production of organic acids as substrates or precursors along with organic compounds, depending on the carried fermentation process. The results suggest that is possible modulate the beverage acidity as well as the fragrance/aroma through organic acids production and alcohols, esters, ketones in a synergistic combination, and this can promote differentiated profiles in order to satisfy the requirements of specialty coffees.

Finding the best strategy to conduct fermentation processing is possible without requiring microorganism inoculation, which becomes a complex process in the conditions of coffee farms. Acetic acid was the main compound that changed its concentration because of variations in the fermentation process. The concentrations of the acids identified were not associated with quality damage.

#### **5 ACKNOWLEDGEMENTS**

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## SIMULATION OF COFFEE FRUIT DRYING USING COMPUTATIONAL FLUID DYNAMICS

Rudney Amaral<sup>1</sup>, Ednilton Tavares de Andrade<sup>2</sup>, Francisco Carlos Gomes<sup>3</sup>, Flávio Meira Borém<sup>4</sup>, Isabela Avila Lemos<sup>5</sup>, Camila de Almeida Dias<sup>6</sup>

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**ABSTRACT**: Drying is a fundamental step in post-harvest handling of coffee because moisture content at the end of drying affects several important aspects, such as sensory quality, storability, and color of the fruit coffee. Within this context, the aim of this study is to determine water distribution within the natural coffee fruit during and at the end of the drying process. For that purpose, simulations were made through finite elements using computational fluid dynamics. Experimental data on moisture content of coffee fruit in the "cherry" stage were collected during drying, which was carried out at a temperature of  $40^{\circ}$ C and relative humidity of 25% to 0.18 decimal l(dry basis – d.b.) to compare the results of the experiment with the results of the simulations. Ten mathematical models of the drying process were developed for the collected data. The two-term exponential model best fit the data. The results of the simulations in computational fluid dynamics were compared to the results from experimental drying, and a satisfactory fit was obtained. The effective diffusivity coefficient ( $D_{eff}$ ) was developed for the model proposed, obtaining the value of  $2.87 \times 10^{-11}$  m<sup>2</sup> s<sup>-1</sup>. At the end of drying, the model exhibited 57.1% of the projection area of the coffee fruit with moisture content below 0.18 decimal (d.b.). Thus, the model can be used for other applications.

Index terms: Moisture content, distribution, natural coffee, finite elements, diffusion.

## SIMULAÇÃO DA SECAGEM DE FRUTOS DE CAFÉ UTILIZANDO FLUIDODINÂMICA COMPUTACIONAL

RESUMO: A secagem é uma importante etapa da pós-colheita do café, visto que o teor de água ao final da secagem influencia questões importantes como qualidade sensorial, armazenabilidade e cor dos frutos de café. Mediante isso, a determinação da distribuição do teor de água no interior de um fruto de café natural durante e ao final do processo de secagem é o objetivo desse trabalho, onde foram realizadas simulações por meio de elementos finitos, utilizando a fluidodinâmica computacional. Foram coletados dados experimentais de teor de água durante a secagem de frutos cereja a qual foi realizada a uma temperatura de 40°C e umidade relativa de 25% e até que os frutos atingissem 0,18 decimal (b.s.) visando a validação do modelo. Foram ajustados aos dados coletados dez modelos matemáticos de secagem. O modelo Exponencial de Dois Termos foi o que melhor se ajustou aos dados. Os resultados das simulações em fluidodinâmica computacional foram ajustados aos resultados da secagem experimental, obtendo um ajuste satisfatório. O coeficiente de difusividade efetivo (D<sub>eff</sub>) foi ajustado ao modelo proposto obtendo-se o valor de 2,87 x 10<sup>-11</sup> m² s<sup>-1</sup>. Ao final da secagem, o modelo apresentou 57,1% da área de projeção do fruto de café com teor de água abaixo de 0,18 decimal (b.s.). Desta forma, o modelo pode ser utilizado para outras aplicações.

Termos para indexação: Teor de água, distribuição, café natural, elementos finitos, difusão.

#### 1 INTRODUCTION

Coffee has a prominent position in Brazilian grain production, with 349,9 million dollars in exports, according to the Informe Estatístico do Café (2018).

The drying stage is fundamental in coffee to prevent the growth of microorganisms and fermentations that can compromise the quality of newly-harvested coffee, which has high moisture content and uneven maturity of the coffee fruit (berries) (RESENDE et al., 2009). The temperature of the coffee fruit must be strictly controlled because an excessive increase in temperature greatly affects its quality. Coffee should be dried at a temperature of 40°C for this operation not to affect beverage aroma and flavor (BORÉM, 2008).

Air flow during coffee drying has an effect only at the beginning of drying (Burmester; Eggers, 2010). In drying coffee fruit at low relative humidity, an increase in temperature brings about an increase in the effective diffusivity coefficient ( $D_{\rm eff}$ ), which ranges from 1.908 to 3.721 x  $10^{-11}$  m<sup>2</sup> s<sup>-1</sup>, as well as in drying rate (Alves, et al. 2013).

Nilnont et al., (2012) modeled drying in parchment coffee through finite elements. Through this modeling, they obtained a model sufficient for predicting moisture content during the drying process. According to the authors, the two-dimensional model provides better understanding of transport processes in different components of parchment coffee. The mean values of the coefficient of liquid diffusion of the coffee bean

<sup>1.2.3.4.5.6</sup>Universidade Federal de Lavras/UFLA - Departamento de Engenharia/DEG - Cx. P. 3037 - 37.200-000 - Lavras - MG rudneymec@yahoo.com.br, ednilton@deg.ufla.br, fcgomes@deg.ufla.br, flavioborem@gmail.com, isa\_lemosti@hotmail.com, camila.almeidadias@gmail.com

and of the coefficient of liquid diffusion of the parchment found by the authors are 7.173 x 10<sup>-10</sup> and 6.737 x 10<sup>-13</sup> m<sup>2</sup> s<sup>-1</sup>, respectively.

During the process of drying agricultural products, conditions are considered isothermal, and water transfer is restricted to the surface of the product (Bergman, et al. 2011). Thus, the aim of this study was to determine the distribution of moisture content within natural coffee fruit during and at the end of the drying process. A computational fluid dynamic model was used to describe the process of heat and matter transfer within the fruit. This model allows analysis of the liquid diffusion rate in the coffee fruit during drying, as well as the distribution of moisture content at the end of drying.

#### 2 MATERIALS AND METHODS

Mature or "cherry" coffee fruit (*Coffea arabica* L. cv. Catuai Vermelho) was used. The fruit was harvested manually. Fruit with lower specific weight (shriveled, floaters, etc.) were removed through separation in water. The fruit had average moisture content of 2.11 decimal (dry basis – d.b.) at the beginning of drying. The moisture content of the coffee fruit was determined by the standard method of ISO 6673 (INTERNATIONAL ORGANIZATION for STANDARDIZATION - ISO, 2003).

Before the beginning, during, and at the end of mechanical drying, the main dimensions of 10 samples of the coffee fruit were measured. These measurements were made with a digital caliper rule with 0.01 mm readability, resulting in the mean dimensions shown in Table 1.

The volume of the coffee berry (V) was calculated by equation 01. The coffee berries were considered as oblate spheroids. The equivalent radius of the coffee berries was also calculated, defined as the radius of a sphere with volume equivalent to the volume of the berry.

$$V = \frac{4 \pi (abc)}{3}$$
 (1)

TABLE 1 - Dimensions of natural coffee fruit

Axis	Dimension (m)	Dimension used (m)
Length (a)	1.4*10-3	7*10 <sup>-3</sup>
Width (b)	1.1*10-3	5.5*10-3
Thickness (c)	$0.9*10^{-3}$	5.5*10-3

In which V is the volume at each level of moisture content (m<sup>3</sup>), a is the length (m), b is the width (m), and c is the thickness (m).

The samples were weighed on a precision digital balance with 0.01 g readability. The true specific weight before drying the fruit was calculated by the simple ratio between weight and volume, as well as the true specific weight at the end of drying. The true mean specific weight of the fruit during the drying process was also calculated.

The system for drying the samples was composed of an air conditioning unit coupled to a fixed bed dryer (Figure 1).

The laboratory air conditioning system (LACS) was used to control the characteristics of the drying air according to the model proposed by Fortes et al. (2006). This equipment allows control of the flow, temperature, and relative humidity of the drying air and is composed of a cooling system with three air conditioning units. The dryer included four removable square trays perforated at the bottom, with sides of 0.3 m and depth of 0.1 m, located over a plenum to make the air flow uniform. The temperature of the drying air was measured in the plenum, under the perforated trays, by thermocouples connected to universal controllers (Novus N1100). A digital hygrometer inserted in the plenum measured the relative humidity of the drying air. A frequency inverter (Weg CFW-10) controlled the rotation of the centrifugal fan that blows the heated air into the plenum; it thus regulated the air flow.

The procedure began by completely filling the perforated-bottomtrays. For each hour of drying, the trays were removed from the dryer and weighed on an analytical balance with 0.01 g readability. This procedure was repeated until the fruit was dried to 0.18 decimal (d.b.); the standard laboratory oven method (BRASIL, 2009) was used to determine the moisture content of the fruit. Total drying time was 109 hours. The tray position was revolved 90° every hour to minimize possible differences of temperature and air flow among the perforated-bottomtrays.

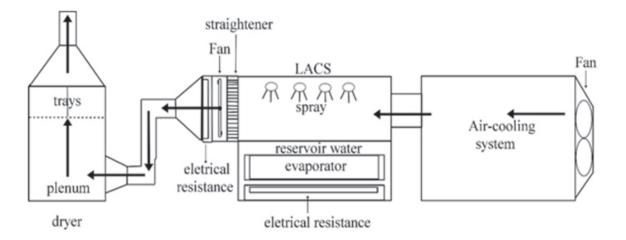


FIGURE 1 - System used in mechanical.

The air flow variables of the drying air were kept constant throughout the procedure – the temperature was 40°C, relative humidity was 25% and air speed was 0.33 m s<sup>-1</sup>, for a flow of 20 m<sup>3</sup>min<sup>-1</sup>m<sup>-2</sup>.

For the drying conditions of this study, the equilibrium moisture content of the fruit was calculated using the model proposed by Afonso Júnior (2001) and cited by Borém (2008) for "cherry" fruit (Equation 2).

$$EMC = (2.0222 + (0.0288T) - (18.7397 RH^{86181}))^{-2.1385}$$
 (2)

In which is the equilibrium moisture content (d.b.), is the drying air temperature (°C), and is the relative humidity of the drying air (decimal).

The moisture ratio was calculated from fitting equation (3) to the data observed using the STATISTICA® 5.0 (Statsoft, Tulsa, USA) software. The moisture ratio (MR) is essential for describing different drying models in a thin layer. Equation (3) is the analytical solution for Fick's second law, considering the geometric form as spherical and considering the surface contour condition as known (BROOKERet al., 1992).

$$MR = \frac{M-EMC}{M_{i}-EMC} = \frac{6}{\pi^{2}} \sum_{n=1}^{\infty} \frac{1}{n^{2}} \exp \left[ -\frac{n^{2}.\pi^{2}.D.t}{9}.\left(\frac{3}{R}\right)^{2} \right]$$
 (3)

in which MR is the moisture ratio of the product, adimensional; M is the moisture content of the product, decimal (d.b.); EMC is the equilibrium moisture content of the product, decimal (d.b.); M<sub>i</sub> is the initial moisture content of the product, decimal (d.b.); D is the effective diffusivity

coefficient (m<sup>2</sup>s<sup>-1</sup>), and R is the equivalent radius of the coffee fruit (m);

The mathematical models disposed in Table 2 by Equations (4) to (14) were fitted to the experimental data of coffee drying.

The choice of the best model was based on the statistical parameters: standard deviation of the estimate (SE), mean relative error (P), coefficient of determination (R2), and the chi-square , calculated by equations 15 to 17. in which SE is the standard deviation of the estimate (decimal), Y is the experimentally observed value, is the value calculated by the model, DFR is the number of degrees of freedom of the model, P is the mean relative error (%), and n is the number of data observed.

For the purpose of understanding heat and mass transfer in coffee fruit during drying, the distribution of moisture content can be identified within the coffee berry by mathematical modeling. Under computational fluid dynamics, hypotheses can be analyzed more easily since computational fluid dynamics is a technique that has been used for evaluation of various physical phenomena using the finite element method. This technique is a set of computational tools based on numerical algorithms to solve and interpret problems in the flow of fluids (SCHNEIDER; MALISKA,2000).

For fluid flows, the mathematical model is based on equations of conservation of the amount of movement, conservation of mass, and conservation of energy. Analytical solution of these equations is only possible for very simple flows.

**TABLE 2** - Mathematical models used to predict drying.

Model	Equation	
Diffusion Approximation	$MR = a \cdot exp(-k \cdot t) + (1 - a) \cdot exp(-k \cdot b \cdot t)$	(04)
Two Term	$MR = a . exp (-k_0 . t) + b . exp (-k_1 . t)$	(05)
Two-Term Exponential	$MR = a \cdot exp(-k \cdot t) + (1 - a) exp(-k \cdot a \cdot t)$	(06)
Logarithm	$MR = a \cdot exp(-k \cdot t) + b$	(07)
Midili	$MR = a. Exp (-k.t^n) + b.t$	(08)
Newton	$MR = \exp(-k \cdot t)$	(09)
Page	$MR = Exp(-k.t^n)$	(10)
Modified Page	$MR = Exp (-(k.t)^n)$	(11)
Thompson	$MR = \exp((-a - (a^2 + 4 \cdot b \cdot t)^{0.5}) / 2 \cdot b)$	(12)
Verna	$MR = a \cdot exp(-k.t) + (1-a) exp(-k1.t)$	(13)
Wang and Singh	$MR = 1 + at + bt^2$	(14)

MR is the moisture ratio (dimensionless); k, k0, k1 are drying constants (s-1); t is the drying time (h); a, b, n are constants that depend on the nature of the product (dimensionless). (BROOKER et al., 1992; Corrêa et al., 2001; Alves et al., 2013; Filho et al., 2015).

$$SE = \sqrt{\frac{\sum (Y - \hat{Y})^2}{DFR}}$$
 (15)

$$P = \frac{100}{n} \cdot \sum_{\gamma} \frac{|\gamma - \hat{\gamma}|}{\gamma} \tag{16}$$

$$\chi^2 = \frac{\sum (Y - \hat{Y})^2}{DFR} \tag{17}$$

To analyze real problems in which strong temporal variations and turbulence are found, numerical methods are used that are able to analyze flows in a general manner through discretization of the equations cited. Computational fluid dynamics is divided into three basic steps: the first is pre-processing, in which the geometry and the numerical mesh is generated and the contour conditions are defined; the second step is processing, which is solution of the mathematical equations using discretization; and the third step is post-processing in which the information, graphs, and analyses generated in the solution are visualized (SILVA et al., 2012).

According to Crank (1975), there is an analogy between thermal conduction and liquid diffusion. The mathematical theory of diffusion in isotropic bodies is based on the hypothesis that

the transfer rate of a substance diffused through the unit area of a section is proportional to the gradient of concentration measured normally at the section. In an analogous manner, the transfer rate of heat through the unit area of a section is proportional to the temperature gradient measured normally at the section. Table 3 shows the analogy between the two processes:

The analogy between liquid diffusion and thermal conduction was used to develop the computational simulation of coffee drying using modeling by the commercial software ANSYS® ED™(version 9.0) through programming in APDL (Ansys Parametric Design Language) language.

To carry out simulations, geometry that corresponds to one fourth of a coffee berry (axisymmetric) was chosen, as shown in Figure 2, considering axial symmetry of the coffee berry.

TABLE 3 - Analogy between liquid diffusion and thermal conduction.

#### LIQUID DIFFUSION THERMAL CONDUCTION

Fick 1855 Fourier 1822

D is the diffusion coefficient K is thermal conductivity

C is concentration of the substance is temperature

 $F(t) = -D \,\partial C/\partial x \qquad \qquad F(t) = -K \,\partial \theta/\partial x$ 

F is the diffusive flux and/or heat flux

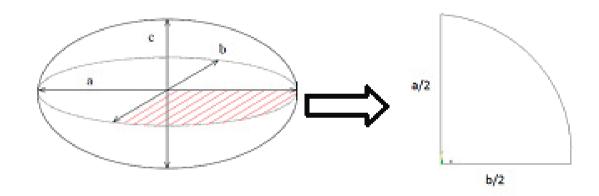


FIGURE 2 - Representation of a coffee berry as an oblate spheroid.

The main dimensions of "cherry" coffee fruit were measured with a digital caliper rule with 0.01 m readability. The main dimensions of a coffee berry are shown in Table 1. The a and b values of  $6.9132 \times 10^{-3}$  m and  $5.7347 \times 10^{-3}$  m, respectively, were used for the geometry implemented.

A thermal element called PLANE77 (Figure 3) was chosen, which is a two-dimensional quadrangular element with eight nodes applicable to two-dimensional thermal analyses according to Ansys (2011) to make up the computational mesh. The mesh was regular with its three sides divided into the same number of elements. This procedure divided the geometry presented into 507 elements and 1600 nodes (Figure 4).

The initial conditions and the contour conditions were also defined in pre-processing.

The effective diffusivity coefficient  $(D_{\text{eff}})$  used in the simulation was fitted to the real data to represent the experimental data.

The total fruit drying time in the computational model was 109 hours, which was the same time obtained experimentally with the fruit in order to dry to mean moisture content of 0.18decimal (d.b.).

The initial condition used for solution of the problem was to apply the same concentration of moisture content to all the nodal points of the mesh. For that purpose, the initial concentration of moisture content was calculated at 861.55 kg(H<sub>2</sub>O).m<sup>-3</sup>. This value was obtained from the product of the fruit moisture content in dry basis and the specific weight of the fruit. This initial condition corresponds to moisture content of 2.11 decimal (d.b.). For the value of specific weight, its mean variation in time was considered, obtaining the value of 1305 kg m<sup>-3</sup>. The dry matter part of this specific weight was considered for use of the model (408.3 kg m<sup>-3</sup>).

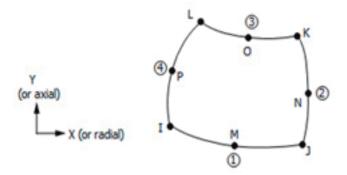
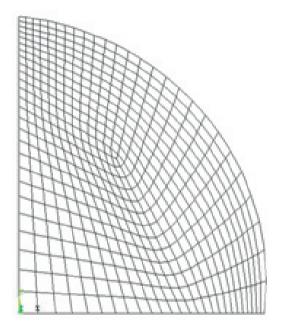


FIGURE 3 - PLANE77 element.



**FIGURE 4** - Representation of the element mesh obtained in the model.

The concentration of equilibrium moisture content was applied on the outside surface of the fruit, represented by the arc in the geometry implemented in the simulations. The concentration of equilibrium moisture content is the product of the equilibrium moisture content of the coffee fruit (calculated by equation 02) and its specific weight. The value of the concentration of the equilibrium moisture content was calculated at 34.53 kg(H<sub>2</sub>O).m<sup>-3</sup>.

Considering the above, the source program was developed from the menu of commands available in the library of the Ansys® software, applying the initial conditions and contour conditions calculated. Thus, the problem of the process for removing water from the coffee fruit was resolved.

Through simulation, the nodal values of concentration of moisture content were obtained in each minute of the drying process, as well as the distribution of moisture content within the fruit. The simulation came to an end exhibiting 720 iterations in all.

To evaluate the efficiency of the model developed computationally in APDL language for liquid diffusion of the coffee fruit, the results of the model were compared to the results obtained experimentally. The moisture ratio curve for the two cases was compared.

The mean relative error (P) was calculated by equation 16 from the drying curves obtained by the computational model in order to carry out statistical parameterization.

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#### **3 RESULTS AND DISCUSSION**

The mean calculated volume of the coffee berry before drying was  $9.922 \times 10^{-5} \, \mathrm{m}^3$ . The mean measured weight of the coffee berry was 1.52g and the true specific weight of the fruit before drying was  $1540.69 \, \mathrm{kg} \, \mathrm{m}^{-3}$ . At the end of drying, when the fruit had a moisture content of  $0.18 \, \mathrm{decimal}$  (d.b.), the true specific weight was  $918.84 \, \mathrm{kg} \, \mathrm{m}^{-3}$ , and the true mean specific weight during drying was  $1305 \, \mathrm{kg} \, \mathrm{m}^{-3}$ .

The initial equivalent ratio of the samples was  $6.18 \times 10^{-3}$  m, and the equilibrium moisture content was  $8.458 \times 10^{-2}$  decimal (d.b.).

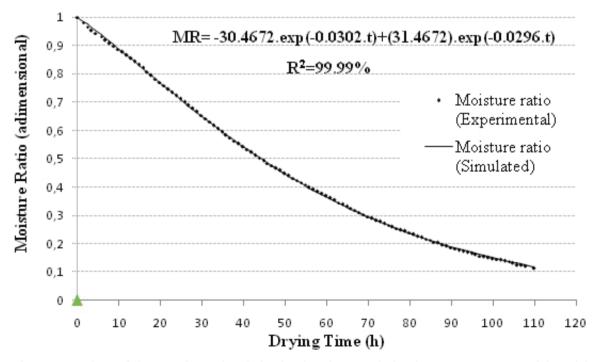
The moisture ratio according to time was calculated by equation (03). Figure 5 shows the comparison of the simulated values and the experimental values for the variation of the

moisture ratio (MR) as a function of time by the two-term exponential model, which best fit the experimental data. In Table 4 are the values of the coefficient of determination (R<sup>2</sup>), mean relative error (P), standard deviation of the estimate (SE), chi-square and the constants of the two-term exponential model. This model exhibited satisfactory values of the mean relative and estimated errors, as well as a high coefficient of determination (R<sup>2</sup>). Values of mean relative error below 10% mean a good fit of data to the drying phenomenon according to Madamba et al. (1996) and confirmed by Mohapatra and Rao (2005).

The effective diffusivity coefficient ( $D_{\rm eff}$ ) was calculated at the value of 1.86 x 10<sup>-11</sup> m<sup>2</sup> s<sup>-1</sup>. This result is corroborated by Alves et al. (2013), who found a variation of this coefficient from 1.908 to 3.721 x 10<sup>-11</sup> m<sup>2</sup> s<sup>-1</sup>.

**TABLE 4** - Coefficient of determination ( $R^2$ , %), mean relative error (P, %), standard deviation of the estimate (SE), chi-square ( $\chi^2$ ), and the constants of the two-term exponential model.

Temperature	Statistical parameter			Constants of the model			
	χ²	P	SE	R <sup>2</sup>	a	b	k
40°C	0.0004	0.73	0.022	99.99	30.467	0.9789	-0.0302



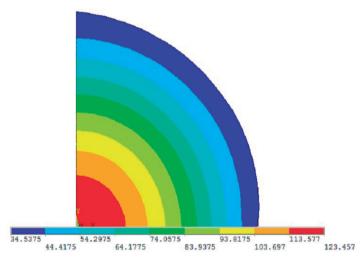
**FIGURE 5** -Values of the experimental and simulated moisture ratio by the two-term exponential model for drying of coffee fruit as a function of time (h).

The value obtained for the effective diffusivity coefficient (Deff)that best fit the experimental data was 2.87 x 10<sup>-11</sup> m<sup>2</sup>s<sup>-1</sup>. This value is in agreement with the results presented in the study of Isquierdo (2011), who studied the drying kinetics for natural coffees under different temperatures and relative humidities of the drying air and concluded that the effective diffusivity coefficient ranges from 1.460 x 10<sup>-11</sup>to 3.993 x 10<sup>-11</sup> m<sup>2</sup> s<sup>-1</sup>. This is also in agreement with the result obtained by Alves et al. (2013), who obtained the effective diffusivity coefficient (Doff) of water in coffee fruit ranging from 1.908 x 10-11 to 3.721 x 10<sup>-11</sup> m<sup>2</sup> s<sup>-1</sup>. The value obtained in a computational manner for the effective diffusivity coefficient (D<sub>eff</sub>) is a mean value since the components of a coffee berry were not considered, such as the parchment. In fact, each component has a specific effective diffusivity coefficient (D<sub>eff</sub>).

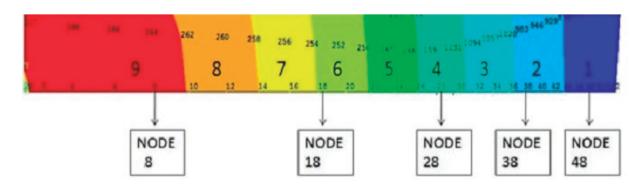
The knowledge of the distribution of water inside the fruit provides better drying, seeking mechanisms to accelerate the process without compromising its structure and quality, enabling the development of new forms and more efficient drying equipment. In addition, it is possible to obtain a better way of handling the drying in order to maintain the quality of the product with a less aggressive drying. Distribution of the moisture content within the coffee berry at the end of the drying process for the berries analyzed in this study is shown in Figure 6. Each color in the Figure indicates a range of concentration of moisture content.

The numbered areas in Figures 6 and 7were calculated, as well as the percentage of each one in relation to the area of the entire berry.

In Table 05, the approximate percentage of each area of Figure 07 is shown.



**FIGURE 6** - Distribution of concentration of moisture content [kg(H<sub>2</sub>O) m<sup>-3</sup>] resulting from simulation of drying of coffee fruit.



**FIGURE 7** - Representation of the nodal points involved in each area on the X axis.

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<b>TABLE 5</b> - Distribution of moisture content in different areas from the center to the outside extremity of the coffee
berry after drying.

No.	Area (m²)	Equivalence (%)	Water concentration (kg(H <sub>2</sub> O) m <sup>-3</sup> )	Moisture content decimal (d.b.)
1	2.09E-05	16.4	34.54 – 44.42	0.08 - 0.11
2	1.75E-05	13.7	44.42 - 54.30	0.11 - 0.13
3	1.67E-05	13.1	54.30 - 64.18	0.13 - 0.16
4	1.77E-05	13.9	64.18 - 74.06	0.16 - 0.18
5	1.21E-05	9.5	74.06 - 83.94	0.18 - 0.21
6	1.19E-05	9.4	83.94 - 93.82	0.21 - 0.23
7	1.12E-05	8.8	93.82 - 103.70	0.23 - 0.25
8	9.65E-06	7.6	103.70 - 113.58	0.25 - 0.28
9	9.72E-06	7.6	113.58 - 123.46	0.28 - 0.30

Area 1 (shown in blue) is the largest and the one that lost most water, as expected, because it is the outside surface of the berry. A band composed of the sum of areas 1, 2, 3, and 4 is equivalent to 57.1% of the total area of projection of a coffee berry. In Table 04, the values of the range of concentration of moisture content in each area are shown. In areas from 1 to 4, the highest concentration of moisture content is 74.06kg(H<sub>2</sub>O) m<sup>-3</sup>, equivalent to 0.18decimal (d.b.) of moisture content.

The evolution of the liquid diffusion process of moisture content in coffee fruit over the drying time is illustrated in Figures 08 and 09, for which each figure represents 24 hours of evolution of the liquid diffusion process.

Figures 8 and 9 show that evolution of the water diffusion process within the coffee berry is inversely proportional to drying time; that is, liquid diffusion is increasingly slower over time. This generates greater difficulty in removing water from the fruit at the end of drying, thus increasing the risks of losses in product quality. This is in agreement with several studies already described in the literature, such as the study of Saath et al. (2010).

Figure 10 shows the liquid diffusion that occurred within a coffee berry during simulation in vector format. The difficulty of removing water from the points nearest the center of the berry can be noted from the figure.

Figure 11 shows the values of concentration of moisture content (kg(H<sub>2</sub>O) m<sup>-3</sup>) for some nodal points of the computational mesh as a function of time. From Figure 11, it can be observed that in the center of the berry, moisture content is higher. The line that represents the drying curve for nodal point 08 is very different from the drying curve of nodal point 48. This occurs because nodal point 08 is at 0.12 x 10<sup>-2</sup> m from the center of the berry and nodal point 48 is at 0.54 x 10<sup>-2</sup> m from the center of the berry. In addition to the drying curves of nodal points 08 and 48, Figure 11 shows the drying curves for nodal points 18, 28, and 38 indicated in Figure 7.

The values of moisture ratio for the experimental data compared to the simulated data as a function of time are shown in Figure 12. The simulation carried out adequately represents coffee fruit drying. The experimental data and simulated data matched satisfactorily. Thus, the model can be used for other applications that require representation of this phenomenon. The mean relative error (P) was 1.8%, which is considered satisfactory.

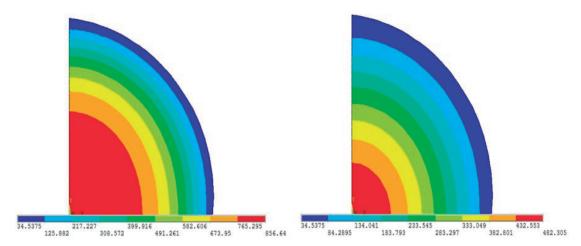


FIGURE 8 - Visualization of the field of moisture content for 24 and 48 hours of drying.

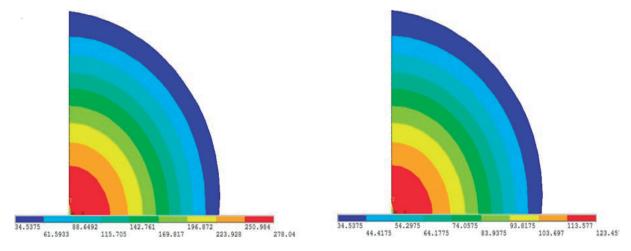


FIGURE 9 - Visualization of the field of moisture content for 72 and 96 hours of drying.

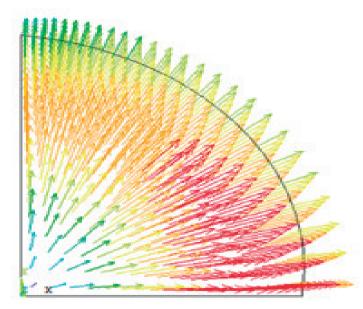


FIGURE 10 -Vector representation of liquid diffusion that occurred within the coffee berry.

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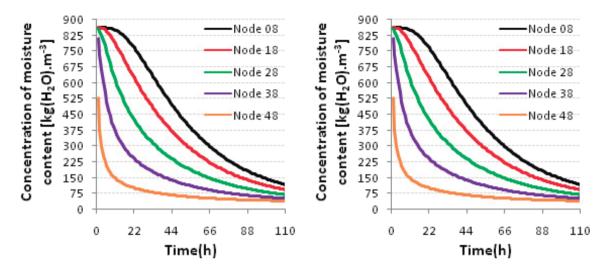


FIGURE 11 - Drying curves for some nodes chosen on the X axis of the geometry.

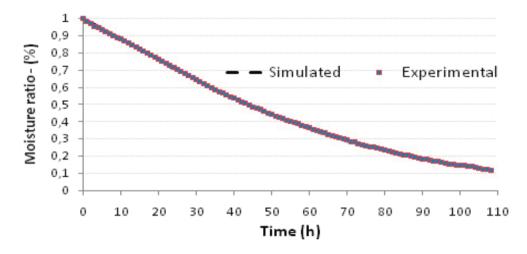


FIGURE 12 - Values of moisture ratio for the experimental data compared to simulated data.

#### 4 CONCLUSIONS

According to the results obtained in this study, we conclude that:

- The model developed in a computational manner by the technique of computational fluid dynamics for drying natural coffee fruit satisfactorily fit the experimental data with a mean relative error of 1.8%.
- For drying air temperature of 40°C and relative humidity of 25% at the end of drying, 57.1% of the area of projection of natural coffee fruit has moisture content below 0.18 decimal (d.b.).
- The effective diffusivity coefficient found by the model was  $2.87 \times 10^{-11} \text{ m}^2 \text{ s}^{-1}$ .

