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Cupuassu from bean to bar: Sensory and hedonic characterization of a chocolate-like product

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ABSTRACT

The processing of cupuassu (*Theobroma grandiflorum* Schum) beans after fermentation gives a chocolate-like product, the cupulate. The high amount of pulp adhered to the seeds hinders the fermentation. Consequently, it is necessary to depulp the seeds to perform the process, even though the pulp contains important substrates for the formation of flavor precursors. To verify whether the complete or partial removal of the pulp influences the sensory characteristics of the product, fermentation was performed with three pulp concentrations (0, 7.5, and 15%) and two schemes of turning for aeration of the mass: fixed (R1) and according to the temperature (R2), in a total of six experiments (0R1, 0R2, 7.5R1, 7.5R2, 15R1 and 15R2). The beans were processed to obtain cupulates, which were submitted to tests performed with consumers, to express their preference and attributes (acceptance, purchase intent, Check All That Apply – CATA), and then to tests with a trained panel, the Quantitative Descriptive Profile (QDP) to characterize the samples. Both tests showed the consumers' perceptions that the cupulates have peculiar sensory characteristics. In the Consumer Test, through the Preference Mapping, all the samples of cupulates obtained from R1 conditions were preferred. In the penalty analysis, these same samples showed positive attributes that mask the negative attributes. Both CATA and QDP results showed that cupulate samples produced from seeds with a higher amount of pulp (15R1 and 15R2) had a higher number of positive mentions, for their fruity and floral flavors. The research also demonstrated that all samples gave the perception of an earthy taste, an important reason for consumer rejection, as well as a bad residual flavor. Thus, the results showed that the presence of the pulp in the fermentation environmental is important to the formation of flavor compounds and improving the sensory acceptance of the products.

1. Introduction

The fermentation of cupuassu (*Theobroma grandiflorum* Schum) seeds allows the obtention of a chocolate-like product with similar sensory characteristics such as brown color, flavor, and texture (Ramos et al., 2016). As with cocoa (*Theobroma cacao* L.), fermentation is an essential step to the sensory characteristics of the product, since important flavor compounds are formed during this process (Munoz et al., 2020). Through the fermentation of cupuassu seeds, some important volatile compounds are formed, including flavor precursors (Ramos et al., 2016). Some compounds are desirable and important for the sensory characteristics of the product (Rodriguez-Campos et al., 2011). The cupuassu fruit has compounds with functional properties such as antioxidant, probiotic, and reduction of hypertriglyceridemia (Avila-Sosa et al., 2019). However, long days (>six) of fermentation of cupuassu seeds can reduce the level of some important bioactive compounds (catechins) and the antioxidant activity (Álvarez et al., 2017).

Cupuassu seeds are still considered by-products, which results in waste of the material. Both seeds and pulp present high nutritional value (Campos-Rodriguez et al., 2021). The seeds also have a considerable amount of carbohydrates, fiber, lipids, and phenolic compounds (Costa, Santos, Lannes, et al., 2020). The presence of essential amino acids and proteins, with high biological value, is considered superior to cocoa beans (Carvalho et al., 2005; Lopes et al., 2008). The fermentation time

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of cupuassu seeds can last for up to 7 days, and the degree of fermentation is evaluated by the cut test, in which cupuassu differs from cocoa by the color of the beans. While poorly fermented cocoa beans are purple, cupuassu are beige (Ramos et al., 2020). In well fermented beans, the brown color exhibited is due to the oxidation of phenolic compounds (Cohen & Jackix, 2005; Lopes et al., 2003). To carry out the fermentation, the cupuassu seeds are usually completely depulped, since abundance and composition of pulp seems to hinder the fermentation, prejudicing the action of yeasts (Ramos et al., 2016, 2020). Moreover, the pulp is the most used part for commercial purposes (Cohen & Jackix, 2005; Matos et al., 2008).

Application of turning during fermentation is essential to make the temperature uniform throughout the mass, increasing aeration and favoring aerobic microorganisms, such as acetic acid bacteria (AAB) (Schwan & Wheals, 2004). AAB play an important role in the production of flavor precursor compounds, such as acetic acid, that leads to the increase of the mass temperature, reaching up to 50 °C (Nielsen et al., 2013; Schwan & Wheals, 2004). After fermentation, the beans must be subjected to the drying process, until they reach 6–8% moisture (Carvalho et al., 2005; Cohen & Jackix, 2005; Vilalba et al., 2004). Processing of the beans to obtain the cupulate is performed through the same steps applied in the production of chocolate (Cohen & Jackix, 2005). Roasting is an important step, in which occurs the main formation of flavor compounds and the destruction of undesirable flavor compounds in the final product (Afoakwa et al., 2008; Serra Bonvehí & Ventura Coll, 2002). Amino acids released by the action of proteolytic enzymes during fermentation react with reducing sugars, especially in the steps of roasting, conching, and tempering, producing important flavor compounds (aldehydes, ketones, and pyrazines) through the Maillard Reaction (Afoakwa et al., 2008; Schwan & Wheals, 2004).

After processing, the products can be subjected to sensory analysis to estimate the degree of acceptance by the consumers and, depending on the test, to obtain sensory attributes according to the characteristics perceived by the tasters. That is important to qualify the product, to accept it or to reject it. In this way, sensory analysis can be used as a tool with trained or amateur assessors, with reference materials associated to the characteristics of the product under study, in an environment with controlled conditions (Jaeger et al., 2015; Minim et al., 2010; Teixeira, 2009). Chocolate-like products elaborated from cupuassu beans have been evaluated by means of acceptance testing with satisfactory results (Cohen et al., 2009; Lopes et al., 2003).