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# SPROUTING INDUCTION FOR MICRO-CUTTING ON IN VITRO CLONED ARABICA COFFEE PLANTS

Paula Cristina da Silva Angelo<sup>1</sup>, Iran Bueno Ferreira<sup>2</sup>, André Moraes Reis<sup>3</sup>, Lucas Bartelega<sup>4</sup>, Carlos Henrique Siqueira de Carvalho<sup>5</sup>, Ana Carolina Ramia Santos Paiva<sup>6</sup>, José Braz Matiello<sup>7</sup>

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**ABSTRACT:** Vegetative propagation of arabica coffee plants selected by their agronomic value has been accomplished routinely in Brazil for scientific purposes, through somatic embryogenesis and rooting of stem cuttings. Somatic embryogenesis is the election method when a very high number of cloned plants is demanded. Nevertheless, the costs of *in vitro* multiplication make difficult to explore it commercially. The experiments described herein aimed to amplify the number of *in vitro* cloned plants, post acclimatization, to reduce costs. Different concentrations of 2,3,5-triiodobenzoic acid (TIBA) and its association with benzylaminopurine (BAP) were applied, as successive pulses, in the 3<sup>rd</sup>, 8<sup>th</sup> and 13<sup>th</sup> months after transference to the greenhouse, on the same set of Catucaí and Siriema *in vitro* cloned plants, to induce sprouting. At the 8<sup>th</sup> month, the experiments with *in vitro* cloned Catucaí plants were reproduced in the nursery, for comparison. Best results were observed for the association TIBA 1000 mg.mL<sup>-1</sup> + BAP 60 mg.mL<sup>-1</sup> applied in the greenhouse, at the 13<sup>th</sup> month, when, on average, 8.5 and 7.0 micro-cuttings above 1 cm in length were produced using sprouts taken from each Catucaí and Siriema acclimatized plant, respectively. Applying this treatment twice a year, and harvesting induced sprouts each six months after the induction treatments, approximately 15 plants per each acclimatized one can be produced. The most important effect of TIBA was the induction of sub-apical sprouting. Greenhouse would be the best environment to apply successive pulses of sprouting inducers to coffee *in vitro* cloned plants.

Index terms: Cloning, micropropagation, cutting, growth regulators, Coffea arabica.

# INDUÇÃO DE BROTAÇÕES PARA MICROESTAQUIA EM CAFEEIROS ARABICA CLONADOS IN VITRO

RESUMO: A propagação vegetativa de cafeeiros arábica selecionados em função de seu valor agronômico tem sido realizada rotineiramente no Brasil para fins científicos, através de embriogênese somática e enraizamento de estacas. A embriogênese somática é o método eleito quando há demanda por um número muito grande de plantas clonadas. No entanto, os custos da multiplicação *in vitro* a tornam difícil de explorar comercialmente. Os experimentos descritos neste trabalho visaram amplificar o número de plantas clonadas *in vitro*, após a aclimatização, com o objetivo de reduzir o custo de clonagem. Diferentes concentrações de ácido 2,3,5-triiodobenzóico (TIBA) e sua associação com benzilaminopurina (BAP) foram aplicadas, na forma de pulsos sucessivos, no 3°, 8° e 13° meses depois da aclimatização, em um mesmo conjunto de plantas de Catucaí e Siriema clonadas *in vitro*, para induzir brotações, em casa de vegetação. No 8° mês, os experimentos com plantas de Catucaí foram reproduzidos em viveiro telado, para comparação. Os melhores resultados foram observados para a combinação de 1000 mg.mL<sup>-1</sup> de TIBA + 60 mg.mL<sup>-1</sup> de BAP, aplicados na casa de vegetação, no 13° mês, quando foram obtidas, em média, 8,5 e 7,0 microestacas para cada planta de Catucaí e de Siriema aclimatizada, respectivamente. Aplicando este tratamento duas vezes por ano, aproximadamente 15 plantas por cada planta aclimatizada podem ser produzidas. O efeito mais importante do TIBA foi estimular brotações subapicais. A casa de vegetação pareceu ser o melhor ambiente para aplicar pulsos sucessivos de indutores de brotações em cafeeiros clonados *in vitro*.

Termos para indexação: Clonagem, micropropagação, estaquia, regulador de crescimento, Coffea arabica.

## 1 INTRODUCTION

The agronomic value of coffee plants highly productive and resistant to diseases, which are selected during long lasting breeding programs, justifies the use of vegetative propagation methods in order to capture all of the plants best characteristics at the end of a reduced period of time (VOS; SNIJDER, 2000).

Vegetative propagation through the rooting of cuttings has been routinely used to clone *Coffea canephora* plants (SANTOS et al., 2013; AQUINO et al., 2017). More recently, *C. arabica* cuttings have been rooted as well (JESUS; CARVALHO; SOARES, 2006; CARVALHO et al., 2008; OLIVEIRA et al., 2010; REZENDE et al., 2010; BALIZA et al., 2013). Micropropagation *in vitro* through somatic embryogenesis also has been used

<sup>&</sup>lt;sup>1</sup> Embrapa Café/ Instituto Agronômico do Paraná - Rod. Celso Garcia Cid, km 375 - Cx.P. 10.030 - 86.057-970 - Londrina - PR paula.angelo@embrapa.br

<sup>2§ -</sup> In memoriam

<sup>4.6.7</sup> Fundação Procafé - Alameda do Café, 1000 - Vila Verônica - 37.026 - 483 - Varginha - MG lucas@fundacaoprocafe.com.br, carol@fundacaoprocafe.com.br, jb.matiello@gmail.com

<sup>&</sup>lt;sup>3</sup>Centro Universitário do Sul de Minas/UNIS - Rod. Varginha-Elói Mendes, Km 232 - 37.100-000 - Varginha - MG - andremoraesreis@gmail.com

Embrapa Café/Fundação Procafé - Alameda do Café, 1000 - Vila Verônica - 37.026 - 483 Varginha - MG - carlos.carvalho@embrapa.br

for *C. arabica* (REZENDE et al., 2012; ETIENNE et al., 2012; CARVALHO et al., 2013). The use of nodal segments taken from the main orthotropic stem to prepare micro-cuttings was demonstrated by Vos and Snijder (2000). A process similar to this last, the use of successive cycles of production and rooting of mini-cuttings obtained in the first cycle from *in vitro* cloned coffee plants, with no use of plant growth regulators. was recently taken to the commercial scale, and proved to be very effective in reducing the costs of the *in vitro* cloning process (GEORGET et al., 2017).

Nevertheless, branching can be useful as a preparation to collect vegetative organs (SANSBERRO; MROGINSKI; BOTTINI, 2006). In trees, the shoot apex exerts apical dominance by inhibiting the outgrowth of the axillary sub-apical buds (MULLER; LEYSER, 2011) and, for arabica coffee plants, only orthotropic stems, which are normally under strong apical dominance, can be used to prepare cuttings. However, coffee seedlings grown from the seeds, decapitated and treated with auxin antagonists were able to produce additional orthotropic branches (CARVALHO et al., 2007).

The aims of this work were to test different concentrations of an inhibitor of auxin translocation (triiodobenzoic acid - TIBA) and its association with a cytokinin (benzylaminopurine - BAP) to produce sprouts, which are sources of micro-cuttings for C. arabica plants cloned in vitro. Two different experimental environments the greenhouse and the nursery - were tested as well. As far as we know, this is the first report on the post-acclimatization application of plant growth regulators as an aid to the vegetative propagation of arabica coffee varieties cloned in vitro. These procedures may contribute to bring down the costs for producing C. arabica plantlets in vitro, promoting their introduction in the coffee market processes.

#### 2 MATERIAL AND METHODS

## Plant material

C. arabica L. varieties Catucaí (567) and Siriema (3) were cloned in vitro through somatic embryogenesis (REZENDE et al., 2012). These varieties are very productive and Siriema is resistant to the leaf rust caused by Hemileia vastatrix Berk. & Broome (MATIELLO et al., 2016). Clones were rooted and hardened in vitro, before the transference to a compost for horticulture made from processed coconut husks. The compost was added with slow release fertilizer granules (NPK 14-14-14) and potted in 300 cm³ plastic conical containers placed in trays. Acclimatization took place in the greenhouse, under controlled fogging, temperature and humidity maintained around 27 °C and 80%, respectively.

### **Experimentation**

From the end of the third month of acclimatization and ahead, the *in vitro* cloned plants were subjected to a series of experiments to stimulate sprouting.

## **Experiment I**

256 in vitro cloned plants, organized in four replicas of eight plants per variety and treatment were maintained in the greenhouse and subjected to successive pulses of sprouting induction. PULSE 1: at the 3<sup>rd</sup> month of acclimatization, decapitation only (control) and decapitation and application of 2,3,5-triiodobenzoic acid (TIBA) as ethanolic solutions in the concentrations of 200. 400 and 600 mg.mL<sup>-1</sup>, sprayed on the leaves of the in vitro cloned plants. Apical sprouts (those coming out of the buds in the most apical node resting in each cloned plant) induced by these treatments were counted, measured and harvested two months after the stimulation pulse and again (spontaneous apical sprouts) five months after the pulse. Sub-apical sprouts (those coming out of buds in any node except the most apical one) were harvested five months after the pulse. PULSE 2: the same as pulse 1, applied to the same plants at the 8th month of acclimatization, with a single sprouts harvest made three months later. PULSE 3: at the 13th month of acclimatization, decapitation only and decapitation and application of 200, 600 and 1000 mg.mL<sup>-1</sup> TIBA or 1000 mg.mL<sup>-1</sup> TIBA plus 60 mg.mL<sup>-1</sup> benzylaminopurine (BAP) as ethanolic solutions sprayed on the leaves of the in vitro cloned plants. Sprouts harvest took place six months later.

## **Experiment II**

128 *in vitro* cloned plants of Catucaí, organized in four replications of eight plants per treatment were transferred from the greenhouse to the nursery (50% shadowing and automated irrigation), on the 5<sup>th</sup> month of acclimatization. In the 8<sup>th</sup> month, these plants were subjected to the same procedures for sprouting stimulation and sprouts harvests described for Experiment I PULSE 2.

After counting and measuring, apical and sub-apical orthotropic sprouts were collected, divided in nodal segments of length equal or above 1 cm, and the leaves in each node were cut by their halves, to prepare the micro-cuttings. Micro-cuttings were planted in multi-cellular plastic trays (50 cells with capacity to hold 90 cm<sup>3</sup> each) filled

with compost for horticulture added of fertilizers, as aforementioned for the acclimatization of the primary clones, and maintained in the greenhouse. Three months after planting, micro-cuttings survival, rooting, stem elongation and production of leaves were evaluated.

#### **Statistics**

The effect of a same concentration of TIBA on the number and length of apical and subapical sprouts produced following the different PULSES of induction were evaluated by linear regression analyses.

PULSES 1 to 3 in Experiment I were compared using multiple comparison procedures and Holm-Sidak's methods to identify the sources of variation influencing the number of microcuttings produced by the same set of *in vitro* cloned plants while subjected to the three different pulses of sprouting induction. The mean values of the different treatments and the control, regardless the PULSE, were compared by analysis of variance and the Tukey's test. Tests between treatments were performed using Student's tests (paired t tests) when necessary.

In vitro cloned plants of Catucaí subjected to PULSE 2 in Experiment I were compared to in vitro cloned plants of the same variety and cloning batch transferred to the nursery for Experiment II through linear regressions and rank sum paired tests of variance.

Overall, the variables analyzed were the number of apical and sub-apical sprouts, the length and the number of nodes in apical and subapical induced sprouts, micro-cuttings produced per in vitro cloned plant using nodal segments of apical and sub-apical sprouts, and the percentage of surviving micro-cuttings that developed roots and retrieved aerial part expansion 90 days after planted. The factors analyzed were the three different PULSES of growth regulators used to stimulate sprouting, the environment (greenhouse x nursery) where sprouting stimulation took place. the dosage of plant growth regulators and the genotype of the in vitro cloned plants (Catucaí x Siriema). Statistical analyses were performed using SigmaPlot (version 11.2).

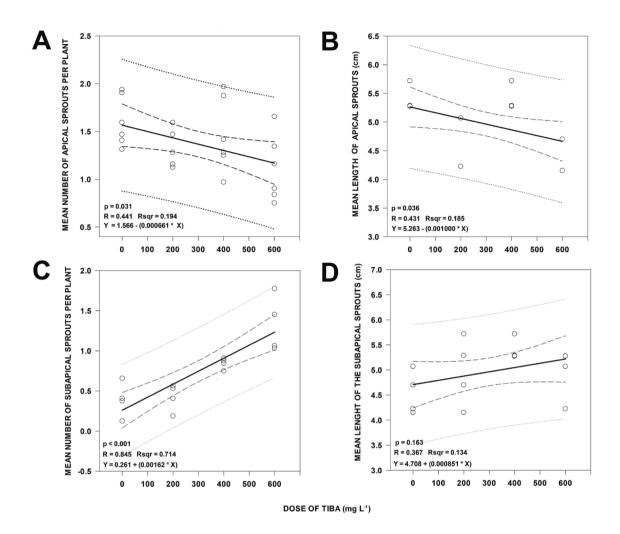
### **3 RESULTS AND DISCUSSION**

A trend to the inhibition of apical sprouting and the stimulation of sub-apical sprouting correlated to the increase in TIBA concentration was found for all the PULSES of

plant growth regulators throughout Experiment I. Invariably, increases in the concentration of TIBA were significantly correlated to the inhibition in number and length of apical sprouts (Figures 1A and 1B). Nevertheless, sprouts were, at least one of them and frequently two or even more than two in control plants and plants treated with the lower concentrations of TIBA, invariably observed coming out of the buds in the most distal nodes of the stems, which had been promoted to apical nodes by decapitation. On the other hand, the occurrence of orthotropic sprouts in nodes below the most apical one was extremely rare in the absence of TIBA. The number and the length of these sub-apical sprouts was improved by the increase in TIBA concentration (Figures 1C and 1D).

TIBA is an inhibitor of auxin translocation and when applied to the distal half of the shoots it reduced the ability of the distal buds to establish dominance in apple (COOK; VERHAEGEN; KEULEMANS, 2000). TIBA can enhance IPT expression by inhibiting auxin transport along the stem (TANAKA et al., 2006). IPT genes code for adenosine phosphate-isopentenyltransferases, which are key enzymes in the cytokinin biosynthesis pathway. By reducing auxin transport and consequently apical dominance, while contributing to enhance the synthesis of cytokinins, TIBA induced axillary buds to develop. And this explains the trends to inhibit apical buds reorganization that was observed. For a while, the sprouting of the axillary buds became favored. Finally, the re-establishment of apical dominance, exerted by new formed apical orthotropic branches, took place. In control plants, the reorganization of newly formed apical meristems was not impaired, apical dominance was more promptly re-established and sub-apical sprouting was inhibited.

To identify the factors influencing the production of micro-cuttings throughout Experiment I, the results obtained for each pulse were compared. The treatments were the only source of variation identified ( $F_{TREATS} = 8.041$ ;  $p_{TREATS} = 0.006$ ) and the subjects (the *in vitro* cloned plants) were not identified as a source of variation that could influence the results by the analysis of repeated measures. Indeed, PULSE 3 was effective (F = 8.445; p = 0.002) in reason of the treatments applied to the plants (Figure 2A). The highest concentrations of TIBA and mainly the association of TIBA and BAP applied to the in vitro cloned plants were essential to attain the best results of Experiment I (Figure 2B).

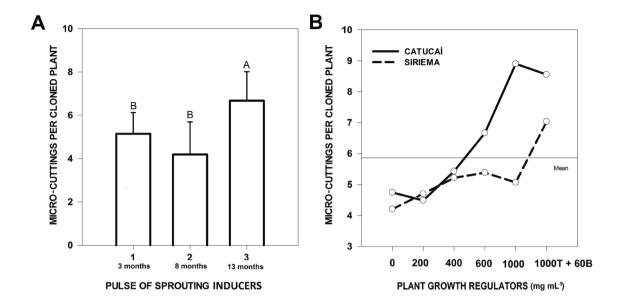


**FIGURE 1** - Number and length of sprouts induced on *in vitro* cloned coffee plants by the use of the auxin translocation inhibitor TIBA, in the greenhouse. Means represent plants of Catucaí and Siriema induced to sprout at the third and at the fifth months, all together (PULSES 1 and 2 of sprouting induction).

The analyses of the results obtained for all the *in vitro* cloned plants regardless of genotypes, supports what is stated above. Micro-cuttings yield observed for TIBA + BAP was significantly (t = 2.904; p = 0.043) higher than the yields observed for control plants of all the PULSES in Experiment I. The association TIBA + BAP did not differ from the highest concentration of TIBA but it promoted higher (t = 2.635; p = 0.039) micro-cutting yields than 600 mg.mL<sup>-1</sup> TIBA applied in the PULSES in Experiment 1.

The statistical difference between genotypes regardless the PULSE could not be detected, certainly in reason of the variability represented by the opposite results of plants of different genotypes to a same concentration

of growth regulator (Figure 2B). Nevertheless, analyzing the results for each genotype independently, it was possible to determine that the micro-cuttings yields of Catucaí in vitro cloned plants treated with 1000 TIBA or 1000 TIBA + 60 BAP were not statistically different. On the contrary, for Siriema in vitro cloned plants, 1000 TIBA or 1000 TIBA + 60 BAP were statistically different (t = 3.517; p  $\leq$  0.001; df = 89) and the higher micro-cutting yield was observed under the association TIBA + BAP (Figure 2B). Regardless genotypes, the association of TIBA + BAP induced a higher number of sub-apical sprouts than 1000 mg.mL<sup>-1</sup> TIBA (t = 5.536;  $p \le 0.001$ ; df = 87) or 600 mg.mL<sup>-1</sup> TIBA (t = 4.709; p  $\leq$  0.001; df = 114).



**FIGURE 2** - Number of micro-cuttings produced with the sprouts induced on *in vitro* cloned coffee plants following decapitation and the use of plant growth regulators, in the greenhouse. **A** - means represented are for micro-cuttings produced by cloned plants of Catucaí and Siriema all together induced to sprout by decapitation and application of three consecutive PULSES (1, 2 and 3) of plant growth regulators. **B** - means represented are for micro-cuttings produced by cloned plants of Catucaí and Siriema decapitated and subjected to the application of different concentration of TIBA and the combination of TIBA (1000T) and BAP (60B) in order to induce sprouting.

BAP, an exogenous cytokinin, contributed to the increase in the concentration of this class of growth regulators in the stem, eventually promoting their absorption by the sub-apical axillary buds. Inside the buds, cytokinins activate the cell cycle and promote cell multiplication (MÜLLER; LEYSER, 2011) for buds to growth. The higher number of sub-apical sprouts contributed for the higher yield of micro-cuttings in PULSE 3 when compared to PULSES 1 and 2. Induction using the highest concentration of TIBA and its association with BAP granted the production of 8.5 and 7.0 micro-cuttings per in vitro cloned plant of Catucaí and Siriema, respectively, in six month. These results can be related to differences between the genotypes: Siriema received contributions from Coffea racemosa Lour., a species described "profusely branching" (SURESHKUMAR; MOHANAN, PRAKASH; 2010). concentrations of regulators could work better for this last variety.

PULSES 1 and 2 together produced around 10 micro-cuttings per *in vitro* cloned plant treated with 600 mg.mL<sup>-1</sup> of TIBA in a year. Harvesting apical sprouts each three months would produce

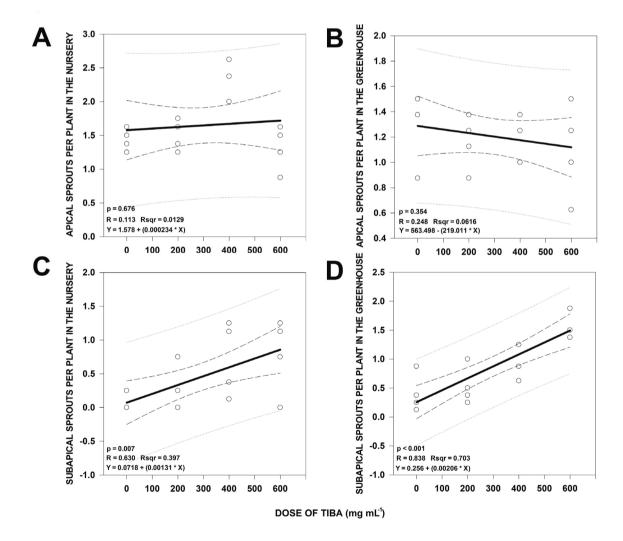
the same 10 to 12 micro-cuttings per decapitated plant in a year with no need to induce sprouting by the application of plant growth regulators. Nevertheless, two pulses of 1000 TIBA + 60 BAP applied to the same *in vitro* cloned plants and two sprouts harvests would result, in average, in 15 micro-cuttings produced per *in vitro* cloned plant each year. If each one of these micro-cuttings, once rooted and already growing, had been subjected to the procedures to induce sprouting or even to the dissection of their main orthotropic stems in nodal segments to produce mini-cuttings with no use of plant growth regulators (GEORGET et al., 2017) clonal amplification in the greenhouse would be exponential.

Experiment II was planned to evaluate the influence of the environment where sprouting induction was accomplished on the production of micro-cuttings. *In vitro* cloned plants of Catucaí at the 8<sup>th</sup> month of acclimatization were tested for the micro-cuttings production in the greenhouse (PULSE 2) and the nursery. Besides being grown in those two different environments, part of the plants under controlled temperature and humidity and maintained under approximately 70% shadowing

in the greenhouse and the other part growing in an environmen deprived of those controls, under approximately 50% shadowing in the nursery, another difference between the two sets of clones was that the plants in the greenhouse had already been subjected to PULSE 1 of sprouting induction and two sprouts harvests during the previous eight months.

As mentioned above, in the greenhouse, the higher the concentration of TIBA the lower the number of apical sprouts (Figures 1A) and, consequently, micro-cuttings prepared with nodal segments taken from apical sprouts produced in the greenhouse following PULSES 1 and 2. However, the correlation between these two factors was not

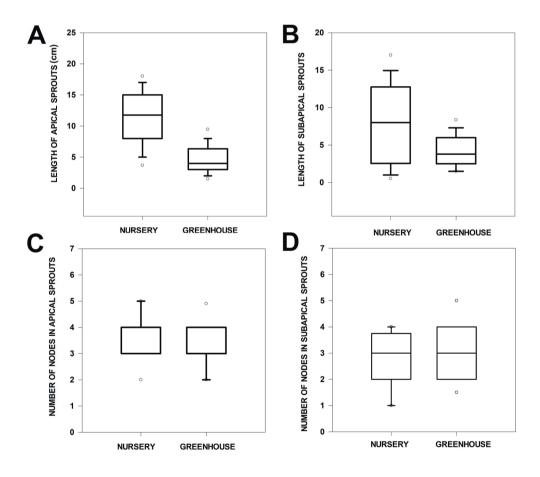
maintained in the nursery (Figures 3A). On the contrary, the trend observed, in the greenhouse, for results coming from three months old plus eight months old plants (PULSES 1 and 2, Figure 1A) to the inhibition of apical sprouting following TIBA application was confirmed for eight months old plants solely (PULSE 2), when cultivated in the greenhouse, despite the reduction in strength and lack of statistical significance (Figure 3B). Regarding sub-apical sprouts, on the other hand, the positive correlation between the concentration of TIBA and the number of sub-apical sprouts for the eight months old plants solely was observed to be significant and considerable in both environments (Figures 3C and 3D).



**FIGURE 3** - Mean number of sprouts induced on *in vitro* cloned Catucaí coffee plants by the use of the auxin translocation inhibitor TIBA at the eighth month of acclimatization. **A** and **C** – induction of sprouts in the nursery. **B** and **D** – induction of sprouts in the greenhouse (part of PULSE 2, Experiment I).

In addition, the length of induced apical (p  $\leq 0.001$ ) and sub-apical (p  $\leq 0.001$ ) sprouts was expressively higher in the nursery (Figures 4A and 4B), and at a first sight nursery could have been inappropriately declared as the ideal place to induce sprouting. Nevertheless, the number of nodes in apical sprouts was just slightly higher  $(p \le 0.001)$  in the nursery (Figure 4C), and the numbers of micro-cuttings produced by in vitro cloned Catucaí plants did not differ significantly (p = 0.079) in the nursery and the greenhouse, for the eight months old plants, because numbers of micro-cuttings are related principally to the numbers of nodes in the sprouts, which reached 7.41 and 6.55, respectively, regardless of the concentration of TIBA applied. In the nursery, longer internodes were produced and apical sprouting was not inhibted as harder as it was in the greenhouse by the application of TIBA, but these reactions were not sufficient to lead to the production of a higher number of micro-cuttings when compared to the greenhouse, where a higher number of sub-apical sprouts (Figure 3C x 3D) displaying shorter internodes (Figure 4B) were produced under the influence of TIBA. Regardless of environments or genotypes, the increase in TIBA concentration was significantly correlated to the increase in the number of micro-cuttings produced by linear regression analysis (p = 0.003, R = 0.418 and  $R^2 = 0.174$ ) throughout Experiment II, in agreement to Experiment I. This positive correlation was undoubtedly a consequence of the induction of sub-apical sprouting. In the nursery, the plants probably produced more auxins, and TIBA provided in the same concentration was not sufficient to inhibit translocation as well as in the greenhouse.

It still needs experimentation, but considering that the stems lignification in the greenhouse was clearly slower, and the diameter of the main orthotropic stem of *in vitro* cloned plants growing in the greenhouse had never reached that



**FIGURE 4** - Mean length and number of sprouts induced on *in vitro* cloned Catucaí coffee plants by the use of the auxin translocation inhibitor TIBA on the eighth month of acclimatization, in the nursery and the greenhouse. **A** and **B** - length of apical and subapical sprouts. **C** and **D** - number of nodes in apical and subapical sprouts.

of in vitro cloned plants, at the same age, while growing in the nursery, the induction of sprouts and rooting of micro-cuttings in short time lapses, induced by successive pulses of plant growth regulators, applied each three or six months. would probably be better in the greenhouse. An additional experiment (results not shown) aiming to induce the five or six basal nodes to sprout. following the harvest of the upper ones, did not succeed. The basal nodes of the in vitro cloned plants lost leaves as the time went by under the harder conditions of the nursery, exposed to variations in the temperature and to diseases. In these reasons sprouting for the basal nodes was not comparable with that of their counterparts in the greenhouse that produced more micro-cuttings after PULSE 3 than after PULSE 1 of inducers (Figure 2B). These last results were in part due to the maintenance of sprouts growing for six months on the in vitro cloned plants following PULSE 3 of plant growth regulators instead of harvesting sprouts in the third month after the stimulation, as done following PULSE 2. The association of TIBA and BAP and the use of a higher concentration of TIBA was advantageous to reach PULSE 3 results, as well. Nevertheless, the good physiological status of the plants, preserved from diseases and stresses in the greenhouse, which made possible their fast reactions to the sprouting inducers, even 13 months after potted, was also important.

Regarding rooting, retrieving of stem elongation and production of leaves, there was no statistical difference among micro-cuttings just above 1 and up to 3 cm in length and those lengthy, prepared with stem segments holding two nodes and four leaves. On the contrary, micro-cuttings below 1 cm in length rooted 11% less frequently and retrieved the growth of aerial parts 30% less frequently than those above 1 cm (p < 0.001). Micro-cuttings of Catucaí rooted 80% and the surviving ones developed new nodes and leaves in 90 days. Micro-cuttings of Siriema that produced new nodes and leaves in 90 days were 90% and all of them rooted.

### **4 CONCLUSIONS**

In conclusion, the most remarkable effect of the treatments with TIBA was the induction of sub-apical orthotropic sprouts, which contributed importantly to the production of micro-cuttings. Regarding the ideal environment to induce sprouting, the number of nodes and nodal segments of the sprouts were not higher in the nursery. In addition, the possibilities to control the environment in the greenhouse would favor the

micro-cuttings production through the application of successive pulses of sprouting inducers. There was no important difference between genotypes, and micro-cuttings above 1 cm rooted and formed new aerial organs 90 days after planting.

#### 5 ACKNOWLEDGMENTS

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# DETERMINATION OF THERMAL PROPERTIES OF COFFEE BEANS AT DIFFERENT DEGREES OF ROASTING

Danilo Barbosa Cardoso<sup>1</sup>, Ednilton Tavares de Andrade<sup>2</sup>, Renso Alfredo Áragón Calderón<sup>3</sup>, Mariane Helena Sanches Rabelo<sup>4</sup>, Camila de Almeida Dias<sup>5</sup>, Isabela Ávila Lemos<sup>6</sup>

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ABSTRACT: The aim in this study was to determine the main thermal properties of the granular mass of coffee (specific heat, thermal conductivity, and thermal diffusivity) for different degrees of roasting, as well as to model and simulate thermal conductivity at different degrees of roasting. For determination of specific heat, the mixing method was used, and for thermal conductivity, the theoretically infinite cylinder method with a central heating source. Thermal diffusivity was simulated algebraically using the results of the properties cited above and of the apparent specific mass of the product. Thermal conductivity was also simulated and optimized through finite element analysis software. As results, at darker roasting there was an increase in specific heat and a reduction in thermal conductivity and thermal diffusivity. Comparing thermal conductivity determined in relation to simulated and optimized conductivity, the mean relative error was 1.02%, on average.

Index terms: Specific heat, thermal conductivity, thermal diffusivity.

# DETERMINAÇÃO DAS PROPRIEDADES TÉRMICAS DE GRÃOS DE CAFÉ EM DIFERENTES PONTOS DE TORRA

RESUMO: Este trabalho teve como objetivo determinar as principais propriedades térmicas da massa granular de café (calor específico, condutividade e difusividade térmica), para diferentes pontos de torra bem como modelar e simular a condutividade térmica em diferentes graus de torração. Para a determinação do calor específico utilizou-se o método das misturas e para a condutividade térmica o método do cilindro teoricamente infinito com fonte de aquecimento central. A difusividade térmica foi simulada algebricamente utilizando-se os resultados das propriedades citadas anteriormente e da massa específica aparente do produto. Condutividade térmica foi, também, simulada e otimizada através do software de análise por elementos finitos. Como resultado foi observado que em torras mais escuras houve um aumento do calor específico e uma diminuição de condutividade e difusividade térmica. Através da comparação entre a condutividade térmica determinada e a condutividade simulada e otimizada verificou-se que o erro médio relativo, em média, foi de 1,02%.

Termos para indexação: Calor específico, difusividade térmica, condutividade térmica.

## 1 INTRODUCTION

Brazil is leader in the world market for production and export of green coffee beans. In postharvest, these green coffee beans pass through some steps until reaching at a very important phase of processing, the roasting. According to the International Coffee Organization - ICO (2017), the world consumption of coffee was 9368 million of Kg in 2017. The roasting is a highly important step in coffee processing because it is responsible for expressive modification of the raw material. During roasting, the coffee bean dehydrates and goes through physical-chemical transformations that provide the final product with the characteristics offered for its consumption (color, aroma, and flavor) through the formation of various volatile compounds (BOTTAZZI et al., 2012; HERNÁNDEZ; HEYD; TRYSTRAM, 2008).

Throughout production process, coffee beans are subjected to changes in temperature and moisture, and to ensure a quality product in the end of process, it is necessary to know how these changes occur. In this sense, the knowing properties such as specific heat, thermal conductivity, and thermal diffusivity are relevant for studies of heat and mass transfer in agricultural seed grains (BORÉM et al., 2002).

The specific heat, by definition, is the amount of heat necessary to raise the temperature of a body by 1°C per unit of mass without change in state. It is essential to know this for determination of the amount of energy required for heating or cooling a food product. The thermal conductivity of a material is the measure of its conduct heat capacity (MOHSENIN, 1980).

According to Mohsenin (1980), numerical values of thermal conductivity of solid, granular, and porous materials can to vary according to chemical composition, fluid material content,

<sup>1.2.3.4.5.6</sup> Universidade Federal de Lavras/UFLA - Departamento de Engenharia/DEG - Cx. P. 3037 - 37.200-000 - Lavras - MG cardosodb@gmail.com, edniltontavares@gmail.com, rensoa.aragonc@gmail.com, marianerabelo1@hotmail.com, camila.almeidadias@gmail.com, isa lemosti@hotmail.com

physical structure, state, density, temperature, and moisture content. According to the same author, for biological materials, thermal conductivity depends more on cell structure, density, and moisture content than on temperature.

For heat to be diffused in the grain mass, for example in a mass of roasted coffee beans, there is dependence on the thermal properties of the product, among which is thermal diffusivity, and on the temperature gradient within the body and between the surface and the environment.

Thus, the aim in this study was to determine variations in specific heat, thermal conductivity, and thermal diffusivity of coffee beans at different degrees of roasting, analyze the correlation of these thermal properties with the lightness colorimetric parameter (L\*) and apparent specific mass, and simulate and optimize thermal conductivity through finite element analysis software.

#### 2 MATERIALS AND METHODS

This study was developed at the Agricultural Product Processing Laboratory (LPPA) of the Universidade Federal de Lavras (UFLA) in Lavras, MG, Brazil.

Coffee (*Coffea arabica* L.) beans were used, with an average moisture content of 0.123 dry basis (10.94% wet basis), which was determined by the laboratory oven method at 105±1°C for 16±0.5 hours, according to the standard method of ISO 6673 (INTERNATIONAL ORGANIZATION FOR STANDARDIZATION - ISO, 1999). The coffee was hulled and the beans were separated according to shape and size. Only the conventional flat coffee beans from sieve size 16 to 18/64 inches were used for roasting, eliminating the flat beans retained in the 19/64 inch sieve and the peaberries retained in the sieve with an oblong screen of 11 x <sup>3</sup>/<sub>4</sub> inch. The beans were then roasted to 5 different levels in an Atilla 5 Kg Gold Plus coffee roaster.

A caliper rule was used for determination of the dimensions of 40 coffee beans from each treatment; and coffee bean volume was calculated through equation (1), approximating the coffee bean shape to the shape of a semi-ellipsoid (BUSTOS-VANEGAS et al., 2018)

$$V = \frac{\pi abc}{6} \tag{1}$$

in which V is volume in mm<sup>3</sup>; a is bean length in mm; b is bean width in mm; and c is bean thickness in mm.

The apparent specific mass of the coffee beans was analyzed using a GEHAKA brand kit, following manufacturer's instructions for determination of the hectoliter weight of coffee beans (BOTELHO et al., 2016). The test was performed in four replications for each roasting degree and for the green coffee bean. The results were expressed in kg.m<sup>-3</sup>.

The color of the roasted coffee was determined using the colorimeter Konica Minolta CR-300. This equipment was used in the configuration of D65 illuminant and calibrated with the white plate with a value determined corresponding to L\*, a\*, b\*. The color of the beans was expressed in parameters of the system CIE L\*, a\*, b\* and also of the system CIE L\* c\* h°. The L\* coordinate indicates lightness, which ranges from 0 (black) to 100 (white). The other parameters, a\* and b\* (chromaticity coordinates), indicate the colors directions of the roasted beans, in which +a\* indicates red color, -a\* green, +b\* yellow, and -b\* blue. Three readings were made for composition of the mean value.

The cylindrical coordinates c\* or chroma, which provides a measurement of intensity or saturation of the color, and h°, which corresponds to the hue angle, were simulated from equations (2) and (3), respectively.

$$c^* = \sqrt{a^{*2} + b^{*2}} \tag{2}$$

$$h^{\circ} = \tan^{-1} \left( \frac{b^*}{a^*} \right) \tag{3}$$

The specific heat of the coffee beans for the different roasting degrees and for the green coffee bean was determined by the mixing method. In this method, the product with known mass (100 g) and temperature (25°C) was placed in a calorimeter (Figure 1) of known thermal capacity (0.048 kJ.°C<sup>-1</sup>), which contains water at a temperature (40°C) and mass (400 g) that are also known. When the thermal equilibrium of the mixture is reached, the specific heat of the product can be simulated, using the equation (4) (MOHSENIN, 1980).

$$C_p \cdot M_p \cdot (T_e - T_p) = C_w \cdot M_w \cdot (T_w - T_e) + C \cdot (T_w - T_e)$$
 (4)

in which  $C_p$  is the specific heat of the product in  $kJ.kg^{-1}.^{\circ}C^{-1}$ ;  $C_w$  is the specific heat of the water in  $kJ.kg^{-1}.^{\circ}C^{-1}$ ; C is the heat capacity of the calorimeter in  $kJ.^{\circ}C^{-1}$ ;  $M_p$  is mass of the product in kg;  $M_w$  is mass of water in kg;  $T_p$  is temperature of the product in C;  $T_w$  is temperature of water in C;  $T_w$  is temperature of equilibrium in C.

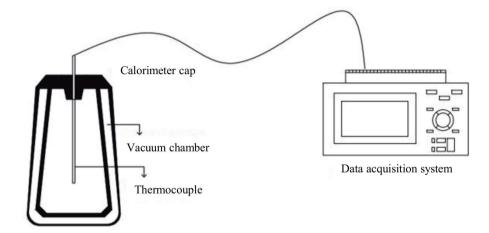


FIGURE 1 - Schematic drawing of the experimental equipment for determination of specific heat.

For the thermal conductivity determination and analysis of transient heat flow through the product granular mass by the theoretically infinite cylinder method (Figure 2), the equation (5) was used to describe heat flow in a linear source (MOHSENIN, 1980).

$$\frac{\partial \mathbf{T}}{\partial \mathbf{t}} = \alpha \left[ \frac{\partial^2 \mathbf{T}}{\partial \mathbf{r}^2} + \frac{1}{\mathbf{r}} \frac{\partial \mathbf{T}}{\partial \mathbf{r}} \right] \tag{5}$$

in which T is temperature in °C; t is the sample heating time in s;  $\alpha$  is thermal diffusivity in m<sup>2</sup>.s<sup>-1</sup>; and r is radial distance of the heat source in m. This method consists of use of an aluminum cylinder with pre-determined diameter and length, with a nickel-chrome varnished conductor wire in the center, through which a low intensity electric current flowed (1 A and 1.8 V). Temperature was obtained by means of three sets equidistant thermocouples (120°) placed at mid-height of the cylinder. Each set was composed of 6 thermocouples at a distance of 1 cm from each other along the radius of the center of the tube (Figure 2). The coffee beans thermal conductivity was obtained on transient regime by cylindrical coordinates through the equation (6).

$$k = \frac{Q}{4 \cdot \pi \cdot (T_2 - T_1)} \cdot \ln \left( \frac{t_2 - t_0}{t_1 - t_0} \right)$$
 (6)

in which k is thermal conductivity in W.m<sup>-1</sup>.°C<sup>-1</sup>; Q is heat provided to the conductor wire in W; t is time in s;  $T_{(t)}$  is temperature at time t in °C; and  $t_0$  is the correction factor in s.

According to Chang (1986), the correction factor  $t_0$  can be calculated as a logarithm function of the values of time and of the differences among the temperatures observed over time and the initial temperature of the system.

The theoretically infinite cylinder is an idealization that allows adoption of the hypothesis of one-dimensional conduction in the radial direction, which is considered a reasonable approximation if the ratio between the length and the radius of the cylinder is greater than or equal to 10 (BERGMAN et al., 2011).

Thermal diffusivity of the granular mass of green and roasted coffee beans was determined using equation (7), after specific heat, thermal conductivity, and apparent specific mass were determined experimentally.

$$\alpha = \frac{k}{\rho \cdot C_p} \tag{7}$$

in which  $\alpha$  is thermal diffusivity in m<sup>2</sup>.s<sup>-1</sup>; and  $\rho$  is apparent specific mass in kg.m<sup>-3</sup>. The equation that governs transient transfer of heat in an infinite cylinder when considering (a) generation of heat coming from a central infinite source of zero diameter and constant wattage; (b) infinite and homogeneous medium; and (c) initial conditions of the medium that is isothermal and in equilibrium with the environment, can be expressed as (BERGMAN et al., 2011):

$$\frac{\partial \mathbf{T}}{\partial \mathbf{t}} = \alpha \left( \frac{\partial^2 \mathbf{T}}{\partial \mathbf{r}^2} + \frac{1}{\mathbf{r}} \frac{\partial \mathbf{T}}{\partial \mathbf{r}} \right) + \mathbf{q}$$
 (8)

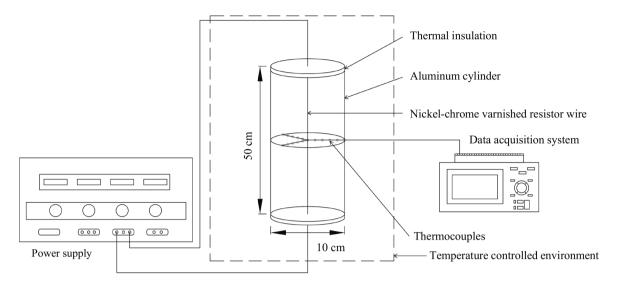


FIGURE 2 - Schematic drawing of the experimental device for determination of thermal conductivity.

in which T is temperature in  ${}^{\circ}$ C; t is time for heating the sample in s;  $\alpha$  is thermal diffusivity in  $m^2$  s<sup>-1</sup>; r is the radius of localization of the thermocouple in m; and q is power dissipated in the source in W m<sup>-1</sup> per unit of volumetric heat capacity ( $\rho$ C-J m<sup>-3</sup>  ${}^{\circ}$ C<sup>-1</sup>). The approximate solution of equation (8) was obtained using the finite element technique. Initially the system was divided into 2925 two-dimensional asymmetric elements of the PLANE77 type, for a total of 9196 nodes.

In the finite element technique, one of the procedures for analyzing equation (8) is by means of the weighted residual method (SEGERLIND, 1984). In this method, the volume integral for an element "e" is given by equation (9).

$$\left\{R^{(e)}\right\} = -\int_{\Lambda} \left[W^{\mathsf{T}}\right] \left(\frac{\partial}{\partial x} \left(k \frac{\partial \mathsf{T}}{\partial x}\right) + \frac{\partial}{\partial y} \left(k \frac{\partial \mathsf{T}}{\partial y}\right)\right) dA + \int_{\Lambda} \left[W^{\mathsf{T}}\right] \left(\rho \, c_{\mathfrak{p}} \frac{\partial \mathsf{T}}{\partial t}\right) dA \tag{9}$$

in which  $\{R^{(e)}\}$  is residual integral;  $[W]^T$  is weight function; k is thermal conductivity; A is area of the element; and dA is elemental area.

The equation (9) is evaluated with respect to the spatial coordinates for a fixed moment of time. Once integrated, equation (9) can be expressed in matrix form as:

$$\left[C_{e}^{t}\right]\left\{T_{e}\right\}+\left(\left[K_{e}^{tb}\right]+\left[K_{e}^{tc}\right]\right)\left\{T_{e}\right\}=\left\{Q_{e}^{c}\right\}+\left\{Q_{e}^{g}\right\}=\left\{F_{e}\right\}\tag{10}$$

in which  $\begin{bmatrix} C_e^t \end{bmatrix}$  is the thermal capacitance matrix;  $\begin{bmatrix} K_e^t \end{bmatrix}$  is the convection matrix on the element surface;  $\begin{bmatrix} Q_e^c \end{bmatrix}$  is the vector of heat flow on the element surface;

 $\{Q_e^g\}$  is heat generation; and  $\{F_e\}$  is the element force vector.

The equation (11) is the general (or global) form of the equation (10) that can be apply to the system set of elements. Thus, the solution of equation (9) produces a linear differential equations system of first order in time.

$$[C] \{T\} + [K] \{T\} = \{F\}$$

$$(11)$$

in which [C] is the global capacitance matrix; [K] is the global rigidity matrix; and {F} is the global force vector.

The discretization of the equation (10) in time can be performed with the aid of approximations by finite differences in the time domain (SEGERLIND, 1984). The result of discretization has the form:

$$([C] + \theta \Delta t[K])\{T\}_{n+1} = ([C] - (1-\theta)\Delta t[K])\{T\}_n + \Delta t((1-\theta)\{F\}_n + \theta\{F\}_{n+1})$$
(12)

in which  $\theta$  is the parameter of transient integration;  $\Delta t = t_{n+1} - t_n$  is the time interval = 2 h;  $\{T\}_n$  is the temperature at time  $t_n$ ;  $\{T\}_{n+1}$  is the temperature at time  $t_{n+1}$ ;  $\{F\}_n$  is the global force vector at time  $t_n$ ; and  $\{F\}_{n+1}$  is the global force vector at time  $t_{n+1}$ .

The equation (12) provides the nodal values of the temperatures at moment  $t_{n+1}$  as a function of the known values of the temperatures at moment  $t_n$  and of forces at moments  $t_n$  and  $t_{n+1}$  and the transient integration parameter  $\theta$ .

The nodal temperatures values at different times were obtained by solving the equations system represented by equation (12) (SEGERLIND, 1984). So, the computational program of finite element analysis ANSYS 14.2 was used.

For implementation of the finite element method, it is necessary to introduce parameters, some related to the product properties, which were determined experimentally, and others relevant to the initial conditions and boundary conditions imposed on the system (Figure 3).

### **3 RESULTS AND DISCUSSION**

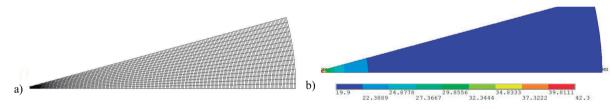
The mean values of color parameters obtained experimentally for green coffee beans that were used for roasting are present in Table 1.

These data reveal that it occurred in the darkening of the sample because the colorimetric parameter (L \*) was of 50.75 for green coffee and 23.85 for roasted coffee in mean.

The mean values obtained experimentally for apparent specific mass and length, width, and thickness of green coffee beans are presented in Table 2.

The apparent specific mass obtained in the present study for hulled coffee was 666.75 kg.m<sup>3</sup> for grains with moisture content of 0.123 dry basis (10.94% w.b.) (Table 2). This result is lower than that obtained by Oliveira et al. (2015), that found data for apparent specific mass from 751 to 758 kg.m<sup>3</sup> for moisture content of 11% (w.b.), but above that found by Giomo, Nakagawa and Gallo (2008), which on average found a value of 498.4 kg.m<sup>3</sup> for mean moisture content of 33.35% w.b, and also above that found by Olukunle and Akinnuli (2012), that found values from 588.2 to 609.8 kg.m<sup>3</sup> for moisture content of 10,7 % d.b.

The experimental values of the thermal properties determined for green coffee beans are expressed in Table 3.



**FIGURE 3** - (a) Boundary conditions applied from discretization of the system in elements; (b) Temperature distribution in the system with coffee beans.

TABLE 1 - Color parameters for green coffee beans used for roasting.

	Cie L a* b*			Cie L c* h°		
	L*	a*	b*	L*	c*	h°
Green coffee bean	50.75	2.07	19.84	50.75	19.94	84.05

**TABLE 2** - Mean dimensions of 40 coffee beans and apparent specific mass of green coffee beans.

Length (mm)	Width (mm)	Thickness (mm)	Specific mass (kg.m <sup>-3</sup> )
8.91	6.59	3.62	666.75

**TABLE 3** - Thermal properties for green coffee beans.

Thermal property	
Specific heat (kJ.kg <sup>-1</sup> .°C <sup>-1</sup> )	1.929
Thermal conductivity (W.m <sup>-1</sup> .°C <sup>-1</sup> )	0.182
Thermal diffusivity (m <sup>2</sup> .s <sup>-1</sup> ) x 10 <sup>-7</sup>	1.415

The values of specific heat and thermal conductivity for green coffee beans in the present study were 1.929 kJ.kg <sup>-1</sup>.°C <sup>-1</sup> and 0.182 W.m<sup>-1</sup>. °C-1, respectively. These values are near those obtained by Borém et al. (2002), which studied five varieties of pulped coffee and obtained specific heat between 1.00 and 3.00 kJ.kg<sup>-1</sup>.°C<sup>-1</sup>, and thermal conductivity between 0.10 and 0.20 W.m<sup>-1</sup>.°C<sup>-1</sup>, with moisture contents that ranged from 0.1 to 0.95 (d.b.), but different from values found by Fabbri et al. (2011), that found 1.256 kJ.kg<sup>-1</sup>.°C<sup>-1</sup> for specific heat and 0.131 W.m<sup>-1</sup>.°C<sup>-1</sup> for thermal conductivity. Burmester and Eggers (2010) found thermal conductivity values for semi-washed coffee from 0.20 to 0.34 W.m<sup>-1</sup>.°C<sup>-1</sup> <sup>1</sup>, determined through the stationary plate method (Eurotechnica GmbH, Bargteheide, Germany).

The diffusivity value of hulled green coffee in the present study was  $1.42 \times 10^{-7} \,\mathrm{m^2.s^{-1}}$ ; which is within the interval of values found by Borém et al. (2002), which was from  $1.0 \times 10^{-7}$  to  $3.0 \times 10^{-7} \,\mathrm{m^2.s^{-1}}$  for all the varieties of pulped coffee, and moisture contents from 0.1 to 0.95 (d.b.) but with the difference that, in the 2002 study, the coffee analyzed had parchment. Fabbri et al. (2011) used the equipment Thermal Analyzer KD2 (Decagon Device Inc., Pullman, USA) and found a thermal diffusivity value of  $0.9 \times 10^{-7} \,\mathrm{m^2.s^{-1}}$ . These differences of values found occur due to different varieties of coffee, product moisture contents, and methods for determination of these properties.

The mean experimental values of apparent specific mass as a function of the roasting degree, which is represented by the lightness (L\*) colorimetric parameter, are present in Figure 4. The results indicate that with a reduction in lightness, there is a reduction in specific mass, which can be explained through the occurrence of an increase in volume the darker the roast is, as present in Table 4.

The mean values obtained experimentally for apparent specific mass and the measurements of the coffee beans for length, width, and thickness of the coffee samples at different roasting degrees are present in Table 4. Degree 1 is considered the lightest roasting degree and degree 5 the darkest, as present in Figure 4.

The apparent specific mass values are in agreement with the values found by Bicho et al. (2012), which were 400 to 300 kg.m<sup>-3</sup>, and by Oliveira et al. (2014), which were 359.4 to 342.26 kg.m<sup>-3</sup>. They are also near the results obtained by Mendonça, Franca and Oliveira (2009) that were 465 to 349 kg.m<sup>-3</sup>, but lower than the values 530 to 370 kg.m<sup>-3</sup>, found by Jokanović et al. (2012).

The mean values of color parameters obtained experimentally for coffee beans at different degrees of roasting are present in Table 5.

Baggenstoss, Perren and Escher (2008) found values of the lightness colorimetric parameter (L\*) from 23.5 to 22.6, which are similar to those found in this study.

Considering that the second degree polynomial is a simple model and satisfactorily explains the variations in the thermal properties studied as a function of the roasting degree, the criterion of highest adjusted coefficient of determination (R<sup>2</sup>) was used as the choice for the equation to be adopted.

Figures 5, 6, and 7 exhibit the experimental values of specific heat, thermal conductivity, and thermal diffusivity for coffee beans roasted as a function of the lightness colorimetric parameter (L\*) and constitutes a scale between black and white, in which values near 20 indicate dark roasting and values near 30 indicate light roasting.

The results reveal that there was an increase in specific heat and a reduction in thermal conductivity and diffusivity with darker roasting,

The Figures 8, 9, and 10 present the experimental values of specific heat, thermal conductivity, and thermal diffusivity for coffee beans roasted as a function of apparent specific mass. These figures present that these properties of roasted coffee had a higher adjusted coefficient of determination (R<sup>2</sup>) compared to the figures related to the lightness colorimetric parameter (L\*). With reduction in specific mass from roasting, there was reduction in thermal conductivity and thermal diffusivity, but an increase in specific heat.

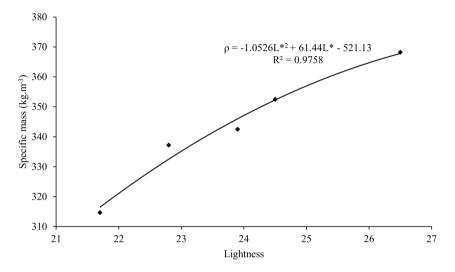
The Table 6 presents the physical properties of green and roasted coffee of the present study.

In the present study of roasted coffee, the values of specific heat ranged from 905.46 to 1245.08 J.kg<sup>-1</sup>.°C<sup>-1</sup>, values of thermal diffusivity ranged from 3.35x10<sup>-7</sup> to 2.45x10<sup>-7</sup> m<sup>2</sup>.s<sup>-1</sup>, and values of thermal conductivity ranged from 0.112 to 0.096 W.m<sup>-1</sup>.°C<sup>-1</sup>. Fabbri et al. (2011) found values of specific heat for roasted coffee from 1256 to 1648 J.kg<sup>-1</sup>.°C<sup>-1</sup>, values of thermal diffusivity from 0.09x10<sup>-7</sup> a 0.82x10<sup>-7</sup> m<sup>2</sup>.s<sup>-1</sup>, and values of thermal conductivity from 0.131 to 0.075 W.m<sup>-1</sup>.°C<sup>-1</sup>. Hammerschmidt and Abid (2016) found a value of specific heat of 1593 J.kg<sup>-1</sup>.°C<sup>-1</sup> for 0°C for roasted coffee. As observed in figures 5, 6, 7, 8, 9 and 10, there was an effect of the roast on all analyzed thermo-physical properties.

The Table 7 presents the data of the property thermal conductivity obtained experimentally and by computational simulation and optimization, and the mean relative error between the two methods studied.

For the suitability of certain models in

the description of a phenomenon, according to Mohapatra and Rao (2005), values lower than 10% mean relative error (P) indicate good fit for practical purposes. The error obtained was acceptable, confirming the results, and, thus, the model developed can be used in other applications.



**FIGURE 4** - Experimental values of coffee beans specific mass as a function of roasting degree (lightness colorimetric parameter).

TABLE 4 - Mean dimensions of 40 coffee beans and apparent specific mass at different roasting degrees

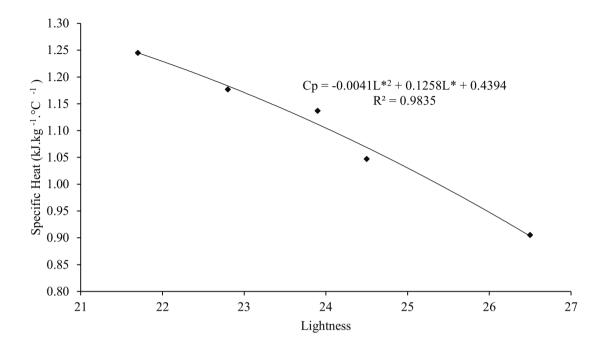
	Length (mm)	Width (mm)	Thickness (mm)	Volume (mm³)	Apparent specific mass (kg.m <sup>-3</sup> )
Degree 1	9.81 a	7.67 a	3.97 a	157.25 a	368.40 a
Degree 2	9.93 a	7.73 a	3.99 a	161.16 a	352.48 b
Degree 3	10.14 b	7.83 b	4.05 a	168.91 b	342.36 c
Degree 4	10.23 b	7.86 b	4.07 a	171.88 b	336.96 d
Degree 5	10.54 c	7.99 c	4.19 a	187.04 c	314.43 e
CV (%)	5.38	3.23	5.37	10.45	0.39

Mean values followed by the same letter in the column belong to the same cluster by the Scott-Knott test at 5% probability.

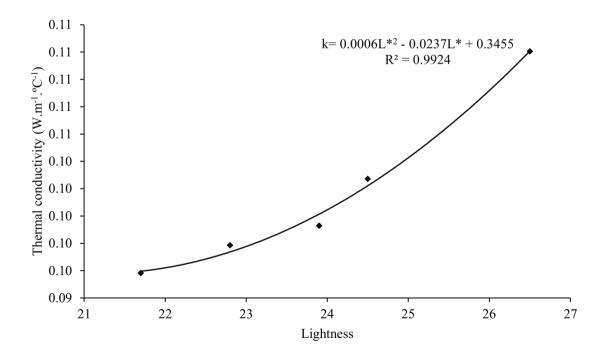
**TABLE 5** - Color parameters for different degrees of roasting.

		Cie L a* b*			Cie L c* h°		
		a*	b*	L*	c*	h°	
Degree 1	26.47	9.66	16.98	26.47	19.53	60.37	
Degree 2	24.46	8.65	17.30	24.46	19.34	63.44	
Degree 3	23.88	8.89	16.84	23.88	19.04	62.18	
Degree 4	22.76	7.91	13.22	22.76	15.41	59.12	
Degree 5	21.66	8.69	13.71	21.66	16.23	57.63	

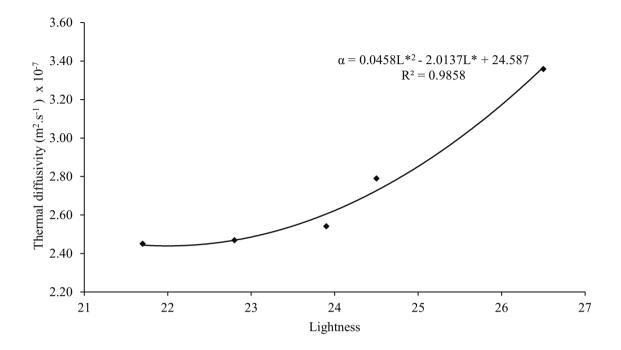
Coffee Science, Lavras, v. 13, n. 4, p. 498 - 509, oct./dec. 2018



**FIGURE 5** - Experimental values of specific heat of coffee beans as a function of roasting degree (lightness colorimetric parameter).



**FIGURE 6** - Experimental values of thermal conductivity of coffee beans as a function of the roasting degree (lightness colorimetric parameter).



**FIGURE 7** - Experimental values of thermal diffusivity of coffee beans as a function of degree of roasting (lightness colorimetric parameter).

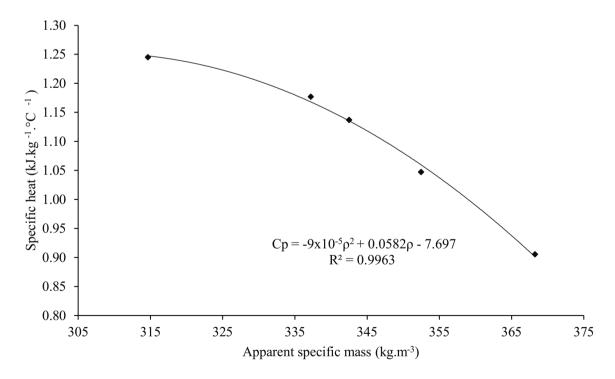


FIGURE 8 - Experimental values of specific heat of roasted coffee beans as a function of apparent specific mass.

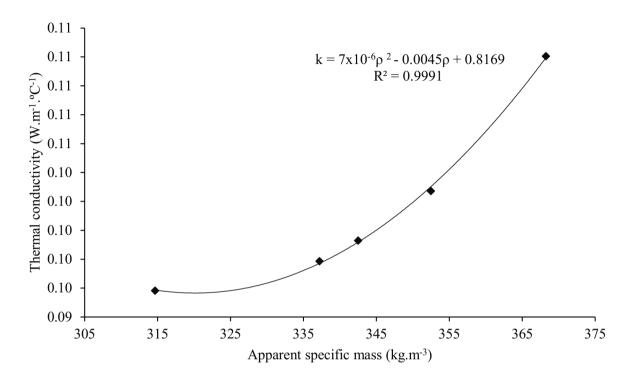


FIGURE 9 - Experimental values of thermal conductivity of roasted coffee beans as a function of apparent specific mass.

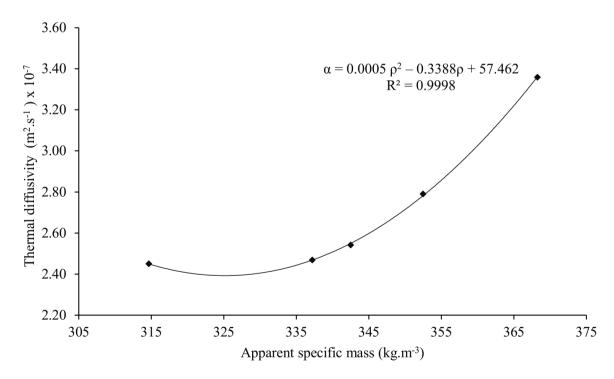


FIGURE 10 - Experimental values of thermal diffusivity of roasted coffee beans as a function of apparent specific mass.

Type of material	Specific mass (kg.m <sup>-3</sup> )	Specific heat (J.kg <sup>-1</sup> .°C <sup>-1</sup> )	Thermal diffusivity (x10 <sup>-7</sup> m <sup>2</sup> .s <sup>-1</sup> )	Thermal conductivity determined	Conductivity simulated and optimized
Green bean	666.75	1929.30	1.4148	0.182	0.18029
Degree 1	368.25	905.46	3.3590	0.112	0.11347
Degree 2	352.48	1047.27	2.7903	0.103	0.10643
Degree 3	342.50	1137.05	2.5421	0.099	0.09945
Degree 4	337.20	1177.00	2.4692	0.098	0.09883
Degree 5	314.65	1245.08	2.4505	0.096	0.09498

TABLE 6 - Physical properties of green and roasted coffee.

**TABLE 7** - Data of the property thermal conductivity obtained experimentally and by computational simulation and optimization.

Type of material	Thermal conductivity determined	Conductivity simulated and optimized	Mean relative error (%)
Green bean	0.182	0.1803	0.93
Degree 1	0.112	0.1135	1.30
Degree 2	0.103	0.1014	1.54
Degree 3	0.099	0.0995	0.45
Degree 4	0.098	0.0988	0.84
Degree 5	0.096	0.0950	1.08
		Mean	1.02

### **4 CONCLUSIONS**

According to the results obtained can be concluded that:

- 1. The thermal properties of roasted coffee bean samples has a correlation with the lightness colorimetric parameter (L\*) and apparent specific mass;
- 2. The specific heat of roasted coffee ranged from 905.46 to 1245.08 J.kg <sup>-1</sup>.°C <sup>-1</sup>.
- 3. The thermal conductivity of roasted coffee ranged from 0.112 to 0.096 W.m<sup>-1</sup>.°C<sup>-1</sup>.
- 4. The thermal diffusivity of the roasted coffee ranged from  $3.35 \times 10^{-7}$  m<sup>2</sup>.s<sup>-1</sup> to  $2.35 \times 10^{-7}$  m<sup>2</sup>.s<sup>-1</sup>.
- 5. The thermal properties of roasted coffee had greater adjusted coefficients of determination (R<sup>2</sup>) in relation to apparent specific mass compared to the lightness colorimetric parameter (L\*).
- 6. With darker roasting, there was an increase in specific heat and a reduction in thermal conductivity and diffusivity.

- 7. With reduction in specific mass after roasting, there was a reduction in thermal conductivity and thermal diffusivity, but an increase in specific heat.
- 8. Comparing the thermal conductivity that was determined in relation to that simulated and optimized by a computational program of finite element analysis, mean relative error was 1.02%, on average.

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# BIG COFFEE VL.: SEED DESICCATION TOLERANCE, SIEVE CLASSIFICATION, AND PHYSIOLOGICAL QUALITY

Ana Cristina de Souza<sup>1</sup>, Sttela Dellyzete Veiga Franco da Rosa<sup>2</sup>, Amanda Lima Vilela<sup>3</sup>, Madeleine Alves de Figueiredo<sup>4</sup>, Ana Luiza de Oliveira Vilela<sup>5</sup>, Ricardo Stéphano Filho<sup>6</sup>, Milene Alves de Figueiredo Carvalho<sup>7</sup>

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ABSTRACT: A coffee plant producing large fruit, seeds, and leaves in relation to conventional coffee plants, possibly generated by genetic mutation, was named Big Coffee VL. Seeds of this coffee plant were classified by size and used to establish the crop, whose progenies were designated as Big Coffee VL. large, Big Coffee VL. medium, and Big Coffee VL. small. The aim in this study was to investigate desiccation tolerance, together with size classification, and evaluate the physiological quality of the seeds of this progeny. Seeds of each type of Big Coffee VL. and of the cultivar Topázio were collected. High moisture seeds and seeds dried to 11% moisture content were evaluated to assess desiccation tolerance. Dried seeds of each Big Coffee VL. progeny and of the Topázio cultivar were classified by size in sieve testing using oblong screens for separation of peaberry seeds, and circular sieves from 22 to 12 for separation of flat seeds. All seeds were subjected to physiological evaluation through the germination test and determination of seedling dry matter. A completely randomized experimental design (CRD) was used; results were subjected to analysis of variance and means compared by the Scott-Knott test. Big Coffee VL seeds tolerate desiccation to moisture content of 11% wet basis. Seeds of the Topázio cultivar have better physiological performance than seeds of the Big Coffee VL progenies. The bigger the seeds of Big Coffee VL., the better their physiological performance, exhibiting greater seedling dry matter.

**Index terms:** Coffea arabica L, seed size, germination test, vigor, particle size analysis sieves.

# BIG COFFEE VL.: TOLERÂNCIA A DESSECAÇÃO DE SEMENTES, CLASSIFICAÇÃO EM PENEIRAS E QUALIDADE FISIOLÓGICA

RESUMO: Uma planta de café que produz grandes frutos, sementes e folhas em relação ao cafeeiro convencional, possivelmente gerada por mutação genética, foi denominada Big Coffee VL. As sementes deste cafeeiro foram classificadas por tamanho e usadas para estabelecer a cultura, cujas progênies foram designadas como Big Coffee VL. grande, Big Coffee VL. médio, e Big Coffee VL. pequeno. O objetivo neste estudo foi investigar a tolerância à dessecação, juntamente com a classificação por tamanho, e avaliar a qualidade fisiológica das sementes destas progênies. Sementes de cada tipo de Big Coffee VL. e da cultivar Topázio foram coletadas. Sementes com alta umidade e sementes secas a 11% de umidade foram utilizadas para avaliar a tolerância à dessecação. Sementes secas de cada progênie Big Coffee VL e a cultivar Topázio foram classificadas por tamanho, jogo de peneiras, utilizando telas oblongas para separação de sementes tipo moca e peneiras circulares de 22 a 12 para separação de sementes chatas. Todas as sementes foram submetidas à avaliação fisiológica por meio do teste de germinação e determinação da matéria seca das plântulas. Um delineamento experimental foi inteiramente casualizado (DIC) foi utilizado; os resultados foram submetidos à análise de variância e as médias comparadas pelo teste de Scott-Knott. Sementes de Big Coffee VL. toleram dessecação para um teor de umidade de 11% base úmida. Sementes da cultivar Topázio apresentam melhor desempenho fisiológico que as sementes das progênies de Big Coffee VL. Quanto maiores as sementes de Big Coffee VL., melhor seu desempenho fisiológico, exibindo maior massa seca de plântulas.

**Termos para indexação:** *Coffea arabica* L, tamanho da semente, teste de germinação, vigor, peneiras de análise granulométrica.

## 1 INTRODUCTION

Coffee is one of the most important agricultural commodities exported and one of the beverages most consumed in the world (CARDOSO et al. 2016). It is an extremely important product in Brazil as an exported product, a source of income, a source of employment, and a channel of manual labor in the rural area

(SANTOS et al., 2009). Coffee fields in Brazil were formed from the arabica cultivar (*Coffea arabica* L. var. typica), the first to be introduced in the country. Little by little, coffee fields began to diversify through introduction of new cultivars (MARTINS et al., 1992).

As the crop is so important for Brazil, a concern is the low tolerance of coffee seeds to desiccation and to storage; they are classified as

<sup>1.3,4,5,6</sup> Universidade Federal de Lavras/UFLA - Departamento de Agricultura/DAG - Cx. P. 3037 - 37.200-000 - Lavras - MG acstina@yahoo.com.br, amanda.li.vi@hotmail.com, madeleineafigueiredo@gmail.com, anavilela\_bio@hotmail.com, ricardodestephano@hotmail.com

<sup>&</sup>lt;sup>2,7</sup>Embrapa Café, PqEB - Av. W3 Norte (Final), Asa Norte, Ed. Sede Embrapa - 70.770-901 - Brasília - DF - sttela.rosa@embrapa.br, milene.carvalho@embrapa.br

intermediate seeds (ELLIS et al., 1990). For that reason, preserving coffee seeds has become a big challenge (BERJAK; PAMMENTER, 2013), especially a problem for seedling producers, who need to conserve seeds up to the suitable time for sowing. To try to resolve this problem, studies are performed for the purpose of finding new characteristics in seeds of new coffee cultivars and of characterizing their drying conditions and to what extent they maintain longevity.

In the midst of a crop field of the cultivar Acaiá (C. arábica) in Capitólio, MG, a coffee plant that had larger than normal fruit and leaves, presumably through having undergone a mutation. was identified and named Big Coffee VL. Seeds of this coffee plant were sown in Piumhí, MG. In the new crop generated, high segregation was observed and the plants were classified, taking into account the size of fruit and leaves and plant height. The seeds were classified by size and used for establishing new crops at the Universidade Federal de Lavras, in Lavras, MG. The plants were classified as Big Coffee VL. small, Big Coffee VL. medium, and Big Coffee VL. large, for the purpose of beginning studies on this coffee plant (CASTANHEIRA et al., 2016; SILVA et al., 2016).

The increase in the size of coffee seeds/beans is one of the purposes of studies in coffee breeding, which may add greater value to the product (GUEDES et al., 2013). There are few studies on the relationship between the size and quality of the seeds of the *Coffea* species. Therefore, it is important to assess the physiological performance of the seeds of Big Coffee VL., since its seeds are larger. This will allow study of the relationship between seed size and physiological quality, comparing Big Coffee seeds with common coffee seeds. The aim in this study was to investigate desiccation tolerance, classify seeds by size, and evaluate the physiological quality of the seeds of these plants, investigating the interaction between the size of the seeds and physiological performance.

#### 2 MATERIALS AND METHODS

#### Harvest of fruit and obtaining seeds

Fruit from the Big Coffee VL. progenies and from the cultivar Topázio, both of the species *C.arabica*, were harvested manually in the cherry maturity stage in two periods, July 2016 and May 2017, in a crop field set up at the Universidade Federal de Lavras.

Fruit from 5 progenies considered as the large type (L), 5 progenies considered as the medium type (M), and 5 progenies considered as the small type (S) were collected at random. For the control, fruit from 5 plants of the Topázio cultivar, of the species *C. arabica* were collected. These plants were also chosen at random.

The fruit from each plant was pulped and fully washed manually and separately so that there was no mixing. Mucilage was removed by fermentation in water in a fermentation chamber at 25°C for 24 hours, until all the mucilage was liquefied. After that, the seeds were washed in running water for total removal of the mucilage.

To remove surface water, all the seeds were placed in a single layer over a screen and kept in the shade, always with attention to identification of the seeds from each plant. After surface drying, initial moisture content of the seeds was determined, which was around 48% wb. These seeds were then studied for desiccation tolerance and the effect of seed size on physiological performance.

# Study of desiccation tolerance of seeds of Big Coffee VL

The seeds of Big Coffee VL. small, Big Coffee VL. medium, and Big Coffee VL. large, with initial moisture content of 48% wb were divided into two parts; one of the parts was dried in a mechanical dryer, and the other part was used without drying.

Drying was performed by forced convection in fixed bed dryers composed of six square trays with a perforated bottom and sides of 0.35 m and depth of 0.4 m, with uniform airflow. The airflow throughout the seed drying process was monitored by a paddle wheel anemometer, which was regulated and maintained at 24 m³.min¹.m². Temperature was adjusted and controlled by thermostats and an electronic panel, which were constantly monitored with the aid of mercury thermometers placed within the seed mass. The seeds were dried at a temperature of approximately 38°C until reaching a moisture content of 11% wb.

Moisture content was determined by the laboratory oven method at 105°C for 24 hours (BRASIL, 2009), with two replications of 5 seeds for each treatment. The results were expressed in percentage based on the wet weight of the seeds.

The samples were monitored until reaching the desired moisture content of 11% (wb).

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The dried seeds and those not subjected to drying of each S, M, and L progeny and of the Topázio cultivar were evaluated physiologically by the germination test and by determination of seedling dry matter.

In the germination test were used four replications of 25 seeds of each treatment, which were sown in germination paper moistened with distilled water at 2.5 times the weight of the dry paper. The rolls of germination paper were placed in a germinator at a temperature of 30°C in the presence of light (BRASIL, 2009).

Percentages of root emergence at 15 days and percentage of normal seedlings at 30 days after sowing were determined; normal seedlings were considered those that had a main root and at least two lateral roots. The germination test also determined the percentage of strong normal seedlings, which were considered those that had a hypocotyl with at least three centimeters length, and the percentage of seedlings with expanded cotyledonary leaves at 45 days after sowing.

Seedling dry matter was determined at the end of the germination test in the seedlings with expanded cotyledonary leaves; the number of seeds sown was considered in the calculation. The shoots of the seedlings were separated from the roots using a scalpel and the plant matter was placed in paper bags and dried in a forced air circulation oven at 60°C for five days, until reaching constant weight. Dry matter was determined on a precision balance.

# Big Coffee VL. seed classification by size and physiological quality

For size classification, seeds collected in 2017 were used from plants of five progenies named in the field as P26, P29, P31, P10, and P32, considered to be of the Big Coffee VL. small type; five named as M11, M17, M27, M8, and M23, progenies of the Big Coffee VL. medium type; and five named as G22, G18, G21, G19, and G25, progenies of the Big Coffee VL. large type. Topázio seeds were collected from plants chosen at random, and their plants do not have a specific designation in the field. A sieve analysis test was carried out on seeds without parchment and with moisture content of approximately 11% using oblong particle size analysis sieves of numbers 17, 15, and 13 for separation of peaberry type seeds, and circular particle size analysis sieves of numbers 22, 21, 20, 19, 18, 17, 16, 15, 14, 13, and 12 for separation of flat seeds. For seed classification, sieves of numbers 11, 12, 13, and 14 were considered as small sieves; 15 and 16 as medium number sieves, and sieves 17, 18, 19, 20, 21, and 22 as large number sieves (BRASIL, 2003). The seeds classified by size were evaluated physiologically through the germination test, performed as already described.

### Experimental design and statistical analysis

For the study of desiccation tolerance of Big Coffee VL seeds the experimental design was completely randomized in a 2 × 4 factorial arrangement (with and without drying; and the progenies S, M, and L and the Topázio cultivar).

In the study of the effects of seed size on physiological quality, the experimental design was completely randomized in a  $2 \times 6$  factorial arrangement (seeds of the Big Coffee VL. and of the Topázio cultivar progenies; and sieve numbers from 13 to 18).

For the study of the effects of seed sizes of the Big Coffee VL. progeny on physiological quality, the experimental design was completely randomized, with 9 sieve numbers (from 12 to 20) and four replications. The data were subjected to analysis of variance and the qualitative means were grouped by the Scott-Knott test at 5% probability (FERREIRA, 2011).

### 3 RESULTS AND DISCUSSION

# Study of desiccation tolerance of Big Coffee VL seeds

The physiological quality of the seeds of the Big Coffee VL. S, M, and L progenies and of the Topázio cultivar was evaluated before and after drying to 11% wb of moisture content. Analysis of variance showed that for most of the variables of physiological quality, there was significant interaction between the progeny or cultivar and drying factors. There was no interaction between the factors studied only for the variables shoot dry matter and root dry matter; however, there was significance for each factor separately (Table 1).

The seeds of the S progeny and of the Topázio cultivar, with moisture of approximately 48% wb, which had not been dried, had a higher percentage of root emergence than the others. Nevertheless, after drying, there was no difference in the values of this variable among the seeds of the progenies studied (Table 1).

At 30 days after sowing, the seeds without drying also had better results of percentage of normal seedlings in the germination test for the S progeny and for the control, represented by the Topázio cultivar. When undergoing drying, only the control seeds stood out from the others (Table 1).

**TABLE 1** - Percentage of root emergence (RE), normal seedlings (NS), strong normal seedlings (SNS), and of seedlings with expanded cotyledonary leaves (ECL) from seeds of Big Coffee VL., S, M, and L progenies and of the Topázio cultivar, without drying and dried to the moisture content of 11% (wet basis).

		RE	N	S	SN	IS	EC	L
Big C. Cultivar	Withou drying		Without drying	With drying	Without drying	With drying	Without drying	With drying
Large	53 Cb	72 Aa	19 Cb	45 Ba	00 Bb	11 Aa	33 Aa	18 Aa
Medium	56 Cb	69 Aa	24 Cb	43 Ba	1 Bb	10 Aa	36 Aa	12 Ab
Small	80 Ba	81 Aa	54 Ba	54 Ba	13 Aa	10 Aa	45 Aa	60 Aa
Topázio	93 Aa	75 Ab	72 Aa	66 Aa	5 Ba	6 Aa	42 Ab	64 Aa
CV	(%)	9.87		27.34	61	.25	32.5	51

Means followed by the same uppercase letter in the column and same lowercase letter in the line belong to the same cluster at the level of 5% probability according to the Scott Knott test.

Considering evaluation of seed vigor, moist seeds of the S progeny had a higher percentage of strong normal seedlings compared to seeds of the other progenies and of the Topázio cultivar.

After drying, there was no difference between the values of the different progenies and of the control seeds. For the results of percentage of seedlings with expanded cotyledonary leaves at 45 days after sowing, the seeds of the S, M, and L progenies and of the control had statistically equal values, regardless of whether or not they were dried.

In general, both seeds of the progenies and of the control had better physiological quality, evaluated by germination and vigor, after being dried, which can be verified by the variables of percentage of root emergence, percentage of strong normal seedlings, and seedlings with expanded cotyledonary leaves. Physiological performance also improved after seed drying for seeds of the M and L progenies.

Better physiological performance can be explained by greater ease of removal of the parchment after drying of seeds to lower moisture contents, minimizing the possibility of damage to seeds. Another explanation would be lower proliferation of fungi in the germination test in the dried seeds, which had better germination and vigor. Rosa et al. (2005) found that in coffee seeds stored at a higher moisture content, there may be fungi and losses in seedling development, resulting in lower germination percentages.

For all the variables analyzed, the seeds of the S progenies and Topázio cultivar (control) had better results, regardless of having been dried or not (Table 1). Coelho et al. (2017) investigated the quality of dried seeds and concluded that water contents below 0.11g H<sub>2</sub>O g<sup>-1</sup> (dry weight-dw) and above 0.43 g H<sub>2</sub>O g<sup>-1</sup> dw hurt the physiological quality of coffee seeds, regardless of the type of drying, temperature, and period of storage. Figueiredo (2017) observed that *C.arabica* seeds have good physiological quality when dried to the moisture content of 20%, being tolerant to the cryopreservation stresses. Corroborating this literature, seeds of Big Coffee VL. tolerate desiccation to the moisture content of 11% wb; however, smaller seeds have better physiological performance. Coelho et al. (2015) state that drying losses for physiological quality of coffee seeds depend on the speed with which water is removed and on the final moisture content of the seeds.

Subjecting seeds to the drying process allowed an increase in shoot dry matter and root dry matter of the plants studied (Table 2).

# Classification by size and physiological quality of Big Coffee VL seeds

Seeds of the Big Coffee VL. S, M, and L progenies and of the Topázio cultivar were classified regarding size through the sieve retention test, and the results are shown in Table 3. It can be seen from the percentages of retention in each sieve that there are small, medium, and large seeds in all the Big Coffee VL. progenies, even if in varied proportions. This is probably due to the segregation that still exists in these plants.

Nevertheless, progeny L, classified in the field as having large seeds, had 61.16% of seeds retained in the sieves with numbers considered large (sieves 17 to 22), 31.70% in the medium numbered sieves (sieves 15 and 16), and 6.88% in the small numbered sieves (sieves 11 to 14) (Table 3).

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**TABLE 2** - Shoot dry matter and root dry matter of seeds of Big Coffee VL. and of the Topázio cultivar, without drying and dried to the moisture content of 11% (wet basis).

Drying	Shoot dry matter	Root dry matter	
With drying	32.67 A	5.68 A	
Without drying	16.52 B	2.58 B	
CV (%)	34.30	39.03	

Means followed by the same letter in the column belong to the same cluster at the level of 5% probability according to the Scott Knott test.

**TABLE 3 -** Percentage of seeds of the progenies S, M, and L of Big Coffee VL., and of the Topázio cultivar of different sizes, classified by retention in particle size sieves.

				S	ieve N	umber						
		Sı	mall		Med	lium			La	rge		
	11	12	13	14	15	16	17	18	19	20	21	22
B. Coffee L	0.00	0.22	2.54	4.12	10.9	20.8	27.6	19.7	10.8	2.96	0.02	0.08
B.Coffee M	0.06	0.96	2.67	7.63	11.9	18.1	24.9	20.3	9.50	3.55	0.13	0.09
B. Coffee S	0.08	1.50	4.18	11.1	18.3	31.7	22.8	8.84	1.27	0.03	0.00	0.06
Topázio	0.00	1.07	4.49	9.45	26.3	36.6	17.4	3.35	0.93	0.11	0.00	0.00

As for progeny M, the greatest proportion of the seeds was retained in large sieves, with 58.47%; in medium numbered sieves, the percentage was 30.00%; and small numbered sieves represented 11.32%. The small, medium, and large numbered sieves retained 16.86%, 50.00%, and 33.00% of the seeds of progeny S, respectively.

For the Topazio cultivar, the results were similar to those of the S progeny; i.e., 15.00% of the seeds were retained in the small sieves; 62.90% in the mean numbered sieves; and 21.70% in the large sieves. The largest proportion of seeds was retained in the medium sieves.

Seeds of the Topázio cultivar were not retained in sieves 11, 21, and 22, and few seeds were retained in sieves 12, 19, and 20. Likewise for the Big Coffee VL. progenies, few seeds were retained in sieves 11, 21, and 22. Thus, the number of seeds from these sieves was not sufficient to perform the germination test.

Classification of the coffee seeds/beans according to size is relevant for uniform roasting. For international trade, coffee beans from sieve numbers 13 to 20 are accepted; however, there is a tendency among exporters to prefer sieve sizes greater than 16 (LAVIOLA et al. 2006;

ROTONDANO et al. 2005). Dias Caldas et al. (2018), who worked with classification regarding the size of *C.arábica* L., cv. Travessia seeds, observed that seeds retained in larger screens were significantly influenced by irrigation and fertilization management practices.

For the sieve-classified seeds, both for Big Coffee VL. and for the Topázio cultivar, physiological performance was evaluated. Analysis of variance showed that for the variables of root emergence, normal seedlings, and seedlings with expanded cotyledonary leaves, there was no significant interaction among the progeny/cultivar factors and sieve number, but there was significance of these factors separately (Table 4). The Topázio cultivar exhibited better physiological performance than the seeds of the Big Coffee VL. progenies, according to the values of root emergence, of normal seedlings, and of seedlings with expanded cotyledonary leaves (Table 4).

Even though significant differences were not observed on physiological performance of the seeds classified by size, a tendency of increase in quality with a decrease in the sieve size number can be seen (Table 4).

Progeny/Cultivar	Root Emergence	Normal Seedlings	<b>Expanded Cotyledonary Leaves</b>
Big Coffee VL.	85.6 b	72.5 b	67.6 b
Topázio	92.8 a	82.3 a	75.8 a
CV (%)	8.04	11.70	11.93
Sieve	Root Emergence	Normal Seedlings	<b>Expanded Cotyledonary Leaves</b>
18	85.5 a	77.5 a	71.5 a
17	87.0 a	75.0 a	68.0 a
16	91.5 a	76.5 a	75.5 a
15	89.5 a	77.5 a	71.0 a
14	89.0 a	79.0 a	72.0 a
13	93.0 a	79.0 a	72.5 a

**TABLE 4** - Effect of the Big Coffee VL. progeny or of the Topázio cultivar and of the number of sieve size classification on root emergence, normal seedlings, and seedlings with expanded cotyledonary leaves of the seeds.

Means followed by the same letter in the column belong to the same cluster according to the Scott Knott test at the level of 5% probability.

11.70

8.04

The seeds of the Topázio cultivar exhibited a higher percentage of normal seedlings, as well as higher vigor, evaluated by the percentage of root emergence and of seedlings with expanded cotyledonary leaves at 45 days. Concerning the effects of sieve size number, the large sized seeds, 17 mm and greater, the medium sized seeds, of 15 and 16 mm, and the small sized seeds, of 13 and 14 mm (BRASIL, 2003) exhibited differentiated physiological performances, with better germination and vigor in the small sized seeds.

In studies on physiological performance according to coffee (*C. arabica*) seed size, there were no significant differences in the results of germination of seeds produced under different conditions of soil, temperature, and rainfall in the phases of fruit expansion and growth (FAVARIN et al., 2003). According to the authors, the separation of coffee seeds by size is a necessary but not sufficient condition for adequate selection for estimation of seed physiological potential and the formation of quality seedlings.

Flores et al. (2014) and Pardo et al. (2015) observed that smaller sized seeds of *braúna* and of soybean, respectively, have low physiological quality compared to those of larger sizes. According to the results presented, plants or progenies arising from seeds with sizes corresponding to lower sieve numbers will not necessarily have lower vigor or less viability than the others.

In this study, the results of percentage of seedlings with expanded cotyledonary leaves, an indication of seedling vigor, do not differ statistically according to sieve size, but did exhibit higher absolute values, the overall mean being 71%.

For the variables of shoot and root dry matter, there was also no significant effect of the interaction of the progeny or cultivar factors and sieve number on physiological response (Table 5). Nevertheless, there were significant differences among the seeds of different sizes and different progenies/cultivar.

The seedlings of the Topázio cultivar had greater shoot dry matter than the seedlings of the Big Coffee VL. progenies, and also had better germination and vigor results (Table 4). Analysis of the effects of sieve number and consideration of the sieves common to the Topázio cultivar and Big Coffee VL. progenies on seedling dry matter showed that the seedlings coming from medium and high sieve size, of size 15 to 18, obtained higher values than small sieves, of size 13 and 14 (Table 5).

Seedlings coming from larger seeds had more developed shoots and root system than seedlings from seeds of smaller sieve sizes, and this was reflected in the values for dry matter. A less developed root system in tropical and/or subtropical species that produce recalcitrant seeds may occur due to the presence of reactive oxygen species (BERJAK; SERSHEN; PAMMENTER; 2011).

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Seeds of the Big Coffee VL. progenies classified by size, of number 12 to 20, were evaluated in regard to physiological performance, and the results are shown in Table 6. In this analysis, seeds of the Topázio cultivar were not considered. Significant differences were observed among the sieve sizes only for the results of seedling dry matter (Table 6); the seedlings coming from seeds of higher sieve size had greater vigor than those from smaller sieve sizes, unlike the analysis in which seeds of the Topázio cultivar were compared with those of Big Coffee VL. (Table 5).

According to these variables that indicate vigor, a decline in the physiological quality of Big

Coffee VL. seeds is observed along with reduction in sieve size, i.e., seeds retained in sieves with larger openings had higher vigor. It is noteworthy that among the Big Coffee VL. seeds classified by size, there was a positive effect of greater size on physiological quality. However, when these seeds were compared to Topázio cv seeds, with same size, there was greater influence of the cultivar, in detriment of the effects of seed size. In other words, when the physiological quality of the seeds of the progenies and cultivar is compared regarding size, there is predominance of the effect of the cultivar; the seeds of the Topázio cultivar exhibit better physiological quality.

**TABLE 5** - Effect of the Big Coffee VL. progeny or of the Topázio cultivar and of the number of sieve size classification on shoot dry matter and root dry matter.

Progeny/Cultivar	<b>Shoot Dry Matter</b>	Root Dry Matter
Big Coffee VL.	0.70 b	0.14 b
Topázio	0.80 a	0.17 a
CV (%)	13.95	24.09
Sieve	Shoot Dry Matter	Root Dry Matter
18	0.83 a	0.20 a
17	0.80 a	0.17 a
16	0.81 a	0.14 b
15	0.68 b	0.13 b
14	0.70 b	0.16 a
13	0.67 b	0.11 b
CV (%)	13.95	24.09

Means followed by the same letter in the column belong to the same cluster according to the Scott Knott test at the level of 5% probability.

**TABLE 6** - Shoot dry matter and root dry matter of seedlings coming from seeds of the Big Coffe VL. progenies classified in different particle size sieves.

	Sieve Number	Shoot Dry Matter	Root Dry Matter
Large	20	1.07 a	0.23 a
	19	0.82 b	0.19 a
	18	0.82 b	0.20 a
	17	0.73 c	0.17 b
Medium	16	0.81 b	0.14 b
	15	0.61 d	0.09 c
Small	14	0.67 c	0.14 b
	13	0.57 d	0.09 c
	12	0.49 d	0.06 c
CV (%)		13.84	26.72

Means followed by the same letter in the column belong to the same cluster according to the Scott Knott test at the level of 5% probability.

#### **4 CONCLUSIONS**

Big Coffee VL. seeds tolerate desiccation to moisture content of 11% wet basis.

Seeds of the Topázio cultivar have better physiological performance than seeds of the Big Coffee VL. progenies.

The bigger the size of the Big Coffee VL. seeds, the better their physiological performance, exhibiting greater seedling dry matter.

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# MODELS IDENTITY GENERATED IN NEUTRON PROBE CALIBRATION IN LATOSOL CULTIVATED WITH COFFEE AND SIGNALGRASS

Inácio Barbosa Borges<sup>1</sup>, Omar Cruz Rocha <sup>2</sup>, Juaci Vitória Malaquias <sup>3</sup>, Marcelo Fagioli <sup>4</sup>, Marcos Aurélio Carolino de Sá<sup>5</sup>, Antônio Fernando Guerra<sup>6</sup>

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ABSTRACT: The cover plants affects soil spatial variability interfering with soil moisture and density, leading to doubts about the need for calibration of the neutron probe for different management systems. The objective of this study was to evaluate the Model Identity Method in the verification of equality of linear regressions resulting from the calibration of a neutron probe for different layers, as a function of signalgrass management as a cover crop between rows of soil coffee crop in the Brazilian Central Cerrado. Aluminum tubes were installed to access the probe and two saturation basins were built in a coffee area, irrigated by a central pivot, in an Oxisol, with two management systems, T (Traditional with bare soil) and Sg (Signalgrass in the row). Samples were taken at depths of 0-0.20; 0.20-0.40; 0.40-0.60; and 0.60-0.80 m for determination of gravimetric moisture and density, and the volumetric moisture was determined to follow the drying of the soil. Concomitantly, measurements were taken with the neutron probe in these layers. Two calibration curves were constructed for each layer, which were compared by the above method. It was observed that treatment Sg yielded differences in soil water storage in the 0.20-0.40 m layer, differing from treatment T. The statistical method allowed the observation of equality of regressions between treatments Sg and T at depths 0-0.20, 0.40-0.60, and 0.60-0.80 m; it also showed the need for different regression curves per layer, besides justifying the need for neutron probe calibration for each specific local condition.

Index terms: Coffea arabica, soil moisture, management systems, sustainability, cover crops.

# IDENTIDADE DE MODELOS GERADOS NA CALIBRAÇÃO DA SONDA DE NÊUTRONS EM LATOSSOLO CULTIVADO COM CAFEEIRO E BRAQUIÁRIA

RESUMO: As plantas de cobertura afetam a variabilidade espacial do solo interferindo na umidade e densidade do solo, levando a dúvidas sobre a necessidade de calibração da sonda de nêutrons para diferentes sistemas de manejo. O objetivo deste trabalho foi avaliar o Método da Identidade de Modelos na verificação de igualdade de regressões lineares resultantes da calibração de uma sonda de nêutrons para diferentes camadas do solo, em função do manejo da braquiária como planta de cobertura nas entrelinhas da cultura do café no Cerrado Central do Brasil. Foram instalados tubos de alumínio para acesso da sonda e construiu-se duas bacias de saturação em área de café, irrigada por pivô central, em um Latossolo Vermelho distrófico, com dois sistemas de manejo, sendo T (Tradicional com solo nu) e B (Braquiária na entrelinha). Retirou-se amostras nas profundidades 0-0,20; 0,20-0,40; 0,40-0,60; e 0,60-0,80 m para determinação propriedades físico-hídricas e acompanhamento da secagem do solo, concomitantemente tomou-se medidas de leitura com a sonda de nêutrons nestas camadas. Construiu-se duas curvas de calibração para cada camada, as quais foram comparadas pelo método citado. Observou-se que o tratamento B proporcionou diferenças no armazenamento de água no solo na camada 0,20-0,40 m, diferenciando-se do tratamento T. O método estatístico: permitiu a observação de igualdade das regressões entre os tratamentos B e T nas profundidades 0-0,20, 0,40-0,60, e 0,60-0,80 m; mostrou a necessidade de diferentes curvas de regressão por camada; justificou a necessidade da calibração da sonda de nêutrons para cada condição local específica.

Termos para indexação: Coffea arabica, umidade do solo, sistemas de manejo, sustentabilidade, plantas de cobertura.

### 1 INTRODUCTION

The sustainability of agricultural production can be improved through the use of cover crops. Variations in exploitation form and depth of the root system of plants used in covers and their capacity of biomass production, in relation to the main crop, allow the recycling of nutrients,

protection against water erosion, besides hindering surface runoff and favoring soil water infiltration by the opening of drainage canals when their roots die (CARDOSO et al., 2012), and by the improvement in structural quality (CALONEGO; BORGHI; CRUSCIOL, 2011; LAL, 2015) and biological activity (MBUTHIA et al., 2015). In addition, it contributes to a reduction in water

<sup>&</sup>lt;sup>1</sup>Instituto Federal do Norte de Minas Gerais/IFNMG - Cx. P. 05 - Rodovia MG 202, Km 407, s/n - Zona Rural Arinos - 38.680-000-Arinos - MG - inacio.ifnmg@gmail.com

<sup>2.6</sup> Embrapa Café - Parque Estação Biológica/PqEB - Av. w3 Norte (final) - 70.770-901 - Brasília - DF - omar.rocha@embrapa.br, antonio.guerra@embrapa.br

<sup>&</sup>lt;sup>3,5</sup>Embrapa Cerrados - Rodovia BR-020, Km 18 - Cx. P. 08223 - 73.310-970 - Planaltina - DF - marcos.sa@embrapa.br, juaci.malaquias@embrapa.br

<sup>&</sup>lt;sup>4</sup> Faculdade de Agronomia e Medicina Veterinária/UnB - Campus Darcy Ribeiro - ICC-Sul - Brasília - DF - 70.297- 400 - mfagioli@unb.br

evaporation and thermal oscillation (MATORANO et al., 2009), with a direct positive effect on soil moisture (BORGES et al., 2014; CARDOSO et al., 2012; SANTOS et al., 2010).

According to Balbinot Junior; Santos; Yokoyama (2017), signalgrass, a plant belonging to the genus *Brachiaria* spp., can improve soil quality from the direct action of its roots in the structure, improving infiltration, water retention, oxygen flow and reducing soil resistance to penetration of crop roots. Adaptability to local conditions, adequate architecture to the coffee production system, perenniality, ease of natural resowing, rusticity, ease of maintenance and resistance to mechanization are characteristics that qualify signalgrass as a cover crop (BULISANI et al., 1993; FIRTH; WILSON, 1995), which can be used between coffee rows.

Soil properties such as texture, structure, porosity and organic matter determine its adsorption and capillarity characteristics, while rainfall, solar radiation, temperature, crop evapotranspiration, leaf area index, plant density and soil management systems affect moisture and drying periods (MARTORANO et al., 2009). Cover plants increase microbiological activity, nutrient cycling (MBUTHIA et al., 2015) and soil moisture directly influencing plant development of coffee; therefore, it is an indispensable information to evaluate the contribution of different management systems.

Among the methodologies used in research to monitor soil water content, the use of the Neutron Probe in both irrigated and non-irrigated areas is considered as the standard method. Although the use of this equipment is repeatedly addressed by several authors (BORGES et al., 2014; MENEZES et al., 2013; SANTOS et al., 2010), it is common knowledge that calibration is a fundamental part of its use (FALLEIROS et al., 1993; KODIKARA et al., 2014). The calibration curve, however, is affected by the spatial variability of the soil and by moisture and density, which result in the main causes of the dispersion of points in the curve (GREACEN, 1981). Although there is a consensus on the need for specific models for each type of soil, there is uncertainty whether different management systems alter calibration curves, as well as whether the various curves generated result in significant differences in soil water content.

The Model Identity Method proposed by REGAZZI (2003) allows to analyze the feasibility of using a single model for situations where

several models are observed. This procedure has been widely used in forestry engineering due to the large number of models generated for administrative planning and prediction of yield capacity and production (ROVEDA et al., 2016). Thus, this procedure could be used in the comparison of models that describe the behavior of the calibration of a neutron probe under different types of soil and / or handling.

Therefore, the objective of this study is to evaluate the Model Identity Method in the verification of equality of linear regressions resulting from neutron probe calibration for different soil layers, as a function of signalgrass management as a cover crop between coffee rows in the Brazilian Central Cerrado.

## 2 MATERIAL AND METHODS

The experiment was set at Embrapa Cerrados, in Planaltina, Federal District, Brazil (15°35'42" S, 47°43'51" W and 1009 m). The climate of the region is CWh1, according to the Koppen classification, with an annual average rainfall of 1,460 mm and average temperature of 21.3°C (ROCHA et al., 2014) with a large variation of precipitation over the months of the year. The months of May to September are considered the dry season and present a historical cumulative average of 67 mm and from October to April the wet season with a cumulative historical average of 1400 mm (CARDOSO; MARCUZZO; BARROS, 2014).

The soil of the experimental area is classified as an Oxisol (LVd), of clayey texture, irrigated by a central pivot irrigation system, cultivated with signalgrass (Brachiaria decumbens Stapf) from January 2000 until December 2007 when coffee (Coffea arabica L.) was planted. The cultivar 'Catuaí vermelho IAC 144' coffee (Coffea arabica L.) was planted, with a 3.50m spacing between rows per 0.70m between plants in the row, with 120 g of triple superphosphate were added, as well as 50 g of magnesium thermophosphate (Yoorin®) and 24.5 g of fritted trace elements (FTE) per well. Liming was performed with 2 mg ha-1 dolomitic limestone, in order to raise base saturation to 50%. In the years after planting, the nutritional management of coffee trees was carried out according to Guerra et al. (2008).

In the irrigation strategy of coffee trees, the controlled water deficit was used to standardize flowering (GUERRA et al., 2007), with a management criterion based on the monitoring of soil water content, according to Rocha et al.

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(2008). Two management systems were used: with Signalgrass (Sg) and Traditional (T), distributed in nine-plant plots; the five central plants were considered useful. For each useful coffee row, two parallel rows were border. Signalgrass was managed with mechanical handling when the plant reached 0.60 m average height, maintaining the crop remains in the area. In the traditional treatment, the soil was kept free of invasive plants with the aid of manual weeding, keeping the same crop remains in the respective plots.

The experimental design was in five randomized blocks in subdivided plots, in which the plot consisted of the two management systems (Sg and T) and the subplots, of the soil layers 1 (0.00 to 0.20 m), 2 (0.20 to 0.40 m), 3 (0.40 to 0.60 m) and 4 (0.60 to 0.80 m) (1, 2, 3 and 4).

In September 2014, the coffee trees were submitted to pruning, cutting the orthotropic branch 0.80 m above ground level, leaving the existing plagiotropic branches up to that height and leading one orthropic branch per plant. In May 2015, aluminum access tubes were implanted in all experimental plots, according to Kodikara et al. (2014) for soil water content monitoring, using a neutron probe (CPN 503 TDR Hydroprobe®), supplied with a 241Am/Be source of 1.85 GBq radioactive activity. Four soil monitoring layers were used, corresponding to the depths of moisture monitoring with the probe 0,10; 0.30; 0.50 and 0.70 m, respectively.

For probe calibration, a 4-m<sup>2</sup> saturation basin formed by an zinc plate (0.30 m x 8m), installed in the form of a square nailed to the soil, was constructed in representative plots of each management system, keeping 0.10 m buried. The basins received a sufficient water volume to saturate the soil profile up to 1.00 m deep. After the complete saturation of the soil profile, the basin was covered with a plastic canvas for 24 hours to control evaporation and deformed soil samples immediately underwent natural drying, from the determination of gravimetric moisture by the greenhouse method and then, with soil density, the volumetric moisture (Empresa Brasileira de Pesquisa Agropecuária - EMBRAPA, 2011). A total of five replicates at the depths of the probe reading (0.10; 0.30; 0.50 and 0.70 m), on seven different dates, between the months of June and September, period without rainfall. Subsequently, at each sampling, readings were taken with the use of the neutron probe. The relative counting (RC) of neutrons in the soil was obtained according to Kodikara et al. (2014), from the division of the soil reading values obtained in the access tubes, by the counting obtained inside the probe head housing.

The models for describing the soil moisture variation for each layer, as a function of the management system, were obtained by correlating the relative counting (RC) with the volumetric moisture  $(\theta)$ .

In order to analyze the chemical attributes and physical and water properties of the soil, deformed and undeformed samples, respectively, were collected, in duplicate in the projection of the crown of the coffee trees in each experimental plot. With the deformed samples, the particle density was analyzed by the volumetric flask method and ethyl alcohol (EMBRAPA, 2011). The undeformed samples, collected in rings (50 mm x 51 mm), were used to determine soil density (SD) and the water retention capacity by the Richards chamber method (EMBRAPA, 2011) at water tensions corresponding to 3, 6, 10, 33, 60, 100 and 1,500 kPa. The experimental points of the retention curves were adjusted using the van Genuchten equation (1980), using the restriction proposed by Mualem (1986). The SWRC (3.00 beta) program, developed by Dourado-Neto et al. (2001), was used to adjust the retention curves.

The total water availability (TWA, in mm cm-1) of the soil was obtained from the following expression: TWA= ((U6-U1,500)/10)SD, where,  $\rm U_6$  and  $\rm U_{1,500}$  correspond to the gravimetric moisture expressed as % equivalent to matrix tensions of 6 and 1,500 kPa determined by the Richards chamber method; SD corresponds to the soil density, in mg m<sup>-3</sup>.

A total of eight linear models resulting from the relation between the Relative Counting (x) and the Average Volumetric Humidity (y) were generated for different management systems and depths, given by:

$$y_i = \beta + b_1 x_{1i} + b_2 x_{2i} + ... + b_k x_{ki} + e_i$$
  
where,

 $y_i$ : i-th value of the response variable, i=1,2, ..., N observationss;

 $x_{ki}$ : i-th value of k-th explanatory variable, k=1,2,...,K variables;

bk: linear model parameter;

e: random errors.

In the linear regression model, the minimum square estimation method can range between ordinary, weighted and generalized, according to the assumptions that residue can assume. In this study, from the verification of the assumptions of residue linearity, normality, homogeneity and

independence, for the adjustment of the models, the Ordinary Least Squares Method was used to estimate the parameters  $\beta$ . The adjustment of a model by this method assumes that the mean of the residue is zero; the error variance is constant and equal to  $s^2$ ; the residue is independent, that is, the errors are random variables with normal distribution.

After the generation of linear regression models, the possibility of constructing a single regression model for each depth was tested, independently of the management system. Thus, a test was applied to verify the equality of two linear regressions, called Model Identity Method (REGAZZI, 2003). With this method, models were created from each coefficient (angular and linear) and from the standard error, which were compared by the F test and as a rule of decision it was determined that if  $F_{cal} > F_{table}$ , for a given level of significance  $\alpha$ , we reject the hypothesis that the coefficients are the same for the two sets of observations.

In order to determine the possible differences in the calibration curves of the probe as a function of the different treatments, the total porosity was determined in the different layers from the expression: TP=1- (SD/PD) 100, where: TP is the total porosity (%); SD is the soil density (mg m<sup>-3</sup>) and PD is the particle density (2.65 mg m<sup>-3</sup>) (EMBRAPA, 2011).

Likewise, in order to understand the distribution of the root system of the coffee tree and signalgrass in the different management systems, in March 2016, soil samples were taken in the projection of the coffee crown, at the same depths of the samples collected in the saturation basins, to evaluate the distribution of the root system. Thus, a 0.10-m auger was used for collecting the soil at the depths of the probe reading. The samples were washed in running water on 0.001- and 0.0005-m sieves. The remaining impurities were collected with tweezers.

The roots were placed in a 150-mL bottle containing 70% ethyl alcohol for conservation. Subsequently, they were treated with gentian violet for 24 hours. They were then washed for the removal of the excess dye, scattered in a glass tray with a white background and water slide, and photographed with an 8-megapixel camera. The root samples were placed in an oven at 60°C for 48 hours in paper bags and then weighed in a precision scale to obtain the root dry matter. With the obtained images, the root area, volume, length and diameter were determined using the free software Safira, version 1.1 (JORGE; SILVA, 2010).

Data were submitted to the normality test (Shapiro-Wilk 5%) and the means were compared by the Tukey test at 5% probability. All statistical analyses were performed using the free software R, version 3.2.2 (R Core Team, 2016).

### 3 RESULTS AND DISCUSSION

The linear regression models for the neutron probe calibration curves of the four depths and the respective studied soil layers  $(1-0.10~\text{m}\ \text{or}\ 0.10~\text{m})$  or 0 to 0.20 m; 2-0.30~m or 0.20 to 0.40 m; 3-0.50~m or 0.40 to 0.60 m; and 4-0.70~m or 0.60 to 0.80 m), as a function of management systems (Sg – Signalgrass and T - Traditional), are found in Table 1.

Only for the linear coefficient in layer 1, there was no statistical significance, independent of the adopted management system. This result can be explained by the sphere of action of the neutron probe, which represents the range of the radiation emitted by the capsule inside the soil. The radiation sphere varies as a function of soil moisture, as reported by Falleiros et al. (1993), who observed that, under saturated soil conditions until the condition of permanent wilting point, it can range from 0.10 m to 0.20 m radius, respectively. Thus, in the reading related to layer 1, part of the radiation can exceed the soil surface as the soil moisture decreases, losing into the atmosphere, which can cause errors in probe reading, resulting in numerical values that are commonly underestimated.

According to Greacen (1981), the margin of error of the probe can be reduced with a comprehensive calibration in relation to the soil moisture range, considering from the conditions near the permanent wilting point to the saturation condition, from the moisture correlation, measured by the neutron probe, with the volumetric moisture layered by depth, as performed in this study. In fact, this comprehensive calibration method reduced the reading margin of error of the probe, both in layer 1 and in the others, as can be observed in Table 1, since the determination coefficients for both treatments (Sg and T) demonstrate that the calibration models represent the accuracy of soil moisture measurement variation with a minimum margin of error, always equal to or less than 0.015 (1.5%).

Table 2 shows the estimates of the parameters of the complete and reduced linear regression models, adjusted according to the management system and layers 1, 2, 3 and 4.

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**TABLE 1** - Adjustment of linear regression models according to the type of management (Sg and T) and soil layer.

Predictor	Coefficient	Standard Error	t - test	P value		
Layer 1		Signalgrass				
$\beta_{0 \; linear}$	-0.03141	0.016314	-1.925	$0.112^{\text{ n.s.}}$		
$\beta_{1 \text{ angular}}$	0.16255	0.009121	17.82	1.02e-5***		
	$R^2 = 0.985$					
		Trad	itional			
$\beta_{0 \; linear}$	-0.00048	0.014024	-0.034	$0.974^{\rm \ n.s.}$		
$\beta_{1 \text{ angular}}$	0.147378	0.008050	18.31	8.94e-6***		
	$R^2 = 0.985$					
Layer 2		Sign	algrass			
$\beta_{0 \; linear}$	-0.23378	0.022017	-10.62	1.28e-5***		
$\beta_{1 \text{ angular}}$	0.22493	0.009718	23.14	2.8e-6***		
	$R^2 = 0.991$					
		Trad	itional			
$\beta_{0 \; linear}$	-0.30629	0.02708	-11.31	9.44e-5***		
$\beta_{1 \text{ angular}}$	0.25082	0.01206	20.80	4.75e-6***		
	$R^2 = 0.989$					
Layer 3		Signa	algrass			
$\beta_{0 \; linear}$	-0.24649	0.02551	-9.661	2.02e-4***		
$\beta_{1 \text{ angular}}$	0.23203	0.01156	20.07	5.67e-6***		
	$R^2 = 0.988$					
		Trad	itional			
$\beta_{0 \; linear}$	-0.27325	0.02599	-10.2	1.34e-6***		
β <sub>1 angular</sub>	0.24609	0.01180	20.85	4.7e-6***		
-	$R^2 = 0.989$					
Layer 4		Signs	algrass			
$\beta_{0 \; linear}$	-0.21239	0.02788	-7.619	6.19e-4***		
β <sub>1 angular</sub>	0.22533	0.01266	17.80	1.03e-5***		
-	$R^2 = 0.985$					
		Trad	itional			
$\beta_{0 \; linear}$	-0.20714	0.02515	-8.236	4.3e-4***		
β <sub>1 angular</sub>	0.22630	0.01167	19.390	6.7e-6***		
	$R^2 = 0.987$					

<sup>\*\*\*</sup> significant at 0.1%; n.s. not significant.

**TABLE 2** - Estimates of the parameters of complete and reduced linear regression models adjusted according to the type of management (Sg and T) per soil layer.

Models	Predictor	Complete Model	Reduced Model I	Reduced Model II	Reduced Model III
				- layer 1	
S.c.	a1	-0.03141	-	-0.01886	-
Sg	b1	0.16255	0.15433	-	-
т	a2	-0.00048		-0.01412	-
T	b2	0.14738	0.15631	<u>-</u>	-
A	a	-	-0.01637	-	-0.01583
Aggregate	b		-	0.15537	0.15499
SQ res.	-	7.00E-04	0.00085	0.00081	0.00089
GL res.	-	10	11	11	12
p-value	-		$0.18207^{\rm ns}$	$0.24278^{\rm ns}$	$0.30762^{ns}$
			1	ayer 2	
	a1	-0.23378	-	-0.2584	· <u>·</u> .
Sg	b1	0.22493	0.2385	-	-
-	a2	-0.30629	-	-0.2728	-
T	b2	0.25082	0.23237	-	-
<i></i>	a	24	-0.26468	-	-0.26861
Aggregate	b	_	-	0.23586	0.2372
SQ res.	-	0.00037	0.00053	0.00047	0.0012
GL res.	-	10	11	11	12
p-value	-	-	$0.06348^{ns}$	0.12397 <sup>ns</sup>	0.00273*
				layer 3	
	a1	-0.24649	~	-0.26303	4
Sg	<b>b</b> 1	0.23203	0.23851	_	-
	a2	-0.27325	-	-0.25893	-
T	b2	0.24609	0.24048	-	_
	a	-	-0.26084	_	-0.26061
Aggregate	b	-4	-	0.23956	0.23939
SQ res.	_	0.00042	0.00044	0.00045	0.00051
GL res.	-	10	11	11	12
p-value	_	_	0.48083 <sup>ns</sup>	0.41619 <sup>ns</sup>	0.38322ns
1				layer 4	
	a1	-0.21239	1000 mm	-0.21352	-
Sg	b1	0.22533	0.22404	-	-
	a2	-0.20714	-	-0.20615	-
T	b2	0.2263	0.22742	-	-
	a	-	-0.20955	-	-0.20593
Aggregate	b	-	-	0.22585	0.22404
SQ res.	-	0.00051	0.00051	0.00051	0,0007
GL res.	_	10	11	11	12
p-value			0.89142 <sup>ns</sup>	0.95589 <sup>ns</sup>	0.20839 <sup>ns</sup>

<sup>(\*)</sup> model reduced with parameters statistically different from the complete model, at 5% probability (p-value(F) <0.05); ns not significant; SQ res. sum of squares of the regression residue, GL res. number of degrees of freedom.

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There is a significant difference in the reduced model III olnly for layer 2, indicating that the generated models, as a function of management systems, differ from each other. Thus, the Model Identity Method showed that there is a need for the use of different calibration regressions in this soil layer as a function of the soil management system to estimate the volumetric soil water content with the neutron probe.

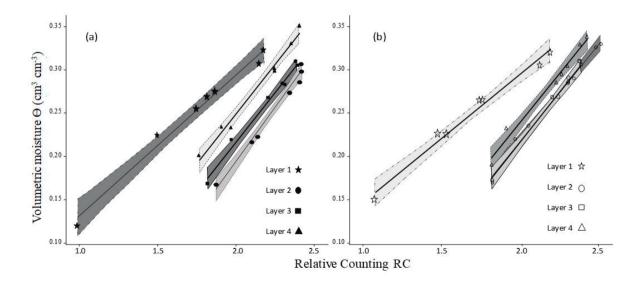
The adjustment of the calibration models of the neutron probe for layers 1, 2, 3 and 4 in the T management system is shown in Figure 1 (a). It is observed that the Model Identity Method demonstrated efficacy in the verification of the equality of the linear regressions, since the difference between the layers is evidenced by the different positioning of the lines in the graph, without the existence of overlapping confidence intervals, proving the statistical difference conditioned by different capacities in water storage, from the superficial (1) to the deepest (4) layer. Figure 1 (b) shows that there is an increase in storage capacity in layer 2, conditioned by the presence of signalgrass, matching layer 3 with overlapping confidence intervals of these two lavers.

In Figure 2 (a, c and d), the adjustment of the calibration models of the neutron probe in layers 1, 3 and 4, respectively, in the Sg and T management systems, show that the differences in water storage, due to the presence of signalgrass, did not alter the positioning of the regressions of these layers. In this condition, the Model Identity

Method demonstrated that it is possible to use a single calibration regression per layer, regardless of the management system, since there were overlapping confidence intervals within each of the different management systems, proving the applicability of the method.

In Figure 2 (b), considering only layer 2 in relation to the two management systems (T and Sg), it was observed that the regression distances resulted in overlapping confidence intervals only at the ends of the equation where the experimental error is larger, indicating that there is a difference between the regressions in this soil layer. Roveda et al. (2016) evaluated eucalyptus growth in six different locations, and observed that two models did not show statistical difference through the Model Identity method, indicating the possibility of using a single plant growth curve for these locations. For the others, as there was a statistical difference, it was concluded by using a specific model for each location.

The difference in layer 2 as a function of the management system can be attributed to the significant increase in water storage capacity in this layer since, while in the soil layers 1, 3 and 4 the presence of signalgrass changed the total soil water availability (TWA) in -0.6%, 8.8% and 4.0%, respectively (Table 3), in layer 2 this increase was higher than 11%, corroborating the significant effect observed in Table 2 and Figure 2 and the results presented by Rocha et al. (2014), who observed an increase in water readily available in a coffee area with signalgrass between rows.



**FIGURE 1** - Adjustment of the neutron probe calibration models for layers 1, 2, 3 and 4 in the Traditional (a) and Signalgrass (b) management systems.

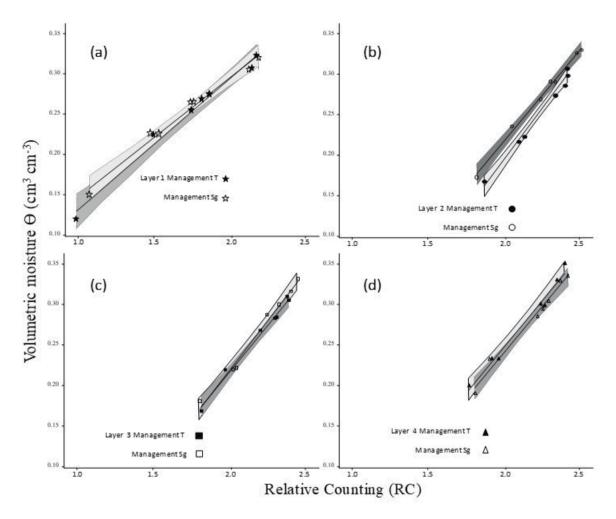


FIGURE 2 - Adjustment of calibration models in management systems T and Sg for layers 1 (a), 2 (b), 3 (c) and 4 (d).

**TABLE 3** - Total water availability (TWA) per soil layer under different management systems between coffee rows irrigated by a central pivot.

Layers	1	2	3	4
		mn	n cm <sup>-1</sup>	
Traditional	1.61	1.62	1.71	1.74
Signalgrass	1.60	1.80	1.86	1.81
Difference	-0.62%	11.11%	8.80%	4.02%

1 Layer (0-0.2 m); 2 (0.20-0.40 m); 3(0.40-0.60 m); 4(0.60-0.80 m) depth.

This increase in TWA can be understood observing Table 4 in layer 2 (0.20 to 0.40 m), where it was possible to verify that, although there was no significant effect on soil density (SD), the presence of signalgrass in this layer significantly altered total porosity (TP), root dry matter (RDM), root area (RA), mean root diameter (RD) and its

length (LR) by 3.0%, 59.9%, 19.6%, -10.8% and 39.6%, respectively (Table 3). In this context, this root dynamics can affect the degree of soil structure, as well as its organic matter, and signalgrass can improve soil physical and water quality (CRUSCIOL et al., 2015).

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**TABLE 4** - Apparent soil density (SD), Total porosity (TP), Root dry matter (RDM), Root area (RA), Root volume (RV), Root diameter (RD) and Root length (RL) of coffee and signalgrass roots in two soil management systems.

Treatments	SD	TP	RDM	RA	RV	RD	RL
	mg m <sup>-3</sup>	%	kg m <sup>-3</sup>	$m^2 m^{-3}$	$m^3 m^{-3}$	mm m <sup>-3</sup>	m m <sup>-3</sup>
				Layer 1 -			
Signalgrass	1.06 a	60.94 a	1.99 a	96.55 a	0.308 a	0.7260 a	4976.4 a
Traditional	1.04 a	58.06 b	1.63 b	77.89 b	0.222 b	0.7202 a	2950.8 b
CV (%)	6.62	3.65	9.34	12.54	9.80	4.33	11.65
				Layer 2			
Signalgrass	1.03 a	61.88 a	0.918 a	49.95 a	0.118 a	0.6592 b	2229.2 a
Traditional	1.03 a	60.06 b	0.574 b	41.76 b	0.110 a	0.7304 a	1596.8 b
CV (%)	5.74	1.84	28.48	15.08	12.77	5.56	10.64
				Layer 3			
Signalgrass	0.99 a	63.22 a	0.554 a	26.76 a	0.062 a	0.7021 b	1092.6 a
Traditional	0.99 a	62.34 a	0.565 a	28.70 a	0.076 a	0.7469 a	1246.2 a
CV (%)	6.32	3.36	15.94	18.43	23.72	6.24	18.35
				Layer 4			
Signalgrass	0.97 a	63.86 a	0.544 a	18.96 a	0.050 a	0.7043 a	856.2 a
Traditional	0.98 a	62.79 a	0.334 b	13.72 a	0.038 a	0.7065 a	736.8 a
CV (%)	6.21	3.07	26.13	25.31	22.83	7.00	16.89

CV coefficient of variation. Means followed by the same letter in the column, by depth, do not differ by the Tukey test at 5% probability; layer 1 (0 – 0.2 m); layer 2 (0.2 – 0.4 m); layer 3(0.4 – 0.6 m); layer 4 (0.6 – 0.8 m)

Dalmago et al. (2009) observed higher water availability in the 0 to 7.5 cm layer under no-tillage system (NTS) compared to conventional planting system (CPS) in an Argissolo with 30% clay and associated this difference to the presence of matter organic matter added to the soil surface by the first system. On the other hand, in the layers below these authors observed a greater availability of water in CPS and attributed this fact to the incorporation of the straw of oats and vetch grown in the winter. More recently, it has been found that organic compounds can improve soil-waterplant relationships by modifying soil density, total porosity and soil water balance (SULTANI et al., 2007), especially at low suctions, since soil moisture content increases linearly with increasing soil organic matter content (BRAIDA et al., 2010).

In layer 2, there was a significant increase in all related measures due to signalgrass, except for mean root diameter, which showed a decrease of -9.7% and no significant in the layer 1. This trend for RD is also true for the layer 3. This may indicate a greater presence of signalgrass

roots that are more branched, thin and grow more abundantly in subsurface than coffee tree roots. While root dry matter (RDM) in the signalgrass system of layer 1 is twice that observed in layer 2, in the traditional system the same measure corresponds to triple when comparing the layers.

The other measures related to roots showed a significant increase, indicating a higher presence and root activity, which may influence soil organic matter and, consequently, a greater water availability and perhaps microrganism activity in this layer, besides the alteration of soil aggregates. Loss et al. (2011) working on a dystrophic Red Latosol with more than 60% of clay and two cultivation systems, where in one of them there was the presence of signalgrass, observed higher aggregate stability indices (weighted mean diameter and geometric mean diameter) of light organic matter and mass of stable aggregates and assign these effects to the roots of that forage. These results corroborate with the present work since these evaluated characteristics favor soil water.

On the other hand, in layer 1 (Table 4), the presence of signalgrass significantly altered TP. RDM, RA, and RL in 5.0%, 22.1%, 24.0% and 68.6%, respectively, in addition to root volume (RV) in 38.7%. However, none of the parameters presented significant differences, either in the full or in the reduced linear regression models (Table 2), indicating that the generated models do not differ from each other, as a function of management systems. This result demonstrates that the changes in the layer due to the presence of signalgrass were not sufficient to significantly alter the water availability of this layer, as can be observed in Table 3, demonstrating that there were no changes in the TWA of layer 1 due to the use of signalgrass.

In deeper layers, 3 and 4, thus more structurally preserved, the management system with signalgrass increased TWA by 8.8% and 4%, respectively, without, however, changing the significance of the calibration curves as a function of management system. The layers just below the soil surface, with lower amounts of organic matter, may present substantial aggregate formation after the entry of organic carbon and can, therefore, be responsive to the root characteristics colonizing the soil (POIRIER; ANGERS; WHALEN, 2014).

### **4 CONCLUSIONS**

The Model Identity Method allowed the verification of equality of linear regressions resulting from neutron probe calibration in different soil layers and management systems of coffee rows in the Brazilian Central Cerrado.

The Model Identity Method allowed to identify the need for differentiated calibration curves of the neutron probe in the different soil layers and management systems.

The method justified the need for neutron probe calibration for each specific local condition.

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# INITIAL PRODUCTIVE PERFORMANCE OF COFFEE PROGENIES IN AN AREA INFESTED BY Meloidogyne paranaensis

Henrique Frederico Santos<sup>1</sup>, Sonia Maria de Lima Salgado<sup>2</sup> Antônio Nazareno Guimarães Mendes<sup>3</sup>, Alex Mendonça de Carvalho<sup>4</sup>, Cesar Elias Botelho<sup>5</sup>, Vinicius Teixeira Andrade<sup>6</sup>

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**ABSTRACT:** In this study, we aimed to carry out a selection of *Coffea arabica* progenies in areas infested by *Meloidogyne paranaensis* in order to select materials that are resistant to this nematode, with desirable agronomic characteristics. The experiment was set on *Guaiçara* Farm, located in the Municipality of *Piumhi* – *MG*, in February 2012. Twenty-one coffee progenies were evaluated in the F5 generation and 5 commercial cultivars were used as a control. A randomized complete block design was used, with three replicates, totaling 78 plots, consisting of eight plants each. The spacing used was 3.00 x 0.50 m in the rows and between plants, respectively. The populations of *M. paranaensis* per gram of coffee roots, productivity (bags. ha<sup>-1</sup>), plant vigor, maturation cycle, maturation uniformity, coffee ranking by grain size (sieve 17 and above), and classification of mocha coffee were evaluated. It was concluded that progenies MG 0179-3-R1-151 and MG 0185-2-R2-132 are resistant to *M. paranaensis* and have good agronomic characteristics in an area naturally infested by this nematode, thus indicated for plantations in this situation. Nine progenies reveal tolerance/resistance characteristics to *M. paranaensis*.

Termos para indexação: Coffea arabica, production, nematode, resistance, amphillo.

# PERFORMANCE PRODUTIVA INICIAL DE PROGÊNIES DE CAFEEIRO EM ÁREA INFESTADA POR Meloidogyne paranaensis

**RESUMO**: Objetivou-se neste trabalho selecionar progênies de *Coffea arabica* em área infestada resistentes a esse nematóide, com características produtivas desejáveis. O experimento foi instalado na Fazenda Guaiçara, situada no Município de Piumhi-MG. Vinte e uma progênies de cafeeiro em geração F<sub>5</sub> e cinco cultivares comerciais (testemunhas) foram avaliadas por meio da população de *Meloidogyne paranaensis*, produtividade de um biênio (sc.ha<sup>-1</sup>), vigor vegetativo, ciclo de maturação, uniformidade de maturação, classificação do café por tamanho do grão (peneira 17 e acima) e classificação do café moca. As progênies MG 0179-3-R1-151 e MG 0185-2-R2-132 apresentaram resistência a *M. paranaensis* e boas características agronômicas em área naturalmente infestada por esse nematoide, sendo indicadas para plantios nessa situação. Nove progênies apresentaram características de tolerância ao *M. paranaensis*.

Termos para indexação: Coffea arabica, produção, nematoide, resistência, amphillo.

### 1 INTRODUCTION

Meloidogyne spp. nematodes stand out as a limiting factor for coffee producing regions (SALGADO; REZENDE; NUNES, 2014). In Brazil, the main species harmful to coffee are Meloidogyne exigua Göldi1887 Meloidogyne incognita (Kofoid & White) Chitwood 1949 and Meloidogyne paranaensis Carneiro et al. 1996 (CAMPOS; VILLAIN, 2005).

M. paranaensis is one of the most harmful root-knot nematodes to the coffee tree, since it drastically reduces the root system and, consequently, plant development and productivity. This nematode is distinguished from other species by its aggressiveness and strong damage to the

coffee root system, with a high degree of plant degradation, commonly culminating in its death (CARNEIRO et al., 2008; SALGADO et al., 2014). The roots parasitized by *M. paranaensis* show peeling and cracking, with some thickening points that show canker lesions and decortication (CASTRO et al., 2008). Lopez-Lima et al. (2015) called 'corky-root' the damage caused by *M. paranaensis*, first diagnosed in coffee trees in several regions of Mexico.

The main strategy for the management of phytonematodes is prevention, that is, to prevent the entry and dissemination of the nematode in the area it occurs through infested seedlings, contaminated soil adhered to machines and

<sup>&</sup>lt;sup>1,3</sup>Universidade Federal de Lavras/UFLA - Departamento de Agricultura/DAG - Cx.P. 3037-37.200-000 - Lavras - MG-henriqueagro7@gmail.com,nazareno.ufla@hotmal.com

<sup>&</sup>lt;sup>2,5</sup> Empresa de Pesquisa Agropecuária de Minas Gerais/EPAMIG - Campus UFLA-Cx. P. 3037 - 37.200-000 - Lavras - MG-cesarbotelho@yahoo.com.br,soniamaria@epamig.ufla.br

<sup>&</sup>lt;sup>4</sup>Universidade Estadual Paulista "Júlio de Mesquita Filho"/UNESP - Campus de Registro - 11.900-000 - Registro - SP - alexcarvalho@registro.unesp.br

<sup>&</sup>lt;sup>6</sup>Instituto Agronômico de Campinas/IAC - 13.020-902 - Campinas - SP - viniciustandrade.vta@gmail.com

implements and mainly runoff water from plots or areas already infested (SALGADO; REZENDE, 2014). Cultural, biological and chemical management have been used as a strategy to reduce the nematode population (GOLÇALVES et al., 1996). However, all these strategies have low efficiency when compared to genetic control, which has been shown to be more efficient and economically viable (ITO et al., 2008).

The sources of resistance are present in some species of the genus *Coffea*, such as *Coffea canephora* Pierre ex A. Froehner *Coffea congensis* A. Froehner (SERA et al., 2007) and *Coffea dewevrei* De Wild. & T. Durand (KANAYAMA et al., 2009). Salgado et al. (2014) observed partial resistance of the Amphillo variety to breed 2 of *M. exigua*. For *M. paranaensis*, these genotypes showed good initial plant behavior in infested areas.

Studies on the behavior of coffee genotypes in relation to *Meloidogyne* spp., in majority, have been conducted under greenhouse conditions. However, there is little research on genetically improved materials in areas infested with M. paranaensis (REZENDE et al., 2013). Even if it requires more time, the selection work carried out in the field is necessary and extremely important for breeding programs, since the data on the performance of coffee progenies become more reliable (OLIVEIRA et al., 2011). Salgado et al. (2014) verified that some genotypes obtained in the active germplasm bank (BAG-Café) of Empresa de Pesquisa Agropecuária de Minas Gerais - EPAMIG present promising initial behavior in an area infested by M. paranaensis in the municipality of Piumhi, Minas Gerais.

In this study, we aimed to carry out a selection of *Coffea arabica* L. progenies in areas infested by *M. paranaensis* in order to select materials that are resistant to this nematode, with desirable agronomic characteristics.

### 2 MATERIAL AND METHODS

### Research installation

The selected experimental area presents a high infestation of *M.paranaensis*, a species identified by electrophoresis (CARNEIRO and ALMEIDA, 2001). The area is located in the Southwest region of Minas Gerais, in the municipality of Piumhi, Fazenda Guaiçara, privately owned, located at 20° 25' 28,7" South latitude, 46° 1' 10,5" longitude and average altitude of 812 m, soil with clayey texture and flat terrain. The average annual temperature is 20.7°C.

From the 42 *C.arabica* progenies in generation  $F_4$  evaluated by Salgado et al. (2014), 21 progenies in the  $F_5$  generation were planted in February 2012, together with five commercial cultivars used as controls (Table 1) in a randomized complete block design with three replicates, totaling 78 plots, consisting of eight plants each. The spacing used was 3.00 x 0.50 m between rows and between plants, respectively.

Phytosanitary management was carried out preventively or curatively, using chemical products, accompanying the seasonality of the occurrence of pests and diseases. The chemical control of the nematode in the area was not carried out, aiming at the identification and selection of progenies resistant to these pathogens.

# Quantification of *M. paranaensis* population in coffee roots and population biological indicator test in the plot soil

Aiming the quantification of *M. paranaensis* population, root samples were removed in October 2015, after the first plant production, avoiding to hinder the initial development of coffee trees. The samples were collected at a depth of approximately 30 cm (Freitas et al., 2007), at four plot points (sub-samples), in the projection of the plant crown, forming a composite sample conditioned in a plastic bag. The nematode extraction from these samples occurred according to the Hussey and Barker (1973) method to obtain the suspension of second-stage juveniles and eggs (J2) of *M. paranaensis*, which were quantified using a counting slide under an inverted objective microscope.

The biological indicator test (Bioteste) used soil samples taken from each experimental plot in trays with 100-mL cells, duly identified according to the representative field plot. A 'Santa Clara' tomato seedling per cell was used as a biological indicator of *M. paranaensis* population in the plot soil. This biotest was evaluated by the quantification of *M.paranaensis* population (eggs + second-stage juveniles - J2) in tomato roots at 50 days of planting.

### Progeny performance in the infested area

The agronomic characteristics evaluated in the first two harvests, 2014/2015 to 2015/2016, represent: production in liters of "bica corrida" coffee, considering an average yield of 480 liters of "farm coffee" for each 60-kg bag of processed coffee.

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**TABLE 1** - Progeny relation and genealogy in generation F5 and of the five commercial cultivars evaluated in the municipality of Piumhi – MG, in an area naturally infested by *Meloidogyne paranaensis*.

N°	Progenies	Origin
1	MG 0185-1-R2-847	C. V. X Amphillo MR 2-474
2	MG 0179-1-R1-776	C. V. X Amphillo MR 2-161
3	MG 0185-1-R2-849	C. V. X Amphillo MR 2-474
4	MG 0185-1-R2-138	C. V. X Amphillo MR 2-474
5	MG 0179-3-R1-151	C. V. X Amphillo MR 2-161
6	MG 0185-1-R2-1182	C. V. X Amphillo MR 2-474
7	MG 0179-1-R1-91	C. V. X Amphillo MR 2-161
8	MG 0185-1-R2-850	C. V. X Amphillo MR 2-474
9	MS Resplendor	Catuai IAC 86 x HT UFV 440-10
10	MG 0179-1-R1-90	C. V. X Amphillo MR 2-161
11	MG 0185-1-R2-132	C. V. X Amphillo MR 2-474
12	MG 0176-9R2-1232	Amphillo MR X H. N. MR 36/349
13	Paraíso MG H 419-1	Commercial cultivar / EPAMIG
14	Mundo Novo IAC 379-19	Commercial cultivar / IAC
15	MG 0185-1-R2-139	C. V. X Amphillo MR 2-474
16	Catuai Amarelo IAC 62	Commercial cultivar / IAC
17	MG 0179-1-R1-1052	C. V. X Amphillo MR 2-161
18	MG 0179-1-R1-89	C. V. X Amphillo MR 2-161
19	MG 0179-5-R1-1004	C. V. X Amphillo MR 2-161
20	MG 0185-1-R2-137	C. V. X Amphillo MR 2-474
21	MG 0185-1-R2-1176	C. V. X Amphillo MR 2-474
22	MG 0179-1-R1-775	C. V. X Amphillo MR 2-161
23	MG 0179-1-R1-1051	C. V. X Amphillo MR 2-161
24	MG 0294-1-R1-342	Hibrido de Timor 408-01
25	MG 0176-8-R1-943	Amphillo X H. N. MR 36/349
26	H504-5-8-2	Catuai IAC 81 x HT UFV 438-01

After coffee processing in 2015 and 2016, a 300-g sample was passed through a set of sieves (8 to 13), and the material was retained in each sieve, determining the percentage of mocha grains (BRASIL, 2003); plant vigor, evaluated by assigning scores according to an arbitrary 10-point scale, with score 1 given to the worst plants, with very reduced plant vigor and marked degradation symptoms, and score 10 to plants with excellent vigor, more unveiled and with marked plant growth of productive branches, as suggested by Carvalho

et al. (1979). In 2016, in the evaluation of the maturation cycle, scores were assigned according to an arbitrary 5-point scale, in which 1 = early; 2 = average to early; 3 = average; 4 = average to late and 5 = late; for maturity uniformity, scores were assigned according to an arbitrary 4-point scale, where 1 = uniform; 2 = moderately uniform; 3 = moderately uniform and 4 = uniform.

Data on the reproductive development of coffee plants and on progeny reaction to the nematode were submitted to analysis of variance by the statistical software SISVAR (FERREIRA, 2008).

From the detection of significant differences between treatments, the means were grouped by the Scott Knott test, at 5% probability. The data transformations  $(\log(x))$  and  $(\sqrt{x+1})$  were used for data on nematode populations in coffee (PGR) and tomato roots (PGRBIOTESTE).

### **3 RESULTS AND DISCUSSION**

The nematode population in coffee roots had a significant difference among progenies in two distinct groups: *M. paranaensis* population below 62 nematodes/g root in group G1, consisting of 15 progenies and genotype HT UFV 408-26; in group G2 are 6 progenies and 'Catuaí Amarelo' IAC 62, 'Mundo Novo' IAC 379-19, 'MS Resplendor' and 'Paraíso' MG H 419-1, with higher values of egg + second-stage juvenile population (J2) of the nematode (Table 2).

The biotest presents a significant difference for *M. paranaensis* population in the plots, indicating a variation in the parasitic population of 2.89 in the progeny soil for progeny 23 (MG 0179-1-R1-1051), up to 1264 individuals in progeny 12 (MG 0176-9-R2-1232), (Table 2).

For the nematode population in coffee roots, 16 treatments had smaller egg and second-stage juvenile populations (J2). Commercial cultivars used as susceptible controls had a high nematode population (Table 2).

According to Salgado, Rezende and Nunes (2014), the plant resistance mechanism to nematodes prevents high reproductive rate from nematode and, consequently, besides not causing damage to the crop, they lead to a decrease in its population in the soil. This explains the results of the biotest, when compared with the evaluations of the nematode population in coffee roots.

The average yields estimated in the biennium 2014/2015 and 2015/2016 ranged from 20.25 sc ha<sup>-1</sup> (treatment 16) to 82.60 sc ha<sup>-1</sup> (treatment 11), and approximately 40% of the progenies had a performance higher than the global average, suggesting a resistance/tolerance of these progenies to *M. paranaensis*. Progenies 4, 5, 11 and 12 had an average yield of 251.30%, higher than the 5 susceptible controls (Table 3).

The superiority in progeny production, associated with *M. paranaensis* population in the roots, indicates a resistance behavior of progenies 4 and 11 and a tolerance behavior of progenies 5 and 12 since, according to Trudgill (1991), tolerance refers to the inherent or acquired capacity of a plant to support pathogen infection (fungus, viruses,

nematodes, bacteria, etc.), without significant damage to its production. On the other hand, the controls represented by susceptible commercial cultivars had low yield (Table 3), confirming the economic damage of this nematode. This marked reduction in yield indicates how much damage *M. paranaensis* can cause to susceptible cultivars, as a consequence of parasitism, with the destruction of the root system, which may cause general degradation.

Regarding vigor scores, the averages were grouped in two classes by the Scott-Knott test at 5% probability, with 10 treatments that showed higher scores. The most vigorous genotypes are 4, 5, 6, 10, 11, 12, 15, 21, 25 and 26, with average vigor scores ranging from 5.12 to 8.0 (Table 3). The susceptible controls obtained the lowest scores, on average, evidencing the impairment of their plant development in the presence of nematodes. In addition to influencing production, according to Carvalho et al. (2012) and Carvalho et al. (2008), plant vigor reduces, to a certain extent, coffee bienniality and is directly related to genotype yield and adaptability to the different edaphoclimatic conditions.

Progenies 4, 10 and 11 showed a low M. paranaensis population in coffee roots, good plant vigor and high yield in the biennium. This indicates that these progenies have the potential for continuation in generation advance and/or backcrossing within the coffee breeding program of EPAMIG. The smallest M. paranaensis population in the roots of these progenies can be attributed to Amphillo, since this genetic material is a possible source of resistance to M. paranaensis. Salgado et al. (2014) identified F plants derived from the crossing between 'Catuai Vermelho', 'Amphillo' MR 2161 and 'Híbrido de Timor' UFV 408-01 as promising, potentially resistant to M. paranaensis and, therefore, important to the coffee breeding program. Silva et al. (2015) observe that progeny 5, from the cross of 'Catuaí Vermelho' and 'Amphillo' 2-161, whose plant in the F<sub>4</sub> generation supplied progeny 5 with seeds in this research, had a capacity for acclimatization to the water deficit, as evidenced by the maintenance of water status, gas exchange and better photochemical performance, culminating in higher yield. Studies of these genotypes in areas infested by other Meloidogyne species should be carried out, as there is the possibility of obtaining coffee cultivars with resistance to several rootknot nematode species (Boisseau et al., 2009).

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**TABLE 2** - *Meloidogyne paranaensis* population (eggs + second-stage juveniles - J2) per gram of *Coffea arabica* progeny roots (PGR) and per gram of 'Santa Clara' tomato roots (PGR Bioteste), cultivated with soil removed from the rhizosphere of coffee plants at 30 months of planting in an experimental area located in Piumhi, MG.

Treatment	PGR	PGR Biotest
1	25.77 a	81.64 a
2	62.36 b	85.46 a
3	173.07 a	43.33 a
4	13.30 a	47.21 a
5	68.54 a	164.88 a
6	139.38 b	10.28 a
7	10.36 a	69.65 a
8	5.04 a	8.16 a
9*	194.14 b	123.99 a
10	17.23 a	33.46 a
11	23.28 a	70.47 a
12	68.85 a	1264.18 b
13*	219.65 b	1067.26 b
14*	171.28 b	740.15 b
15	278.82 b	128.90 a
16*	103.39 b	399.21 b
17	254.29 b	382.44 b
18	20.04 a	14.26 a
19	8.07 a	17.77 a
20	191.7300 b	144.95 b
21	148.7333 b	208.18 b
22	5.2900 a	51.91 a
23	9.8800 a	2.89 a
24	63.2266 b	316.15 b
25	95.7900 a	337.18 b
26*	97.8200 a	27.69 a
Mean	94.9773	224.6816
CV(%)	36.53	61.54

<sup>\*:</sup> Controls. Means followed by the same letter, vertically, belong to the same group, by the Scott-Knott test at 5% probability.

Progenies MG 0179-3-R1-151 (5), originated from the cross between C. V. X Amphillo MR 2-161 and MG 0185-2-R2-132 (11), coming from the cross between C. V. X. Amphillo MR 2-474, were classified as resistant by Pasqualoto (2015) and Peres (2013), respectively, corroborating the results obtained in this study. Pasqualoto (2015) evaluated progenies in infested areas and greenhouse environments, and Peres (2013) evaluated the reproductive factor of *M. paranaensis* and *M. incognita*, breed 1, in a greenhouse and compared the results with those obtained from areas infested with *M. paranaensis*.

Progenies MG 0185-2-R2-138 (4), from the cross between C. V. X Amphillo MR 2-474 and MG 0176-2-R2-943 (25), obtained from the crossing of Amphillo X. H. N. MR 36/349, were

pointed out by Peres (2013) as moderately resistant to M. paranaensis, and the latter was highlighted by Pasqualoto (2015) as potentially resistant.

Treatment 23 presents good yield and low nematode population in the roots, but with low vigor. However, Pasqualoto (2015) concluded that this progeny, MG 0179-1-R1-1051, from the cross between C. V. X Amphillo MR 2-161, is resistant to *M. paranaensis*.

Progenies 10, 18, 19 and 22 can be considered of intermediate performance, according to plant yield and vigor. Peres et al. (2017) concluded that plants of progenies MG 0179-1-R1-90, MG 0179-1-R1-89 and MG 0179-1-R1-775, from which seeds were obtained for treatments 10, 18 and 22 of this study, respectively, were resistant to the nematodes *M. paranaensis* and *M. incognita*.

**TABLE 3** - Estimated average yield, in sc/ha, in the biennium 2015/16, vigor of coffee progenies evaluated in an area infested by *Meloidogyne paranaensis* in Piumhi, MG.

Progenies	Yield	Vigor
1	40.27 b	3.91 b
2	38.48 b	4.58 b
3	25.95 b	4.12 b
4	74.21 a	5.91 a
5	78.69 a	6.66 a
6	58.44 a	5.86 a
7	28.64 b	4.37 b
8	33.85 b	3.87 b
9	39.23 b*	3.79 b*
10	44.41 b	6.83 a
11	82.60 a	8.00 a
12	64.81 a	5.50 a
13	31.30 b*	3.25 b*
14	27.95 b*	3.08 b*
15	56.85 a	6.33 a
16	20.25 b*	2.46 b*
17	30.26 b	3.75 b
18	45.85 b	4.83 b
19	48.17 b	3.75 b
20	59.60 a	4.75 b
21	55.33 a	5.50 a
22	46.43 b	4.71 b
23	59.59 a	4.96 b
24	24.45 b	2.63 b
25	55.11 a	5.92 a
26	51.50 a	5.12 a
Means	46.9726	4.7896
CV(%)	35.47	31.99

<sup>\*:</sup> Cultivars used as controls. Means followed by the same letter, vertically, belong to the same group, by the Scott-Knott test at 5% probability.

In the evaluation and selection of coffee plants, researchers seek an ideotype whose performance includes, besides other characteristics, high yield and percentage of grains classified in high sieves (FERREIRA et al., 2005). In general, increased grain size provides greater uniformity of the batch to be processed and directly influences the physical appearance of the product, which is desirable, mainly for use in espresso machines (FERREIRA et al., 2013). For this characteristic, it was observed that three groups were formed, and only 'Mundo Novo' IAC 379-19 was in the superior group; in the intermediate group, there were 6 treatments, with 5 progenies and 'Catuai Amarelo' IAC 62 and, in the inferior group, 19 treatments, with 16 progenies and 'MS Resplendor', 'Paraíso' MG H 419-1 and HT UFV 408-26.

Mocha beans originate under lack of fertilization conditions or even when fruit development occurs in one of the stores GASPARI-PEZZOPANE et al., 2005). Carvalho et al. (2009) define mocha beans as grains that have ovoid shape, with a crack in the longitudinal format. The evaluation of their average percentage in the biennium 2014/2016 had two classes, and only three progenies with percentages above the average (Table 4). In addition to the percentage of mocha, grain size (% sieve 17 and above) may have been affected by rain scarcity in the period, compromising plant nutrition, flower setting and fruit granulation. During this period, there was a water deficit of approximately 500 mm in relation to the historical average (Table 4).

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**TABLE 4** - Mean values of sieve 17 and above, mocha grains, maturation cycle and maturity uniformity evaluated in the 2014/2015 to 2015/2016 harvests in an area naturally infested by *Meloidogyne paranaensis*, in the municipality of Piumhi-MG.

Treatment	Sieve 17	Mocha	Maturation	Maturity uniformity
1	13.61 c	21.16 a	1.61 a	1.82 a
2	27.02 b	24.90 a	3.33 a	3.21 b
3	24.28 b	32.49 b	2.72 a	2.65 a
4	7.38 c	18.01 a	2.89 a	3.54 b
5	17.85 c	19.12 a	3.22 a	3.41 b
6	11.96 с	27.30 b	3.62 a	3.00 b
7	27.98 b	18.08 a	2.44 a	2.51 a
8	15.60 c	22.35 a	2.55 a	2.86 a
9	8.16 c*	15.12 a*	2.40 a*	2.89 a*
10	16.22 c	16.78 a	2.44 a	2.16 a
11	9.78 c	14.53 a	3.37 a	3.96 b
12	21.99 b	25.42 b	3.23 a	3.70 b
13	14.06 c*	16.51 a*	2.77 a*	2.71 a*
14	36.51 a*	19.40 a*	2.39 a*	2.22 a*
15	12.53 c	15.22 a	2.88 a	3.15 b
16	20.21 b*	18.02 a*	2.56 a*	2.76 a*
17	8.36 c	20.89 a	2.10 a	2.27 a
18	23.35 b	19.31 a	2.36 a	2.06 a
19	14.84 c	18.46 a	1.78 a	2.70 a
20	10.16 c	18.79 a	2.85 a	3.52 b
21	8.21 c	19.15 a	3.10 a	2.75 a
22	7.72 c	22.23 a	2.82 a	3.10 b
23	13.67 с	17.37 a	2.99 a	3.25 b
24	14.26 c*	20.88 a*	3.33 a*	3.50 b*
25	13.95 с	19.55 a	2.82 a	3.31 b
26	12.79 c	18.47 a	3.09 a	3.76 b
Mean	15.8664	19.9843	2.7597	2.9557
CV(%)	36.35	24.50	24.13	18.30

<sup>\*:</sup> Cultivars used as controls. Means followed by the same letter, vertically, belong to the same group, by the Scott-Knott test at 5% probability.

For maturation season, the treatments did not present significant differences, occurring concurrently in 'Catuaí Amarelo' IAC 62, 'Mundo Novo' IAC 379-1, 'MS Resplendor', 'Paraíso' MG H 419-1 and HT UFV 408-26; the maturation cycle was classified as medium.

As for the average maturity uniformity scores, two distinct groups were formed: 9 progenies and the controls 'Catuaí Amarelo' IAC 62, 'Mundo Novo' IAC 379-1, 'MS Resplendor', 'Paraíso' MG H 419-1 were in the first group, with average scores ranging between 1.82 and 2.89, classified as moderately uniform; the other 12 progenies and cultivar HT UFV 408-26 make up the other group, with average scores ranging from 3.00 to 3.96, classified as moderately uneven to uneven.

### **4 CONCLUSIONS**

Progenies MG 0179-3-R1-151 and MG 0185-1-R2-132 were resistant to *M. paranaensis* and had good agronomic characteristics in an area naturally infested by this nematode, being indicated for plantations in this situation.

Progenies MG 0185-1-R2-138, MG 0179-1-R1-90, MG 0176-9-R2-1232, MG 0179-1-R1-89, MG 0179-5-R1-1004, MG 0179-1-R1-775, MG 0179-1-R1-1051, MG 0176-8-R1-943 and MG 0294-1-R1-342 present tolerance characteristics to *M. paranaensis*.

The high yield of progeny MG 0185-1-R2-132, above 80 sc/ha, evidences its great productive potential in areas infested with *M. paranaensis*.

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## THE CONTINUAL IMPROVEMENT IN THE CERTIFICATION OF COFFEE FARMS: A CASE STUDY

Claúdio Vieira Castro<sup>1</sup>, Jean Marcel Sousa Lira<sup>2</sup>, Eduardo Gomes Salgado<sup>3</sup>, Luiz Alberto Beijo<sup>4</sup>

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**ABSTRACT:** The certification of agricultural products is considered a strategy that promotes sustainability and appreciation of product. In coffee production, the *Certifica Minas Café* is the only public coffee certification program in Brazil. However, over the years, many coffee farms that have this certification reduce their competence to meet the requirements, stabilizing at minimum levels of compliance, which allows maintaining the certificates, but does not promote continual improvement. Therefore, this study aimed to evaluate the effect of *Certifica Minas Café* certification on the improvement of sustainability over time on coffee farms. For this, a case study with two properties in the south of the state of Minas Gerais was carried out in which one of them quest to not achieve continuous improvement, while the other does. On the coffee farm of the first case was observed a reduction of number of requested fulfilled by the certification program over time. While coffee farm of the second case there was a reduction in investment and low concern to maintain the requirements established by the certification. Lastly, this paper hypothesizes that improvement of sustainability depends on understanding the benefits of certification in the long term and is related to a behaviour change by the certified farm.

Index terms: Certifica Minas Café, improvement of sustainability, coffee production.

# A MELHORIA CONTÍNUA NA CERTIFICAÇÃO DAS FAZENDAS DE CAFÉ: UM ESTUDO DE CASO NO SUL DE MINAS GERAIS

RESUMO: A certificação de produtos agrícolas é considerada uma estratégia que promove a sustentabilidade e a valorização do produto. Na produção de café, o *Certifica Minas Café* é o único programa público de certificação de café no Brasil. No entanto, ao longo dos anos, muitas fazendas de café que possuem esta certificação deixam de atender vários dos requisitos reduzindo o nível de conformidade, o que as permite manter a certificação, porém não promove uma melhoria contínua. Portanto, este estudo teve como objetivo avaliar o efeito da certificação *Certifica Minas Café* na melhoria da sustentabilidade ao longo do tempo nas lavouras de café. Para isso, foi realizado um estudo de caso com duas propriedades no sul do estado de Minas Gerais, em que uma delas não buscou a melhoria contínua, enquanto a outra buscou. No primeiro caso de estudo foi observado que a fazenda reduziu o número de exigências solicitadas pelo programa de certificação ao longo do tempo. Enquanto no segundo caso a fazenda sofreu uma redução no investimento e demonstrou pouca preocupação em manter os requisitos estabelecidos pela certificação. Por fim, este artigo supõe que a melhoria da sustentabilidade depende do entendimento dos benefícios da certificação em longo prazo e está relacionada a uma mudança de comportamento da fazenda certificada.

Termos para indexação: Certifica Minas Café, melhoria da sustentabilidade, produção cafeeira.

### 1 INTRODUCTION

In 2006, the Government of the State of Minas Gerais established a certification program for coffee plantations, which stands out as the only one proposed and administered by the Government. Although, there are at least ten other certifications in Brazil for the coffee market, all of them of private character, such as the Rainforest Alliance, Organic, Nespresso AAA, 4C (Community Code Commonwealth), UTZ Certified, Fair Trade, Globalgap, Starbucks CAFE Practices and Brazilian Specialty Coffee Association.

This work had access to certification audit reports of a total of 1,347 properties that were certified in the criteria established by the *Certifica Minas Café* program, with all the scores

awarded to each property, from the first year of certification until the year 2015. The examination of the material reveals that, comparing the scores from the first year of certification and from 2015, a total of 337 properties had a decrease in the scores obtained; another 80 properties kept the same scores and 930 properties have increased scores since the first year of entry into the program. In the light of these preliminary data, two fundamental questions emerge: Why do some rural properties in certification, over the years, improve their socio-environmental performance, while other properties have a worsening of these same results? What factors may be contributing to positive or negative certification performance? These were the central questions that this study sought to answer.

<sup>1.2.3.4</sup> Universidade Federal de Alfenas/UNIFAL - Instituto de Ciências Exatas/ICEX Cx.P. 3037- 37.130-001 - Alfenas - MG claudio@vieiracastro.com.br, jmslira1283@gmail.com, eduardosalgado@bcc.unifal-mg.edu, luiz.beijo@unifal-mg.edu.br

The Certifica Minas Café regulations provide that the program is intended to establish a certification system for coffee plantations in the State of Minas Gerais that can be applied to all properties, regardless of location or possible differences in technology standards applied to productive processes (CASTRO; SALGADO; BEIJO, 2017). The proposal is to establish their own certification, with an independent verification system, but at the same time allowing the exchange of technologies and regulations with national and international entities related to certification of coffee, in order to encourage the adoption of good agricultural practices in the production. contributing to the safety and reliability of the coffee produced in the State (MINAS, 2009).

Upon entering the program, the rural property is able to adjust from an initial assessment made on the basis of specific requirements and through the monitoring of a technical staff provided by the State itself. Since the beginning of the program, certification requirements have been modified, added or suppressed, adapting to the parameters desired by the managing bodies. The intention evidenced through the evolution of the requirements is to promote a gradual resurgence of certification requirements, aiming to reach the desired quality standards, through a process of raising producers' awareness about the need to adapt and improve the coffee production activity. In the version adopted from 2016, there are 102 requirements:

- a) 26 requirements considered mandatory, evaluated with weight 3, whose non-compliance results in non-certification of the property;
- b) 55 restrictive requirements, evaluated with weight 2, whose non-compliance does not compromise the achievement or continuity of certification; and
- c) 21 recommended requirements, where certification occurs, regardless of compliance, but they are evaluated with weight 1 (MINAS, 2016a).

The criterion "Plantation" includes requirements related to propagation material, cultivation areas, pest and disease control, irrigation, and also requirements related to techniques adopted at harvest and post-harvest. In this criterion, appropriate conditions are required for coffee seedlings, field identification of the soil, soil fertility assessment, laboratory analysis for liming and fertilization recommendations, use of agrochemicals in accordance with prescriptions and technical recommendations, use of personal protection equipment, appropriate storage and disposal of packaging, records, training and grants for irrigation activities.

In the "Traceability" criterion, the aim is to record all stages of production, from the planting, harvesting, post-harvesting, storage and marketing of the coffee produced. Records need to be sufficient to identify the origin of the coffee, separating certified coffees from others that may be produced on the property.

criterion of "Environmental Responsibility", presents requirements related to compliance with environmental legislation, soil conservation and management of weeds (brushcutting, manual weeding, chemical control, among others), water, air (deforestation, fires), biodiversity (trade in wild fauna and flora species) and the appropriate disposal of waste generated on the property (including domestic sewage) and those residues resulting from agro-industrial and agricultural activities. The highlight is the requirements related to water conservation, with 37.03% of the total, requiring identification of springs that may exist in the property, adoption of protection practices for springs and measures to protect the areas considered as recharge (hill tops), prohibition of interventions in watercourses without the authorization of the competent bodies, re-use of the water from post-harvest processes and the proper disposal of wastewater from the washing or processing of coffee. Certification also intends to make the producer aware of the need to adopt water preservation and conservation measures.

In turn, the "Social Responsibility" criterion addresses requirements related to labour and health and occupational safety aspects, in a context where child labour, forced labour, and discriminatory practices of any kind and related to trafficking of people are strictly prohibited, and must be "excluded from and banished from the property" (MINAS, 2016a). In addition, certification aims to deal with the regularization of labour activities (registration of employees), appropriate transport, appropriate eating areas, appropriate sanitary facilities and access to health systems, identification of risk areas, adoption of internal commissions for the prevention of accidents (where applicable), periodic medical examinations. Certification also includes the adoption of measures, such as taking care of containers that can accumulate water, to prevent the occurrence of the Aedes aegypti (Linnaeus, 1762) mosquito, associated with diseases such as the dengue fever.

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On the other hand, the criterion "Training" has requirements related to the training of workers. These trainings are associated with work safety, pest and disease management, application of agrochemicals and operation of production equipment, such as tractors, harvesters, brush cutters, coffee stripping machines and chainsaws. It also includes the training of workers in the preparation, harvesting, drying, storage and processing of coffee.

Finally, certification presents the criterion called "Property Management", with only two requirements: the first one related to production cost control, where it is expected that the producer can annually evaluate the profitability of at least one field or tract of land; and the second requirement refers to the adoption of a methodology that allows the producer to handle complaints from interested parties if he develops the roasting activity and has his own brand of the coffee produced.

After the training period, the property is submitted to the certification audit, being obliged to meet 100% of the mandatory requirements and overall to at least 80% of the total requirements. If the property is disapproved, it can take corrective actions to obtain or maintain certification, which will only be cancelled in the event of not taking the required corrective actions. When the requirement is considered as compliant or noncompliant, it receives a score of 1 or zero and may be considered as not applicable. There is the possibility of imposing suspension penalties and even cancellation of certification, if producers fail to comply with the contractual rules, spread the word that the coffee produced has characteristics not included in the certification, such as informing that the coffee is organic, using the seals in uncertified coffees or the use of counterfeit seals or if contaminations are found in certified coffees (Empresa de Assistência Técnica e Extensão Rural

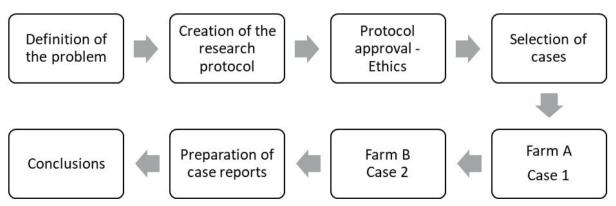
do Estado de Minas Gerais - EMATER, 2011). Certification maintenance audits are conducted annually at all properties participating in the program on previously scheduled visits.

### 2 MATERIALS AND METHODS

In order to respond to the inquiries of this work, the case study methodology was adopted, because it is appropriate for the intended scope, since it allows investigating a contemporary phenomenon under such conditions where, in the definition of Yin (2015) Figure 1 illustrates the steps that were implemented in the present study:

Based on the finding of differences in socioenvironmental performance among properties in the Certifica Minas Café, the research sought to identify two rural properties that were located in nearby geographic areas, with similar planting and productivity areas. One of the properties should show increasing performance, and the other, a decreasing performance, from the first year of certification. Once grouped according to these characteristics, the cases were chosen by drawings of lot. The proposal was to make a more homogeneous choice, avoiding that the differentiation factors of the properties, such as their location, size and technological standard of production could interfere in a relevant way in the performance analyses.

The research protocol involved conducting a semi-structured interview with the property manager (or the owner), based on a predefined questionnaire and analysis of the available audit reports on the property. The protocol was approved by the Ethics Committee (CAAE 50770215.0.0000.5142), and the interviews were submitted to terms of consent and authorization for the recording of voice and images.



**FIGURE 1** - Flowchart of the case study. Prepared by the authors, adapted from Yin (2015).

The study procedure provided for the maintenance by the authors of the strict confidentiality of personal data of participants and also of the data that could identify the rural properties involved, even in the hypotheses of publication of the data in specialized scientific journals. Because of this, in this study the chosen properties were identified as Farm A (did not continuous improvement) and Farm B (continuous improvement). The interviews and visits to the property occurred between the months of May and December 2016.

#### **3 RESULTS AND DISCUSSION**

Farm A was visited during the coffee harvest, at the beginning of May, 2016. The property started the certification process in 2012 and was certified for the first time in 2013. The farm has 60.6% of the property with coffee plantations, with an average yield of 37.78 bags per hectare. The property manager, who has been employed for 26 years and directly responsible for certification since its implementation, was interviewed.

Our study had access to scores obtained by Farm A, from 2013 to 2015. Figure 2 below indicates the reduction of socio-environmental performance of the property, based on the certification requirements.

In addition to the scores obtained in the certification, two audit reports on Farm A were analysed, the first in 2013, 98.3% of the requirements, and the second in 2015, 94.8% of the requirements. Table 1 below presents only those requirements that have changed, with the exception of item 5, in the assessment for the years indicated.

Thus, based on the percentages of compliance with the certification requirements, Farm A, over time, had a deterioration in its socioenvironmental performance.

Items 1, 3, 4, 6 and 8 of Table 2 were responsible for the reduction in the score obtained in 2015, when compared to that obtained in 2013. Items 2 and 7 revealed improvements, but not enough to prevent a decline in property performance. In item 5, there was a negative evaluation (noncompliance of the requirement) for the entire period considered. The property manager was asked to explain each of the items mentioned.

Regarding item 1, the manager clarified that the negative evaluation occurred due to the time at which the audit was performed. In 2015, they have been audited at the beginning of the harvest, when they started harvesting with more green grains,

"to avoid sweating coffee", but that this criterion was met at the end of the harvest, although the information was not evidenced. It was explained as a change of strategy used in recent years. However, the certification requirement is related to the beginning of the harvest and not to the end as clarified by the manager, who associated the practice to a greater use of the fruits, with reduction of the sweeping coffees. Certification allows a maximum of 30% of green grains, considered unripe because these grains do not have a balanced chemical composition, as they have not yet fully developed, which compromises the full development of the organoleptic properties of coffee and gives rise to drinks that are "harsh, astringent and with marked bitterness "and therefore with inferior quality to that drink prepared with ripe grains (GIOMO, 2012). Although not expressly stated by the manager, the practice of harvesting green beans indicates the preference for a quantitative harvesting of the crop, to the detriment of the qualitative aspect of the coffee produced.

The analysis of the presence of faecal coliforms in the water used in the post-harvest process (item 2) was an improvement evidenced by the audit carried out in 2015, after being designated as non-compliance in 2013. According to the manager, the activity was performed by a company specially hired for this purpose and the point highlighted in the interview was the additional cost of this hiring.

The cleaning of the facilities for processing and storage of the coffee beans before reuse (item 3) was not evidenced. According to the manager, it was just a case of forgetting about the requirement of recording the activity:

[...] It was not registered because we forgot. We did, but we did not record. We used to do the cleanings, but we did not have to record. We still do the cleaning, but we miss some details, because of the rush. We did not have an Internal Accident Prevention Committee (CIPA) set up yet, and there was only me and the crop supervisor. Today we have the workplace safety technician, who has the responsibility of making these records.

The same explanation was given for noncompliance with item 4. Farm A failed to renewal the environmental permit after its expiration, although the procedure is not considered by the manager as difficult to carry out. When questioned about the item, the manager replied that it is not complicated to renew the permit, "but we did not do it for lack of time", drawing a brief smile, denoting a certain embarrassment for the lack of a plausible explanation for not performing an activity required by certification.

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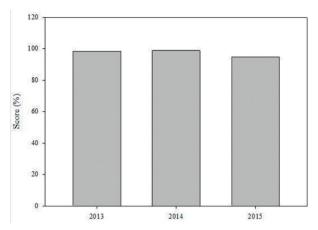


FIGURE 2 - Graphic of Annual scores obtained by Farm A, verified in the certification audits. Source: IMA MG.

**TABLE 1** - Farm A. Requirements with changes in the audits performed in 2013 and 2015

Item	Requirement	2013	2015
1	Harvesting should start when there is the least amount of green grains	X	
2	The inlet water in post-harvest processes should meet minimum faecal coliform standards		X
3	Processing and storage facilities must be sanitized before reuse	X	
4	The property must prove the existence of a permit or environmental authorization or a certificate of exemption from licensing	X	
5	Household waste must have appropriate treatment (use of septic tanks)		
6	Implementation of work safety training	X	
7	Training of manual trimmers operators		X
8	Training of chainsaw operators	NA	

Requirement completed is marked with X, Requirement not completed is empty and Not applicable (NA)

The requirement for the treatment of domestic sewage (item 5) through the use of septic tanks or other recommended treatments (digester, filters, among others) remains a problem in Farm A since the beginning of certification. The inappropriate disposal of domestic sewage occurs in most coffee farms, according to Araujo et al., (2016), in a research on the socio-environmental impacts of certifications in cooperatives of family producers in the states of Minas Gerais and Espírito Santo. In most cases, domestic sewage is taken to cesspools, "when not directly to watercourses," the authors point out. Farm A manager is fully aware of the requirement and the need for its implementation, but has not yet been able to allocate the financial resources required to execute the service. Farm A has 17 houses used by its employees and none of them has a septic tank.

We still do not have the septic tank, we have not done it yet. This makes it expensive, because of the construction, but we have not done it yet because of the time, which did not let us do. There is a lot of stone in the colony [referring to the place of residence of employees] and this makes it difficult to execute. The ones they have there are very shallow, but the boss has decided that he will do it. The *Certifica Minas Café* staff has been asking us for these tanks in all their visits. The boss is who decides when to do it. He said he will do it, but the time is passing and we are doing other things.

Items 6, 7 and 8 are all associated with worker training. Farm A stopped practicing work safety training, between 2013 and 2015 it carried out the training for the use of brush cutters and did

not carry out the training for the use of chainsaws in 2015, and in this item, in 2013 there was no the equipment on the property and therefore the evaluation was considered not applicable in that year. The manager explained that in the case of using chainsaws, a minimum of 8 participants is required for the training, but the farm has only two employees assigned to these activities and therefore did not do so, also claiming that the distance from the training has made the participation of employees impossible. Previous research has pointed out advances in the aspects related to the training of workers in certified farms, although the numbers are still inexpressive, pointing out irregularities in 55% of the properties certified by the Certifica Minas Café (SANTOS et al., 2015).

When asked to reflect on the impact of certification on the property, Farm A manager reported that Certifica Minas Café brought good results and improvements for employees, but that certification did not add value to the price of certified coffee, although the comparison of prices can reveal incorrect data, given the differences in demand between certified and non-certified coffees, as Barham and Weber (2012) have pointed out. The manager recognizes that several practices with the crop were only implemented in the property after certification, such as the appropriate programming and the technical recommendation for the application of agrochemicals. The traceability of coffee, in the manager's view, was a beneficial measure brought about by certification. But the focus of his analysis is geared especially to the final price aspect of certified coffee:

The boss says that so far it has not brought improvement in price. We'll have to get certified because everyone is doing it, but it needed to add value from 5% to 10%, which he commented to the bank staff a few days ago. He will continue, but it needed to add value. This gives a certain discouragement, because we spend more, we have to hire personnel, but it has no financial return.

[...] some things we leave behind because it does not have much importance, like a simple annotation, for example. Then we do the things that are more important.

The manager's explanation makes it clear that there is a selective attitude to the measures recommended by certification that will or will not be implemented in the property. The cost of the measures to be adopted is a relevant factor, but it is not the only one, because as presented in the

results presented some simple measures are no longer adopted because they have been forgotten or were not considered important by those who have the obligation to adopt the requirements of certification.

Farm B was visited during the off-season, at the end of November 2016. It began preparations for certification in 2009 and was certified for the first time in 2010. It occupies 56% of the area of the property with coffee plantations, with an average yield of 30 sacks per hectare. The interview was held at the property's headquarters with one of the partners, who is directly responsible for managing the certification.

Unlike the previous case, Farm B actively disseminates property certification, from the entrance gate to the various production and processing areas. The owner acquired the farm 10 years ago, when he began planting the first lands, with the purpose of making the property a model in the production of coffee. He has brought a business vision to management, results oriented, to achieve productivity, profitability and socio-environmental sustainability. In this sense, the owner understands that the implementation of certification was a very important dividing point for the management of the processes, improvement of the productive base and the quality of the coffee produced, being able to add all these aspects to the products and the people involved. The economic aspect related to the price of coffee is seen in another way:

[...] We were able to generate value to our business, to our product. So, I'm sure certification is not what you get on certified products, that's what's left over, because a certified property has a very efficient management ... and I felt it improved a lot, not only my life but the lives of those who work with me.

When asked to explain the gains from certification, the owner can list a number of topics by dividing them into environmental, social and economic aspects. In the first aspect, he points out that certification brought environmental licensing, the construction of septic tanks, the appropriate disposal of waste, the reuse of coffee straw in fertilization and the reutilization of wastewaters from the coffee washing in composting, with the use of bacteria. In the social aspect, the highlight is the intensive training of permanent employees (the property has four employees) and of those who are hired during the harvests (from 10 to 20 employees, depending on the harvest), emphasizing that investments in training and qualification are indispensable, since "You 545 Catro, C. V. et al.

do not make a special coffee without special people". And in the economic aspect, the owner emphasizes that certification adds value to the business, even allowing the export of the product, but understands that it is not the certification that ensures better prices, but the quality of the final product. Therefore, what drives the owner to stay certified is not the immediate economic result, but the combination of environmental, social and economic aspects. The view of the owner of Farm B contrasts with other works where the economic aspect was pointed out as the main driver of certification, being this the determining factor of the continuity of the producer in the programs, as Ibnu et al. (2015) stated when researching certified and non-certified coffee producers in Indonesia. From an economic perspective, the owner of Farm B explained his view as follows:

"What opens doors is not the *Certifica Minas Café*, but quality. When coffee has quality and is certified, then the producer is able to sell it for a higher price. Anyone who sees otherwise has difficulties and becomes less involved with certification. And when the producer has this resistance, he loses even the opportunity to open doors to other markets, he loses opportunities."

For Farm B, the improvement of production processes, involving crop planning, abandonment of practices that may compromise quality, such as the use of sweeping coffee and the cultivation of organic matter in the soil are also relevant, as they provide what is defined as "the emergence of a microclimate" in agriculture, which impacts the quality and sustainability of the business. To a certain extent, it is the same as mentioned by Leme and Gandia (2013) that have proposed an explanatory model for the analysis of the market for certified and special coffees and pointed to other benefits deriving from certification, besides price, such as the organization of work, which results in the greater control of the producer to his production system, with consequences in reducing costs and improving the quality of the final product. The owner of Farm B understands that all these benefits cited by Leme and Gandia (2013) are aggregated by certification and in a simple and direct way explains the partnership relationship that develops between the certifier and the property:

"Certification brings this vision. When you are not certified, you do not have this vision. It's as if you have a person, someone to whom you are subordinated to and have to render account. It is

a partnership relationship, because our results are theirs too. I celebrate all my scores. This year, we almost scored 10. We fell short ... "

The Research had access to the 2013 and 2015 reports of Farm B and access to the scores obtained from the first year of certification until 2016, when Farm B was visited. The data reflects the long-term vision of the enterprise. In seeking to achieve a sustainable productive arrangement, the results are gradually being incorporated into the business. The owner is aware that the major changes are behavioural.

Therefore, the challenge is precisely to make these changes in the way of producing be able to modify the behaviour of managers and employees, becoming routine in the business. Figure 3 below shows the clear improvement of property performance in the requirements, when observed all the certification time.

The changes indicated in the audit reports for 2013 and 2015 were consolidated in Table 2.

For the owner of Farm B, all nonconformities identified in certifications resulted in opportunities for improvement and were important to raise awareness of the need to be implemented and this implies changes in behaviour. For example, the use of personal protective equipment (PPE) where "employees do not like to use it, they feel uncomfortable, but they must be aware that it is important to use it for their safety". Therefore, he explained that some of the nonconformities occurred only after the certification audits warned about the need for improvements, not only because of the scores, but especially because of the results that these improvements have on quality of life, as occurred with the treatment of household waste (item 3) non-compliance indicated in 2013 and corrected in the audit of 2015. Weighing on the importance of certification to improve the quality of life of producers and communities is in harmony with the results obtained by Rueda and Lambin (2013) when assessing the potential of certification in Colombia to promote socioecological systems more resistant to the processes of globalization. The owner of Farm B explains his view as follows:

"That was an adjustment we had to make. First, we did at our employee's house because he lives there permanently. Then we did at the farm headquarters. It demanded time and logistics, more than cost. [...] If the producer understood that he is the one who gets the improvement, it would be much easier and he would be much more evolved in the certification processes. "

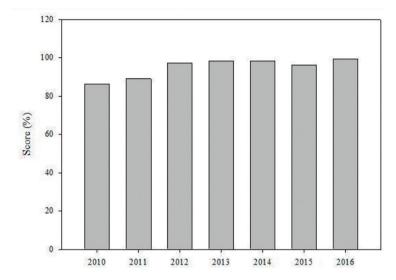


FIGURE 3 - Annual scores obtained by Farm B, verified in the certification audits.

**TABLE 2** - Farm B. Requirements with changes. Audits carried out in 2013 and 2015.

Item	Requirement	2013	2015
1	Harvesting should start when there is the least amount of green grains	X	
2	The inlet water in post-harvest processes should meet minimum faecal coliform standards	X	
3	Household waste must have appropriate treatment (use of septic tanks)		X
4	Implementation of work safety training		
5	Training in the integrated management of pests and diseases		

Requirement completed is marked with X, Requirement not completed is empty and Not applicable (NA)

The problem of the records is a major challenge to certification, according to the owner of Farm B, especially the record of activities that, prior to certification, were already routine in the management of the property, such as fertilization. The challenge, therefore, is to incorporate the requirements added by certification into the already established culture. This observation corroborates the results found by Vriesman et al. (2012), which, in the consolidation of 149 case studies involving technical assistance for the certification of organic products from family agriculture, pointed out the records as the main difficulty for producers. Therefore, among the processes implemented in Farm B is carrying out activities through work orders. All employees leave for farming with written work orders that were previously planned.

At the end of the day, they return with the orders filled out, informing what was done. The owner clarifies: "Our mission is to interpret these data and turn them into decisions," which takes time for this new culture to be fully absorbed by those involved.

The training activities, whether in work safety (item 4) or in the integrated management of pests and diseases (item 5) were carried out, reported the owner, but the evidence of its realization was lacking. Regarding item 5, the fault was attributed to service providers, who would have been hired specifically for this activity. In the view of the audit finding, the owner considered the possibility of termination of the outsourcing of those services, in the event of persistence of implementation flaws. What the owner wanted to

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emphasize was that behavioural change should affect not only the internal environment of the property (managers and workers) but that it should also extend to the contracted service providers, who should be guided on the importance of meeting the certification requirements

Regarding items 1 and 2, the owner was not able to answer what happened in the specific case, but he informed and presented the audit report where it was stated that the only non-compliance in 2016 was again the lack of evidence of training in the management of pests and diseases (item 5) and because of this item, it was considered that the property met 99.4% of all requirements of the standard.

"As we get involved in all activities, all without exception, we end up forgetting to ask for some items. So I told him [referring to the contracted technician for pest and disease management] that if he does not do it as required, he cannot stay with me because I cannot do everything. If he doesn't do it, I'll have to replace him. "

The last question of the interview with the owner of Farm B was related to the future of the property and the coffee industry itself, since he understands that certification is a "no-return path", and anyone who takes time to understand this may have difficulties in the future to reach those who came out first. And who is focused only on the immediate financial results, according to his vision, has not yet understood how to achieve higher prices for coffee produced, as pointed out in previous studies that verified the percentage of 60% of owners of certified coffee farms that understand certification as a necessity for the future and not for the present (BÓCOLI et al., 2013). The owner of Farm B explains his view as follows:

"The world is looking for quality coffees. The consumption of quality coffees is increasing. We are the only country in the world that can meet world demand for quality coffees. We have a market full of opportunities. We have many challenges too, such as climate change that are very cruel. So if we do not start doing differently, producing sustainably, we will have great difficulties ahead. We have to start now: in the choice of products, in the productive arrangements, and to know how to use all that we do as competitive advantages of our coffees. And this coffee will be worth much more. "

From the data obtained in this research it is possible to affirm that there is a distinct difference in the certified rural properties performance in terms of meeting the requirements required by certification. Table 3 illustrates the main points that were evidenced in this work regarding the differences found in the comparison of Farms A and B.

What is observed in the cases studied was that Farm A faces accommodation in meeting the criteria, while Farm B has presented a continuous improvement in its evaluations. The case study allows us to hypothesize that these differences are directly associated with the degree of owner's commitment to certification.

In Farm A, the focus is on the immediate results that certification can bring, notably on the impact on the final price of the product. The expectation is that certification could result in an increase in the price of coffee marketed. On the other hand, the owner of Farm B understands certification as a process, whose objectives are achieved in the long run. Thus, it does not expect an immediate economic result that can positively impact the price of its product, but understands certification as an ally in obtaining better quality coffees, which is responsible for the increase in market prices (MAEKAWA, CARVALHO; OLIVEIRA, 2013; PEREIRA et al., 2013; MARTINS; SILVA, 2014). Thus, in the face of the finding that occurs after some time of permanence in the program that these results may not happen with this logic, there is a process of accommodation and sometimes resistance in realizing the investments that certification may require, which in extreme cases leads to even the abandonment of the program. This work also contributes to the debate on the continuity of certifications, allowing management agencies to plan the activities of publicity and awareness of the target audience.

In addition to the limitations of the adopted methodology, it should be noted that, although the farms studied are in the same geographic region and have similarities in size and productivity, there is a difference of time in which they are certified. Farm A has 4 years of certification, while Farm B is in the seventh year, and it is possible to assume that this factor may in some way interfere in the degree of maturity and consequently in the results of certification. Future work may include the analysis of the time of certification factor in the performance of certified properties.

Criteria for analysis	Farm A	Farm B
Improvement in score in the program	No	Yes
Meeting certification requirements	Accommodation	Improvement
Investments in the program	Restrictive	Substantial
Results expected	Short term	Long term
Expectation of better coffee prices	Yes	No
Understanding of certification	Distortions	Yes

**TABLE 3** - Criteria for analysis and differences in the socio-environmental performance of farms.

### **4 CONCLUSIONS**

The certifications and management systems benefits is closely related to a behavioural change of the organization, involving motivation, improvement of image and production processes, increase of satisfaction, worker involvement, with direct impact on customer service by offering quality products. However, Certifica Minas Café can lead to improved sustainability of coffee farms in the long term, but the maintenance of the requirements established by the certificate is still low, which makes it difficult to observe sustainability improvements in the future. In this way, the constant technical monitoring for the farmers is fundamental for the effect of the certifications improvements.

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### ATR-FTIR FOR DISCRIMINATION OF ESPRESSO AND AMERICANO COFFEE PODS

Andrés Felipe Bahamon Monje<sup>1</sup>, Lina Ximena Parrado<sup>2</sup>, Nelson Gutiérrez-Guzmán<sup>3</sup>

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ABSTRACT: Roasted and ground coffee for encapsulation in single-serve capsules compatible with keurig® and coffee powder obtained from Nespresso® commercial capsules were analyzed for pH value, titratable acidity, moisture content, water activity and color (lightness); a data matrix that contains the physicochemical properties and the absorbance measurements using a baseline of 1600 to 1800 cm–1 by FTIR-ATR technique, was evaluated through the combined methods of principal component analysis (PCA) and cluster analysis in order to discriminate between the types of capsules. In the PCA biplot two distinct groups can be identified and in the cluster analysis two groups are that correspond to the two types of capsules. The results reveal that FTIR-ATR based methods seem to be a promising alternative for the discrimination of coffee samples for the pods industry or for the type of consumption.

Index terms: Coffee brewing Single-serve brewer, Coffee pods, Extraction brew physiochemical properties, FTIR spectra.

### ATR-FTIR PARA DISCRIMINAÇÃO DE CÁPSULAS DE CAFÉ ESPRESSO E AMERICANO

RESUMO: Café torrado e moído encapsulado para consumo individual compatível com keurig® e pó de café obtido cápsulas comerciais Nespresso®, foram analisadas para valores de pH, acidez titulável, teor de água, atividade de água e cor (luminosidade); uma matriz de dados com as propriedades físico-químicas e medições de absorbância usando uma linha de base de 1600 a 1800 cm–1 pela técnica ATR-FTIR, foi avaliada por meio de métodos combinados de análise de componentes principais (PCA) e análise de clusters a fim de discriminar entre os tipos de cápsulas. No gráfico do PCA podem ser identificados dois grupos distintos, e na análise de clusters, dois grupos que correspondem a dois tipos de cápsulas. Os resultados mostraram que métodos baseados em ATR-FTIR podem ser uma alternativa promissora para a discriminação de amostras de café na indústria de cápsulas ou para o tipo de consumo.

**Termos para indexação:** Máquina de café em dose única, Cápsulas de café, Propriedades físico-químicas no extrato de café, Espectro de FTIR.

#### 1 INTRODUCTION

Espresso and *americano* coffee are a widely consumed beverages worldwide; while the espresso coffee has been defined as a beverage prepared by submitting ground roasted coffee beans to hot water at a certain pressure for a short time, the *americano* coffee is defined how a diluted espresso, extracted with higher water volume; after the extraction, an *americano* coffee has a similar volume to a filtered coffee.

Traditionally, the espresso and *americano* coffees were extracted in a same big and expensive coffee machines, recently new devices was development to make this beverages based on pods systems, they have gained market share because they are user-friendly and they also make it easy to prepare good-quality coffees through the reduction of uncontrolled preparation variables (PARENTI et al., 2013); although encapsulated coffee is increasingly popular, little research information exists about this product. WANG et al. (2016) evaluated the effects of the capsule parameters on extraction yield and physiochemical properties of

coffee brews prepared using a commercial single serve brewer Keurig®; DESBROW et al. (2017) quantified the caffeine content of the Nespresso® coffee pod and BELCHIOR et al. (2017) evaluated the potential of ATR-FTIR and chemometrics to discriminate espresso coffees with different sensory characteristics described by a panel of coffee tasters.

Some food analysis techniques are complicated instruments and require time and more costs; FTIR- ATR spectroscopy is a simple technique that requires no sample pretreatment, is fast and provides an overall infrared fingerprint of the specimen; FTIR- ATR have been recently employed for coffee analysis (REIS et al., 2013a); FTIR analysis was used to detect multiple adulterants in roasted and ground coffee CRAIG et al. (2014); to discriminate between defective and non-defective coffee beans prior to roasting (CRAIG et al., 2012) and to evaluate the chlorogenic acid isomer profile and antioxidant capacity of coffee beans (LIANG et al. 2016b).

Since Chlorogenic acids (CGAs) are a major acid group present in Coffee (FARAH &

<sup>1.2.3</sup> Centro Surcolombiano de Investigación en Café CESURCAFE/Universidad Surcolombiana - Departamento de Ingeniería Agrícola Cx.P. 410001 - Neiva - Colombia - u2010297360@usco.edu.co, u20142131808@usco.edu.co, ngutierrezg@usco.edu.co

DONANGELO, 2006), and caffeine content is relevant in coffee analysis, authors have proposed for the identification and quantification of caffeine and CGAs using infrared spectra finding that peaks are usually located within the wavelength range from 1600 cm-1 to 1800 cm-1 (Table 1).

The objective in this work was to investigate the feasibility of using some physicochemical properties and the bands of the FTIR-ATR spectrum where the CGA's and Caffeine peaks are highlighted (1600-1800 cm-1), in conjunction with PCA and HCA to discriminate between *americano* and espresso coffee pods.

### 2 MATERIALS AND METHODS

### 2.1 Samples

Six commercial Nespresso® coffee pods (7g coffee net weight) (EC) and twelve *americano* coffee pods (12g coffee net weight) (AC) of which three commercial Keurig® k-cup coffee pods and nine roasted and grinding coffee samples packed

in the single-serve coffee capsule EZ-CUP<sup>2.0</sup> compatible with keurig K-cup brewers were used in the analysis; the single origin coffee and encapsulated process is presented in the table 2. The manually encapsulated and sealed process occurred in a laboratory prototype of coffee pod packaging with a single chamber (Figure 1).

### 2.2 Physicochemical analysis in coffee powder Moisture content

Before being packed, the roasted and ground coffee for the Keurig® pods and the coffee powder inside of the Nespresso® pods (Factory packed) were evaluated for moisture content with MB45 OHAUS infrared moisture analyzer.

### Water activity measurement

3±1 g of coffee powder were placed in the sample chamber of a vapor sorption analyzer, at 25oC. (VSA Aqualab Decagon Device, Inc. Pullman, WA).

**TABLE 1** - Reported bibliography of infrared spectral analysis in coffee.

Molecules	Wavelength (W) - peaks (P)	Author(s)/year
	W(1660-1600) cm <sup>-1</sup>	(KEMSLEY et al, 1995)
	P(1700, 1658, 1600) cm <sup>-1</sup>	(DAGHBOUCHE et al, 1997)
	P(1659, 1554, 1710) cm <sup>-1</sup>	(BOUHSAIN et al, 1999)
	W(4000-800) cm <sup>-1</sup> , P(1700,1659) cm <sup>-1</sup>	(GARRIGUES et al, 2000)
	W(1800-1300) cm <sup>-1</sup>	(NAJAFI et al, 2003)
	P(2924, 1591, 1659, 1585) cm <sup>-1</sup>	(ESTEBAN-DÍEZ et al, 2004)
Caffeine	P(1659, 1705) cm <sup>-1</sup>	(GALLIGNANI et al, 2008)
Carrente	P(1650, 1600) cm <sup>-1</sup>	(WANG & LIM, 2012)
	P(1650, 1745) cm <sup>-1</sup>	(RIVERA et al, 2013)
	P(2923, 2854, 2925, 2848, 1659, 1655) cm <sup>-1</sup>	(REIS et al, 2013b)
	P(1659) cm <sup>-1</sup>	(BARBIN et al, 2014)
	W(1700-1600) cm <sup>-1</sup> , P(1642) cm <sup>-1</sup>	(ABDALLA et al, 2015)
	P(1625) cm <sup>-1</sup>	(CRAIG et al, 2018)
	P(1655, 1591, 1072) cm <sup>-1</sup>	(BELCHIOR et al, 2017)
	W(1450-1000) cm <sup>-1</sup> , P(1300, 1150) cm <sup>-1</sup>	(KEMSLEY et al, 1995), (BRIANDET et al, 1996), (REIS et al, 2013b)
	P(1375, 1074)	(BUFFO & CARDELLI-FREIRE, 2004)
GG 4	P(1160, 1250, 1380) cm <sup>-1</sup>	(RIVERA et al, 2013)
CGAs	P(1868, 1741) cm <sup>-1</sup>	(BARBIN et al, 2014)
	P(1605, 1627, 1680, 1742) cm <sup>-1</sup>	(LIANG et al, 2016a)
	P(809, 1032, 1120, 1165) cm <sup>-1</sup>	(LIANG et al, 2016b)
	W(1375-1074) cm <sup>-1</sup> , P(1072) cm <sup>-1</sup>	(BELCHIOR et al, 2017)

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**TABLE 2** - Samples, origin and packed process.

Type of capsule	Origin	Encapsulated
AC	Colombia	Manually packed
AC	Colombia	Manually packed
AC	Bolivia	Manually packed-
AC	Guatemala	Manually packed-
AC	Ethiopia	Manually packed-
AC	Colombia	Factory packed
AC	Colombia	Factory packed
AC	Colombia	Factory packed
AC	Colombia (Huila)	Manually packed-
AC	Colombia (Huila)	Manually packed-
AC	Colombia (Huila)	Manually packed-
AC	Colombia (Huila)	Factory packed
EC	Colombia	Factory packed
EC	Colombia	Factory packed
EC	Colombia	Factory packed
EC	Colombia (Medellin)	Factory packed
EC	Colombia	Factory packed
EC	Colombia	Factory packed



FIGURE 1- Laboratory prototype of coffee pod packaging with a single chamber.

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**Color.** CIE L\*a\*b\* coordinates were obtained in the coffee powder directly in the 50 mm (diameter) cell, in triplicate with Minolta CR-410 Colorimeter (Konica Minolta Sensing Inc., Osaka, Japan).

**Titratable acidity**. Determined by titration, with the Ph meter (BP-3001 manufactured by Trans Instruments, Singapore), 50 ml of distilled water at  $90 \pm 2$  ° C is filtered with 5 grams of roasted ground coffee. After that, 50 mL of the filtered extract, filtered by waterman qualitative filter paper, was titrated against 0.1 N NaOH solution to pH 6.5 Measurements were taken in duplicate.

### 2.3 FTIR analysis

A FTIR Spectrophotometer (Cary 630 manufactured by Agilent, EEUU) was used in the ATR-FTIR measurements that were performed in a dry atmosphere and at room temperature (20 ± 0.5 °C). A horizontal ATR sampling accessory (Diamond ATR) equipped with ZnSe cell was employed. Approximately (1 g) of the roasted and ground coffee was placed in the sampling accessory and pressed; the background material was obtained from readings of the accessory without any sample. All spectra were recorded within a range of 1800-1600 cm-1 where the CGA's and Caffeine peaks are variable importance projection scores (VIP)(BELCHIOR et al., 2017), the resolution used was 4 cm-1 with 20 scans and submitted to background subtraction; all samples were analyzed in triplicate. The chlorogenic acid standard SIGMA ALDRICH CAS 327-97-9 purity ≥99.0% and caffeine standard CAS 58-08-2 purity >99.0% was used for determination of spectrum patterns, in the same range defined with VIP scores.

### 2.4 Coffee brewing

The packed coffee (AC) was brewed using a commercial single serve brewer (K50 Classic Series manufactured by Keurig®, USA), using Lungo method with 177 ml (6 oz) hot water; the Nespresso® pods (EC) were brewed with a commercial single serve (C50-US-CW-NE manufactured by Nespresso®, Switzerland) with pressure of 19 bar and 40 ml (1,35 oz) hot water.

### 2.5 Coffee beverage characterization

The pH value of coffee AC and EC beverages was measured with pH-meter BP-3001 (Trans Instruments, Singapore); the Refractive index with digital refractometer PR-201a (Atago, United States) and the beverage lightness (L\*),

was obtained in the coffee beverages, using a CR-A33e Light Protection Tube with Glass of the digital CR410 (Konica Minolta, japan).

### 2.5 Statistical analysis

All data obtained in this study were analyzed statistically, the results are expressed as the mean  $\pm$  standard deviation; differences among average values were estimated. Comparisons between the two groups were made by Student's t test using the statistical package Statgraphics Centurion XVI. Average values were considered significantly different when P $\leq$  0.05. Cluster and PCA analysis was applied to the matrix with the absorbance values of the peaks related to representative functional groups in 1600 to 1800 cm<sup>-1</sup> region and physicochemical parameters in coffee powder and beverages.

### **3 RESULTS AND DISCUSSION**

### 3.1 Coffee powder and beverages characterization

Table 3 presents the results of Student's t test for the comparison of the coffee powder contained in EC and AC, with the addition of the comparison of the parameters obtained in the EC and AC beverages. The moisture content, a<sub>w</sub> and the degree of roasting in coffee powder from EC presented statistically significant differences (P<0.05) compared to AC; likewise, in the analysis of the beverages, pH, °Brix and beverage lightness (L\*) presented statistically significant differences (P<0.05) in the two types of drinks.

The moisture content and a in AC is significantly higher than in EC, this result may be influenced by the storage conditions or because the AC package material allowed vapor exchange between the inside of the pods and the surrounding environment; the color of roasting in the coffee powder of EC and AC showed significant differences (p< 0.05), the magnitudes of the coordinate L obtained (23.20±0.53 and 24.79±1.5 respectively) correspond to the degree of dark roasting according to the classification proposed by FRANCA et al. (2009), although the color in samples are statistically different this result is appropriate the preparation of espressos and americano coffee.

The refractive index in the beverages had higher result in espresso than in *americano* coffee as shown in Table 3, similar results were obtained by GLOESS et al. (2013) where he determined that espresso coffee has a refractive index closer to 4.0%.

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**TABLE 3** - Physico-chemical parameters in EC and AC coffee powder and beverages

T	Dl	Type of capsule	
Type	Physico-chemical parameters	EC	AC
	Moisture content (%)	$2.71 \pm 0.62^a$	$4.51 \pm 1.52^{b}$
	Water activity (Aw)	$0.25\pm0.11^{\text{a}}$	$0.38\pm0.11^b$
Roasted and ground coffee	Color (°L)	$23.20 \pm 0.53^a$	$24.79 \pm 1.50^{b}$
	Titratable acidity (0.1 M NaOH / ml)	$1.68\pm0.40^{a}$	$1.85\pm0.36^{\text{a}}$
	(Ph)	$5.21\pm0.13^{a}$	$4.92 \pm 0.11^{b}$
Beverage coffee	Refractive index (°Brix)	$3.92\pm0.53^a$	$1.66\pm0.45^{b}$
	Color (°L)	$32.04 \pm 2.89^a$	$29.80 \pm 1.15^{b}$

n=3, mean ± SD. Different letters, in the same row indicate significant difference (P<0.05).

The author evaluated these parameters in different machines: a semi-automatic espresso machine, an automatic machine and a Nespresso® brand single-use machine. In the americano coffee, the same author determined that the refractive index was slightly higher than 1.0%; finding for the filtered coffee extract a refractive index of  $1.03\pm0.01\%$  and for the French press  $1.43\pm0.01\%$ , these results are similar to those found for the AC. The pH in beverages was similar as those obtained by FUJIOKA & SHIBAMOTO (2008) in seven types of commercial coffees varying between  $4.95\pm0.01$  and  $5.99\pm0.01$ , this author comments that the pH in extracted coffee is related to the presence of chlorogenic acids, although WANG & LIM (2012) comments that high values in the pH of coffee beverages is presents in dark roasted coffee beans and it is associated with the presence of organic acids (citric and malic acids).

### 3.2 FTIR spectral analysis

Figure 2A shows the CGAs (blue) and Caffeine (red) spectrum patterns; the figure 2B shows the approach of the region between 1600 to 1800 cm<sup>-1</sup> in the CGAs standard, three peaks are clearly defined, the carbonyl (C=O) group at 1685 cm<sup>-1</sup>, the ethylene (C=C) group at 1636 cm<sup>-1</sup> and the phenyl ring stretch at 1599 cm<sup>-1</sup> according to LIANG et al, (2016a); likewise, the figure 1C presents the Caffeine standard enlargement, two peaks are clearly defined, the carbonyl (C=O) group at 1642 cm<sup>-1</sup> and the amines (C=N) group at 1692 cm<sup>-1</sup>.

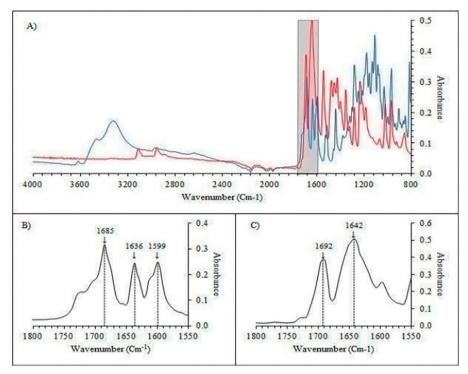
Figure 3 presents the *americano* coffee pods AC (red) and espresso coffee pods EC (blue) spectrums in the region 1600 to 1800 cm<sup>-1</sup>. The spectrums obtained in this research are similar with those reported by LIANG, et al. (2016a) in commercial coffee samples; the figure 3 shows the CGAs and caffeine molecules superimposed with the functional groups located in the corresponding Wavenumber, which were taken to obtain the respective absorbance values. These absorbance values were included in the multivariate matrix for PCA and HCA analysis.

### 3.3 Cluster Analysis

Figure 4 shows the dendrogram based on similarity of physical-chemical parameters and absorbance values of the functional groups peaks in 1600–1800 cm-1 region. Two clusters are defined for the AC and EC coffee pods samples. All of the americano coffee pods samples AC are included in the Cluster 1, though it is not possible to find a separation pattern for different origins or between commercial or manual encapsulated type. The cluster 2 includes the six espresso coffee pods samples EC.

### 3.4 Exploratory analysis

The biplot of the PCA on the matrix composed with the absorbance values of the peaks related to representative functional groups in 1600 to 1800 cm-1 region and physicochemical parameters in coffee powder and beverages is shown in figure 5.



**FIGURE 2 - A)** FTIR spectral of pure chlorogenic acid and caffeine standards. **B)** and **C)** enlargement of the 1600 to 1800 cm<sup>-1</sup> region in CGAs and Caffeine standards

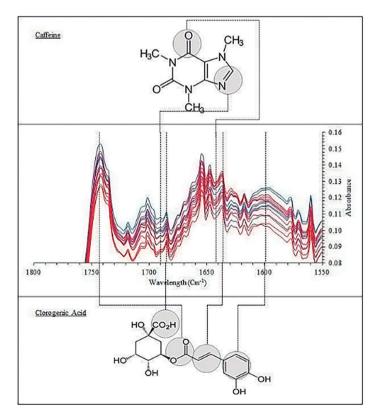


FIGURE 3 - FTIR spectrums for coffee pods AC (red) and EC (blue) samples (CGAs and caffeine molecules superimposed).

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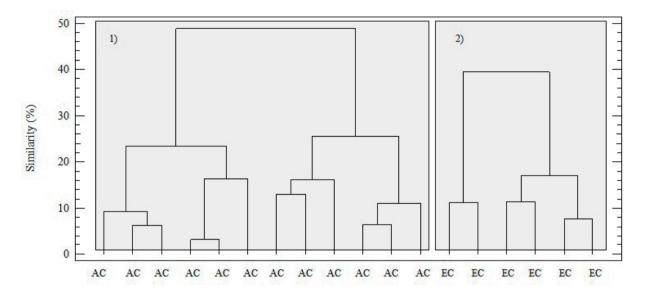


FIGURE 4 - Dendrogram for cluster analysis in americano (AC) and espresso (EC) coffee pods.

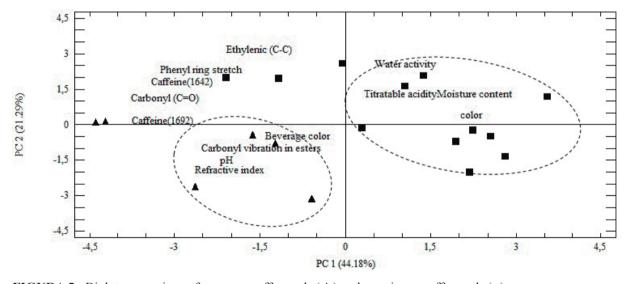


FIGURA 5 - Biplot comparison of espresso coffee pods (▲) and americano coffee pods (■).

The first two principal components, PC1 and PC2, accounted for approximately 65% of the variability. Two distinct groups can be perceived: AC coffee pod samples are located in positive PC1, while EC coffee pod samples are located in negative PC2 quadrant.

These results reveal that FTIR-based methods seem to be a promising alternative for the discrimination of coffee samples for the pods industry or for the type of consumption; in this study, the FTIR technique allow a discrimination between type of capsules for different extraction methods; so, it could be defined the type of capsules (espresso or *americano*) can be elaborated according to the type of coffee available.

### **4 CONCLUSION**

A high moisture content and a was found in coffee powder AC. This can significantly affect sensory acceptance since high moisture contents can be indicators of product interaction with the environment given the high degree of hygroscopicity of the coffee.

A differentiation was identified between EC and AC coffee pods, PCA and cluster analysis results indicated that coffee samples could be separated into distinct groups, based on both. The absorbance values of the peaks related to representative functional groups in 1600 to 1800 cm<sup>-1</sup> region and physicochemical parameters in coffee powder and beverages.

### **5 ACKNOWLEDGMENT**

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### EFFECT OF COVER CROPS AND BIOACTIVATORS IN COFFEE PRODUCTION AND CHEMICAL PROPERTIES OF SOIL

Kleso Silva Franco Junior<sup>1</sup>, Ana Beatriz Carvalho Terra<sup>2</sup>, Tiago Rezende Teruel<sup>3</sup>, José Ricardo Mantovani<sup>4</sup>, Ligiane Aparecida Florentino<sup>5</sup>

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ABSTRACT: Coffee cultivation has great socio-economic relevance in Brazil for the employment and income generation and there is currently a constant search for sustainable management techniques. Among them, we can mention the use of cover crops and soil bioactivators. However, studies relating the use of these two techniques are still incipient. Based on this, the objective of this research was to evaluate the effect of the Penergetic® bioactivator associated with different cover crops on chemical properties of soil and coffee productivity. The experiment was carried out in a coffee field with Catuaí Vermelho cultivar IAC 144, in a randomized block design in a factorial scheme 4 (soil cover) x 2 (use or not of the Penergetic® bioactivator), consisting of control (without plant cover); oats (*Avena strigosa*) + forage turnip (*Raphanus sativus*); oats (*Avena strigosa*) + forage turnip (*Raphanus sativus*); *Brachiaria brizantha* (*Urochloa brizantha*), associated or not with the use of the Penergetic® bioactivator. The experiment was conducted for 6 months and after that period, the chemical properties of soil, the nutrient contents of the coffee plants, the development of the branches and the coffee productivity were analyzed. Data were analyzed by the Scott Knott test at 5% probability. It was verified the interaction between cover crops and the use of the Penergetic® bioactivator, positively influencing soil chemical characteristics, coffee nutrition and productivity.

**Index terms**: Green fertilization, coffee nutrition, fertility.

# EFEITOS DAS PLANTAS DE COBERTURA E BIOATIVADOR NA PRODUÇÃO DE CAFÉ E AS PROPRIEDADES QUÍMICAS DO SOLO

RESUMO: A cafeicultura apresenta grande relevância socioeconômica no Brasil pela geração de renda e empregos e atualmente verifica-se uma busca constante por técnicas de manejo sustentáveis, dentre as quais, podemos citar o uso de plantas de cobertura e de bioativadores de solo. No entanto, estudos relacionando o uso dessas duas técnicas ainda são incipientes. Baseado nisso, o objetivo desta pesquisa foi avaliar o efeito do bioativador Penergetic® associado a diferentes plantas de cobertura nas características químicas do solo e na produtividade do cafeeiro. O experimento foi instalado numa gleba de café com a cultivar Catuaí Vermelho IAC 144, em delineamento de blocos casualizados em esquema fatorial 4 (cobertura do solo) x 2 (uso ou não do bioativador Penergetic®), sendo constituído por: Controle (sem planta de cobertura); aveia (*Avena strigosa*) + nabo forrageiro (*Raphanus sativus*); aveia (*Avena strigosa*) + nabo forrageiro (*Raphanus sativus*) + tremoço (*Lupinus albus*) + centeio (*Secale cereale*) + ervilhaca (*Vicia sativa*); *Brachiaria brizantha* (*Urochloa brizantha*), associados, ou não ao uso do bioativador Penergetic®. O experimento foi conduzido por 6 meses e após esse período foram analisadas as características químicas do solo, teores de nutrientes foliar das plantas do cafeeiro, desenvolvimento dos ramos e a produtividade do cafeeiro. Os dados foram analisados pelo teste de Scott Knott a 5% de probabilidade. Foi verificada a interação entre plantas de cobertura e o uso do bioativador Penergetic® influenciando positivamente as características químicas do solo, na nutrição e produtividade do cafeeiro.

Termos para indexação: Adubação verde, nutrição do cafeeiro, fertilidade.

### 1 INTRODUCTION

Most Brazilian soils present low natural fertility (SOUSA and LOBATO, 2004), associated with high acidity and aluminum concentration, therefore requiring high doses of correctives and fertilizers to guarantee satisfactory production (CARVALHO et al., 2017), however, there is an increase in costs once that fertilizers are mostly imported (RODRIGUES et al., 2015).

Coffee stands out among the crops of great economic importance, and Brazil is the world's largest producer and exporter (SUPLICY, 2013), with an estimated harvest of approximately 60 million bags in 2018 (CONAB, 2018). Coffee is considered a highly nutrient-demanding crop, requiring adequate soil fertility management in order to achieve nutrient balance and high productivity. This culture is the one with the lowest nutrient utilization among large crops according to Stipp and Casarin (2012).

¹Centro Superior de Ensino e Pesquisa de Machado/CESEP - Av. Dr. Athaide Pereira de Souza s/n - 37.750 -000 -Machado - MG kleso.junior@yahoo.com.br

<sup>&</sup>lt;sup>2,3,4,5</sup>Universidade José do Rosário Vellano /UNIFENAS - Rod 179 Km 0 - 37.132-440 - Alfenas -MG - anabeatriz.terra@hotmail.com, tiago.teruel@unifenas.br, jose.mantovani@unifenas.br, ligiane.florentino@unifenas.br

Cover crops are presented as an ecological practice and with lower costs for agricultural management, promoting balance in the physical, chemical and biological characteristics that compound the soil-plant system. Besides being an organic fertilizer alternative without great resources, they can be grown in the same environment where they will be produced for this purpose (SOUZA et al, 2008). Several species are recommended for cultivation between the lines of coffee with this objective, as reported by Matos et al. (2008).

In relation to soil fertility, the use of green fertilization contributes to the increase of organic matter and the effective (cation exchange capacity) CEC of the soil due to the availability of negative loads. Most of the green fertilizers are legumes, which establish symbiosis with rhizobia, reducing the cost with nitrogen fertilizers (SILVA, 2015).

Another strategy that has been used with the proposal to reduce the need for chemical fertilizers are the soil bioactivators, which are constituted by organic substances, humic and fulvic acids, amino acids, algae extract and vitamins, which may or may not, being associated with micronutrients (CASTRO et al., 2007). These products act in plant development by various mechanisms, such as ion transport and enzymes responsible for metabolism, which may influence the secondary metabolism, promoting the synthesis of precursors of important plant hormones and, consequently, contributing to plant nutrition (CASTRO et al. al., 2007). Külen et al. (2011) state that they act in all stages of the development of cultivated plants, and can benefit different crops, such as soybeans, corn, wheat, and vegetables. These substances may also contribute to seed germination (O'BRIEN, FOWKES e BASSOM., 2010).

The Penergetic® bioactivator has been used in Brazil and, among its mechanisms of action, it is possible to highlight the greater soil microbiota balance, increasing the organic matter decomposition and the nutrient cycling, reducing the use of inputs, thus contributing for the sustainability of the agroecosystem (CALEGARI, 2013). However, despite all the benefits of green manuring and bioactivators, studies involving the use of these two techniques are still incipient. Based on this, this research aims to evaluate the effect of the Penergetic® bioactivator associated with different cover crops on soil chemical characteristics and coffee productivity.

### 2 MATERIAL AND METHODS

The experiment was carried out at Farm Boa Esperança, located in Serrania, South of Minas Gerais, in 2016, from May to December. The experimental area has the following geographical coordinates: Latitude: 21 ° 36'18.29 "S, Longitude: 46 ° 07'46.29 ° W and 982 m Altitude. The selected coffee farm, cultivated with Red Catuaí IAC 144, was planted in 2011, spacing 3.5 m between rows and 0.7 m between plants.

The experimental design was a randomized complete block design in a 4x2 factorial scheme, with 4 treatments containing or not cover plants. distributed as follows: control treatment, without cover plant, with traditional farm management (weed control by chemical and mechanical); treatment containing oats (Avena strigosa) + forage radish (Raphanus sativus L); (Lupinus albus) + rve (Secale cereale) + vetch (Vicia sativa) (ANTCE) and the treatment containing only brachiaria ( *Brachiaria brizantha*) as cover plant. All of these treatments were associated or not with Penergetic® soil bioactivator product. Four replicates per treatment were used, with a total of 32 experimental plots. Each plot of the experiment was constituted by 10 plants, being considered as a useful plot the six central plants for evaluation.

The planting densities of the cover plants were: brachiaria and forage turnip, 10 kg ha<sup>-1</sup>, oats 40 kg ha<sup>-1</sup>, lupine and rye 20 kg ha<sup>-1</sup> and vetch 15 kg ha<sup>-1</sup>. The bioactivator was used at a dose of 0.6 kg ha<sup>-1</sup>, as recommended by the manufacturer.

Before the installation of the experiment, samples of soils in the 0 - 10 cm and 10 - 20 cm layers were carried out to carry out the chemical analysis, according to table 1.

The cover plants were kept in the area from May to October, a time that was mowed, and kept on the coffee tree line. Two months later, soil samples were removed in the 0-10 cm and 10-20 cm layers to perform the fertility analysis.

In the same period, leaves were collected in the middle third of the plants, where the fourth pair of leaves were removed from both sides of the plant and, subsequently, the plants were sent to the laboratory for analysis of nutrient contents, according to Silva (2015). The number of internodes of the plagiotropic branches was also evaluated. Six lateral branches were randomly selected at the height of the middle third in six plants of each plot. It was also evaluated the number of internodes developed in the period (ALFONSI, 2008).

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**TABLE 1** - Result of soil chemical analysis 0 - 10 cm and 10 - 20 cm depth in the installation in the experiment.

Chemical Parameters	0 - 10 cm	10 - 20 cm
pH (CaCl <sub>2</sub> )	5,8	5,6
MO (g/kg)	30	27
P (mmol <sub>c</sub> /dm <sup>3</sup> )	77	22
K <sup>+</sup> (mmol <sub>c</sub> /dm <sup>3</sup> )	5,7	4,4
$Ca^{2+}$ (mmol <sub>c</sub> /dm <sup>3</sup> )	62	45
$Mg^{2+}$ (mmol <sub>c</sub> /dm <sup>3</sup> )	27	18
H+Al (mmol <sub>c</sub> /dm <sup>3</sup> )	29	34
S (mmol <sub>c</sub> /dm <sup>3</sup> )	14	15
SB (mg/dm <sup>3</sup> )	95	68
$T (mg/dm^3)$	123	102
V (%)	76,9	66,6
$Cu^{2+}$ (mg/dm <sup>3</sup> )	4,4	2,6
$Fe^{2+}$ (mg/dm <sup>3</sup> )	72	81
$Mn^{2+}$ (mg/dm <sup>3</sup> )	5,8	4,2
$Zn^{2+}$ (mg/dm <sup>3</sup> )	4,5	2,5
$B (mg/dm^3)$	0,98	0,64

In order to evaluate the productivity, six plants of each plot were collected, measuring the drying and processing of the coffee. and later the classification and the granulometry to determine the percentages of sieve 16 coffee were performed according to Brazil (2003).

The data were submitted to the Scott Knott test at 5% of probability, using the Sisvar computer program (FERREIRA, 2011).

### **3 RESULTS AND DISCUSSION**

For the results presented in the chemical analysis of the soil, performed in the 0-10 cm depth layer, all verified parameters, except for sulfur, presented significant interaction between soil cover and use of the Penergetic® bioactivator (Table 2).

Regarding the pH values, it was verified that, with the use of the bioactivator, no alteration of soil pH occurred. However, without the use of bioactivator, the pH was modified, and the highest value was observed in the control treatment, and the lowest in the soil cultivated with oats + turnip + lupine + rye + vetch (ANTCE).

These results can be justified by the cultivation of green manure plants, which, although favoring the cycling of nutrients and

the decomposition of the organic material by soil microorganisms, promote a reduction of pH at the beginning of the process (OLIVEIRA, 2005).

When the action of the bioactivator is observed within the treatments, it is noticed that in the control treatment and with brachiaria there were significant differences in some of the evaluated parameters. In the control treatment, in the absence of the bioactivator, the pH was higher and the CTC lower when compared to its presence. In the treatment with brachiaria, it was possible to observe that in the presence of the bioactivator, the pH was lower, but the contents of CTC and Organic Matter were higher when compared to the treatment where there was no use of the bioactivator. For the coverage plants ANTCE the bioactivator had a superior effect compared to the same plant without its use.

Regarding the values of organic matter, no statistical difference was observed in the different soil cover treatments in the presence of the bioactivator. In the absence of the bioactivator, the soil cultivated under brachiaria presented lower value. However, according to Bressan et al. (2013), nutrient content and organic matter levels were higher in the areas covered by millet and brachiaria, and these plants were similar in relation to changes in soil chemical attributes.

**TABLE 2** - Chemical parameters of soil samples collected in the 0-10 cm layer, in the interline of coffee plants grown under different cover crops, associated or not with the use of the Penergetic® bioactivator.

Chemical parameters	Bioactivator	Control	Oat and turnip	ANTCE*	Brachiaria
nII (CoCl.)	With	4,48 A b	4,33 A a	4,73 A a	4,53 A a
pH (CaCl <sub>2</sub> )	Without	5,50 A a	4,35 B a	3,95 C b	4,63 B a
MO (g dm <sup>-3</sup> )	With	30,25 A a	28,00 A a	27,25 A b	29,50 A a
WO (g dili*)	Without	30,25 A a	29,25 A a	30,25 A a	25,25 B b
V (0/)	With	58,50 A a	33,50 Ba	68,50 A a	51,50 A a
V (%)	Without	70,50 A a	33,25 Ba	30,50 B b	45,25 B a
Potential Acidity	With	44,00 A a	46,25 A b	33,00 A b	54,25 A a
mmolc/dm <sup>3</sup>	Without	31,00 C a	70,50 A a	82,00 A a	54,25 A a
Sum of bases	With	64,33 A a	63,15 A a	71,55 A a	56,92 A a
mmolc/dm <sup>3</sup>	Without	74,60 A a	42,03 B b	35,75 B b	43,38 B a
CTC potential	With	116,25 Aa	120,00 A a	104,50 B b	120,25 A a
mmolc/dm <sup>3</sup>	Without	106,00 Bb	115,75 A a	118,00 A a	101,50 B b

Average followed by distinct letters, uppercase in the row and lowercase in the column, differ by Scott Knott's test at 5% probability. \* Abbreviation of treatment: oats + turnip + lupine + rye + vetch.

Analyzing the interaction between soil cover *versus* bioactivation, it was observed that the treatment of oat + turnip + lupine + rye + vetch (ANTCE) showed a lower value of organic matter in the presence of the bioactivator. According to Calegari (2013), these results occur because of this product favoring the soil microbiota and, consequently, the decomposition of organic matter.

In addition, in this treatment, lupine and vetch may have contributed to reducing the C/N ratio, favoring the decomposition of organic matter justifying the lower values of this parameter. Cunha et al. (2011), researching cover plants as: crotalaria (Crotalaria juncea), pigeonpea (Cajanus cajan (L.), (Mucuna aterrima), sorghum broom (Sorgum technicum) in fallow on the physical attributes of soil cultivated with organic beans and com under direct seedding and conventional preparation verified that although providing a good amount of biomass did not reflect the increase in the increase of the organic matter since this also depends on the quality of the residues, the C/N ratio, recalcitrant among other factors for its increase.

When observing the different types of soil cover without the bioactivator, the treatments oats + turnip and oats + turnip + lupine + rye + vetch (ANTCE) presented higher values of potential acidity. Tomé Júnior (1997 apud Paulett, 2012) confirms the trend of higher values of potential acidity (H + Al<sup>+ 3</sup>) in soils with higher organic matter content, especially in more acidic soils.

Regarding the saturation of bases, when the bioactivator was used, only the oat + turnip treatment presented a lower result than the others. However, without the use of the bioactivator, the soils cultivated with the different cover plants were statistically inferior to the control treatment.

As for the effect of the bioactivator on the different cover plants, it was observed that the only treatment in which interaction occurred was oat + turnip + lupine + rye + vetch (ANTCE), and the bioactivator promoted a higher base saturation value. The same was observed by Callegari et al. (1993), which states that cover plants provide increased base saturation promoting soil fertility improvement by reducing the oxidation rate of organic matter in the soil.

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For the soil analyzes collected in the 10-20 cm layer, the values of pH, organic matter, phosphorus, potassium, magnesium, and sulfur showed the interaction between soil cover versus the use of the bioactivator, as can be observed in table 3.

Regarding pH, in the presence of the bioactivator, the control treatment was the one with the lowest value. In the absence of the bioactivator, the soil cover with *Brachiaria* spp. promoted a lower pH value, a similar result for the 0-10 cm layer (Table 2). Analyzing the interaction between the soil cover *versus* the bioactivator, it was verified that the only treatment that showed influence was the control treatment, in which the use of the bioactivator reduced the pH of the soil, as observed for the 0-10 cm layer.

For the organic matter contents, the use of the bioactivator provided higher levels in the control and ANTCE treatments. In the absence of the bioactivator, the highest values were observed in the oat and turnip and ANTCE treatments.

In relation to phosphorus, with the use of the bioactivator, the highest value was for the soil covered with oats, turnip, lupine, rye and vetch (ANTCE). When the bioactivator was not used, the highest values in the phosphorus contents were for the control and oat and turnip treatments. Analyzing the interaction between soil cover *versus* bioactivator, it was observed that the bioactivator® worked positively, increasing phosphorus levels, in the treatments containing ANTCE and Brachiaria. According to Tirloni et al. (2009), bioactivators reduce phosphorus adsorption sites, increasing their availability in the soil for plants.

**TABLE 3** - pH value and organic matter content of soil analyzes collected in the layer 10-20 cm, in the interline of coffee plants grown under different cover crops, associated or not with the use of the Penergetic® bioactivator

Chemical Parameters	Bioactivator	Control	Oatmeal + turnip	ANTCE*	Brachiaria
nU (CoCl )	With	4,30 Bb	4,65 Aa	4,85 Aa	4,90 Aa
pH (CaCl <sub>2</sub> )	Without	4,98 Aa	5,00 Aa	4,85 Aa	4,50 Ba
Organic matter	With	27,50 Aa	24,00 Bb	28,25 Ab	25,75 Ba
$(g/dm^3)$	Without	27,00 Ba	30,00 Aa	31,50 Aa	24,25 Ca
Phosphor	With	15,25 Ca	17,00 Cb	137,50 Aa	81,50 Aa
(mmolc/dm³)	Without	33,50 Aa	47,50 Aa	24,00 Bb	13,75 Cb
7 (1.1)	With	4,40 Aa	2,95 Bb	3,30 Ba	3,03 Ba
Potassium (mmolc/dm³)	Without	3,01 Bb	4,90 Aa	3,50 Ba	2,09 Ba
	With	7,00 Bb	6,50 Ba	15,25 Aa	7,50 Ba
Magnesium (mmolc/dm³)					
	Without	11,75 Aa	7,00 Ba	5,75 Bb	5,75 Ba
Sulfur (mmolc/dm³)	With	6,00 Aa	5,50 Aa	7,00 Aa	8,75 Ab
	Without	4,50 Ba	6,75 Aa	3,75 Ba	20,25 Aa

Means followed by distinct letters, uppercase in the row and lowercase in the column, differ by Scott Knott's test at 5% probability. \* Abbreviation of treatment: oats + turnip + lupine + rye + vetch.

Potassium showed a statistically significant difference between the treatments, if it was superior to the coverings oats + turnip and the control treatment, however, in relation to the bioactivator, the control treatment was statistically superior when compared to the use of the bioactivator.

Magnesium in the ANTCE treatment with the use of the bioactivator was shown to be statistically superior, showing effect both in the coverage plant and in the use associated to the bioactivator.

For the phosphorus, the bioativador the highest levels were found in the treatments with Oat + turnip and in the consortium Oat + turnip + lupine + rye + vetch. When comparing the results of the treatments with and without bioactivator, it was observed that the product provided higher phosphorus contents in the *Brachiaria* and Oat + turnip + + rye + vetch plots, corroborating with Foloni et al. (2008) that Brachiaria presents high capacity in the recycling of P. Pavinato and Rosolem (2008), on the other hand, they showed the possibility of solubilization of phosphorus of the soil, in less labile forms, in the presence of plant residues, which may have occurred in the plots of the consortium between Oat + turnip + lupine + rye + vetch and in the treatment with Brachiaria.

The values of calcium, base sum, CTC and base saturation of the soil analysis results collected in the 10 to 20 cm depth profile did not show interaction with or without the use of the bioactivator (Table 4), having only soil cover influence.

Observing the values shown in Table 4, of the calcium contents, the treatments Control and ANTCE were the ones that presented better results, being statistically equal among themselves. The same can be observed for the other parameters (base sum, CTC and base saturation). For the results of leaf analysis, there was a significant interaction for the nitrogen, potassium, calcium, magnesium, sulfur, boron, iron and manganese contents. The values for zinc were significant only for cover plants, while the phosphorus and copper contents were not significant, as shown in Tables 5 and 6.

The results of the plant tissue analysis showed that, in relation to the Nitrogen element, without bioactivator the treatments did not differ statistically. On the other hand, with the use of the bioactivator it was verified that the N was lower in the treatments with Oat + turnip, demonstrating that no interaction occurred.

For the potassium nutrient, the interaction of the bioactivator, with a statistical difference, occurred only for the treatment with Brachiaria, and among the cover plants, the lowest statistically found result was with *Brachiaria*.

In relation to Calcium, the treatments with bioactivator use were higher than Brachiaria without this association.

When considering Magnesium, the best treatments were oats + turnip and oats + turnip + lupine + rye + vetch without bioactivation and oats + turnip using bioactivator. It is also evidenced that the ANTCE was higher without the bioactivation when compared to the same treatment with the use of the bioactivator.

For the data of the Sulfur, the Control and Oat + turnip and oat + turnip + lupine + rye + vetch with bioactivators showed statistical differences. The treatments Oats + turnip and oats + turnip + lupine + rye + vetch and Brachiaria without the use of the bioactivator did not differ among themselves. However, in the control treatment the presence of the bioactivator showed positive response, with higher results than the treatment without bioactivator, however, *Brachiaria*, the bioactivator had no positive interaction.

**TABLE 4** - Values of calcium, base sum, CTC and base saturation of soil samples collected in the layer 10-20 cm, in the interline of coffee plants grown under different cover plants.

Soil cover	Calcium mmolc/dm <sup>3</sup>	Base sum %	CTC mmole/dm³	Base saturation %
Control	41,38 A	57,46 A	104,75 A	58,13 A
Oat and turnip	34,00 B	47,34 B	96,88 B	48,63 B
ANTCE*	49,00 A	65,15 A	107,50 A	61,25 A
Brachiaria	32,38 B	41,99 B	93,88 B	44,38 B

Averages followed by letters differ from one another by the Scott Knott test at 5% probability. \* Abbreviation of treatment: Oats, Turnip, Lupine, Rye and Vetch.

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**TABLE 5** - Foliar contents of the macronutrients nitrogen, potassium, calcium, magnesium and sulfur of coffee plants grown under different cover crops, associated or not with the use of the Penergetic® bioactivator.

Chemical Parameters	Bioactivator	Control	Oatmeal + turnip	ANTCE*	Brachiaria
Nitrogen	With	27,79 Aa	22,64 Bb	28,34 Aa	30,75 Aa
(g/Kg)	Without	27,41 Aa	26,88 Aa	29,86 Aa	28,38 Aa
Potassium	With	23,31 Ab	22,94 Ab	23,44 Aa	25,00 Aa
(g/Kg)	Without	26,56 Aa	25,50 Aa	25,00 Aa	22,50 Bb
Calcuim	With	9,06 Aa	9,13 Aa	8,69 Aa	8,81 Ab
(g/Kg)	Without	8,60 Ba	8,69 Ba	9,13 Aa	10,00 Aa
Magnesium	With	2,92 Ba	3,38 Aa	3,02 Bb	3,013 Ba
(g/Kg)	Without	3,05 Ba	3,022 Ba	3,52 Aa	3,024 Ba
Sulfur	With	2,39 Aa	1,87 Ba	2,44 Aa	1,78 Ba
(g/Kg)	Without	1,59 Bb	1,56 Ba	2,46 Aa	2,07 Aa

Average followed by distinct letters, uppercase in the column and lowercase in the row, differ by Scott Knott's test at 5% probability. \* Abbreviation of treatment: Oats, Turnip, Lupine, Rye and Vetch.

**TABLE 6** - Foliar contents of boron, iron and manganese micronutrients of coffee plants grown under different cover crops, associated or not with the use of the Penergetic® bioactivator.

Soil cover	Boron (	mg/Kg)	Iron (r	ng/Kg)	Manganes	se (mg/Kg)
Son cover	With Bio	Without Bio	With Bio	Without Bio	With Bio	Without Bio
Control	79,21 A a	63,46 A b	138,25 A a	132,50 A a	140,75 A a	111,38 A b
Oat and turnip	63,92 B a	67,10 A a	138,25 A a	118,25 A b	119,25 B a	110,25 A a
ANTCE*	61,92 B b	73,64 A a	131,50 A a	100,00 B b	132,50 A a	116,25 B a
Brachiaria	69,67 A a	69,03 A a	120,00 A a	117,50 A a	112,50 B a	124,00 A a

Averages followed by distinct letters, uppercase in the column and lowercase in the row, differ by Scott Knott's test at 5% probability. \* Abbreviation of treatment: Oats, Turnip, Lupine, Rye and Vetch

For the Boron element, in the presence of the bioactivator the control treatment presented higher results than the others, being also superior to the same treatment in the absence of the bioactivator. Regarding iron, in the treatment with bioactivator there was no statistical difference between the different sources of cover in the soil, however, in the absence of the product, the ANTCE treatment was the one that presented the lowest result. Analyzing the interaction, it was verified that the bioactivator had a positive effect when associated with oat and turnip treatments and ANTCE.

In relation to the Manganese, it was observed that the control treatment with bioactivator use was superior to the others, whereas in the absence of the product there was no significant difference between the treatments. Analyzing the effect of the bioactivator, there is a positive effect on the control treatment.

Table 7 shows the results obtained with respect to the zinc micronutrient.

Regarding Zinc, only the soil cover was significant, and the treatments oats and turnip and ANTCE cultivated in the coffee interlayer provided higher levels of this micronutrient in the coffee leaves.

Table 8 shows the number of internodes developed in the period, coffee production per plant and productivity.

Regarding the number of internodes, in the presence of the bioactivator, all the treatments cultivated with cover plants were superior to the control treatment. Analyzing the interaction, it was verified that the bioactivator had a positive effect when associated with Brachiaria.

TABLE 7 - Zinc leaf contents of coffee plants grown under different cover crops

Soil cover	Zinc(mg/Kg)
Control	10,83 B
Oat and turnip	14,19 A
ANTCE*	15,75 A
Brachiaria	11,63 B

Averages followed by distinct letters differ from one another by the Scott Knott test at 5% probability. \*Abbreviation of treatment: Oats, Turnip, Lupine, Rye and Vetch.

**TABLE 8** - Number of internodes and productivity coffee (bags ha-1) yield under different cover crops, associated or not with the use of the Penergetic bioactivator

Soil cover	Number of i	Number of internodes		gs ha <sup>-1</sup> )
	With Bio	Without Bio	With Bio	Without Bio
Control	5,91 B a	5,99 A a	16,74 C a	14,91 C a
Oat and turnip	5,99 A a	5,87 B a	25,10 B a	19,21 B b
ANTCE*	6,37 A a	5,91 B a	26,54 B a	21,08 B b
Brachiaria	6,91 A a	5,80 B b	30,19 A a	30,20 A a

Averages followed by distinct letters, uppercase in the column and lowercase in the row, differ by Scott Knott's test at 5% probability. \* Abbreviation of treatment: Oats, Turnip, Lupine, Rye and Vetch.

This data confirms Partelli's (2013) apud Dubberstein et al., (2017) demonstrating that a greater number of internodes comes from the longer productive branches that promote the production of more rosettes, allowing a greater emission of flowering buds leading to a greater quantity of fruits in the plant.

Analyzing coffee productivity, it was verified that the cover treatment with Brachiaria was superior to the others, both in the presence and absence of the bioactivator. Analyzing the interaction, the bioactivator had a positive effect on ANTCE and oat and turnip treatments.

### **4 CONCLUSIONS**

It was observed that the bioativador promotes effect on soil chemical characteristics, nutrition and productivity of coffee, however, these results were variable according to the cover plant cultivated in the coffee plant.

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