In the present study, cupuassu beans from six fermentation conditions (depulped seeds, seeds with 7.5% and 15% of pulp, and two schemes of turning for each) were processed to obtain 50% cupuassu bars. After that, the products were submitted to the Quantitative Descriptive Profile (QDP) and Consumer Test by Check All That Apply (CATA) and Purchase intention. In this way, this study aimed to evaluate the influence of pulp content and turning schemes applied to cupuassu seeds during fermentation on the sensory perception with trained and untrained consumers to improve technologies applied post-harvest and to add value to an important native product of the Amazon rainforest.

2. Material and methods

2.1. Sample preparation

2.1.1. Fermentation

About 1 t of ripe and healthy cupuassu fruits were collected after their fall, in Presidente Figueiredo-Amazonas, Brazil, in January 2013. The fruits were broken and cupuassu seeds were depulped (0%) and partially depulped (7.5 and 15%). The fermentation process was performed and two schemes of turning were applied to each condition: R1 (fixed) made 48 h after the beginning of the fermentation and then each 24 h; and R2, where the first turning occurred once the temperature had doubled inside the fermenting mass and the following turns occurred when the average fermentation temperature dropped. The experiments were identified as 0R1 and 0R2; 7.5R1 and 7.5R2; 15R1 and 15R2. The total fermentation time for the experiment 0R1 was of 60 h; to 0R2, 7.5R1 and 7.5R2 was 84 h; and 15R1, 15R2 was 108 h.

2.1.2. Drying

After fermentation, the beans were subjected to natural drying (solar) in the first 24 h, and then to artificial drying in an oven with air circulation (MARCONI MA 035/5/10P São Paulo, BRAZIL), set at 40 °C for 5–7 days until they reached 6–8% moisture. The combination of natural (solar) and artificial (mechanical dryers) for the drying process was performed due to the rainy weather in the period with temperature range between 23.4 and 34.7 °C (INMET, 2013) in Manaus, Amazonas State, Brazil.

2.1.3. Processing

The cupulate processing was held in the pilot plant of the Fruit, Vegetable, Beverages and Confectionery Products of the School of Food Engineering of UNICAMP (São Paulo, Brazil), following the same steps used for chocolate production. The beans from each condition (0R1, 0R2, 7.5R1, 7.5R2, 15R1, 15R2) were roasted at 120 °C / 120 min in batches of 4.0 kg in a rotary electric oven (JAF INOX São Roque, Brazil). The beans acquired a characteristic browning color inside. This condition used as a standard was defined based on previous studies (Cohen et al., 2003; Carvalho et al., 2005). After roasting, the beans were broken in ICMA knife mill (ICMA, Campinas, Brazil) to obtain the nibs. Then the nibs were separated from the shell according to their granulometry (separation made in a vibrating device with sieves - PRODUTEST - Model T, series 3244, Brazil) and using a cocoa winnower equipment (Capco, Ipswich, UK). The nibs were ground in a knife mill to obtain the cupuassu mass. To prepare the cupulate, the following ingredients, cupuassu mass (48.9%), refined, and icing sugar ("Glaçucar" União) (48.9%) were mixed in a planetary mixer (KitchenAid® Artisan Model 5KSM150, USA). The mixture was refined in a three-cylinder mill (PILLON, Brazil), internally cooled with water at 15 °C to obtain a powder with particle size smaller than 25 μm, so that the granules were imperceptible during tasting. The refined ingredients were placed in a homogenizer, a device equipped with a steam jacket connected to other equipment with a pump to circulate water at 60 °C. Then, cupuassu butter extracted from cupuassu mass using a hydraulic press (1.4%), soybean lecithin (CH Solec - the Solae Company) (0.4%), and polyglycerol polyricinoleate-PGPR (Grindsted Super - Danisco) (0.4%) were added. The mixture was kept in this condition for up to 4 h, until complete melting. After that, conching was performed in the conching equipment (INCO, Brazil) under stirring at a temperature of 70 °C for 12 h to reduce the acidity and humidity, to improve the flavor and to promote pasteurization at the same time. After the conching time, the cupulate was subjected to manual tempering in a controlled temperature room (20.0 ± 1.0 °C). Tempering was performed on a preheated marble surface, with continuous movements being applied to the melted cupulate, with time and temperature control until the formation of stable crystals to guarantee a homogeneous and shiny mass, in addition to facilitate the demolding of the product. The initial mass temperature (50 °C) was reduced to 30–32 °C at a rate of 2 °C/min. After the tempering step, the cupulate was molded into polycarbonate molds. After filling the molds, manual vibration was performed to remove air bubbles. The molds with cupulates were cooled at 4–8 °C, for approximately 30 min. The cupulate bars were then demolded and packed in aluminum foil, identified, and stored at ± 20 °C until the sensory analysis and consumer tests.

2.2. Consumer and sensory tests

The project was submitted and approved by the Research Ethics Committee of the University of Campinas (Unicamp), in Brazil under N° CAAE 07336612.0.0000.5404. All subjects signed a consent form to participate in the sensory and consumer tests.

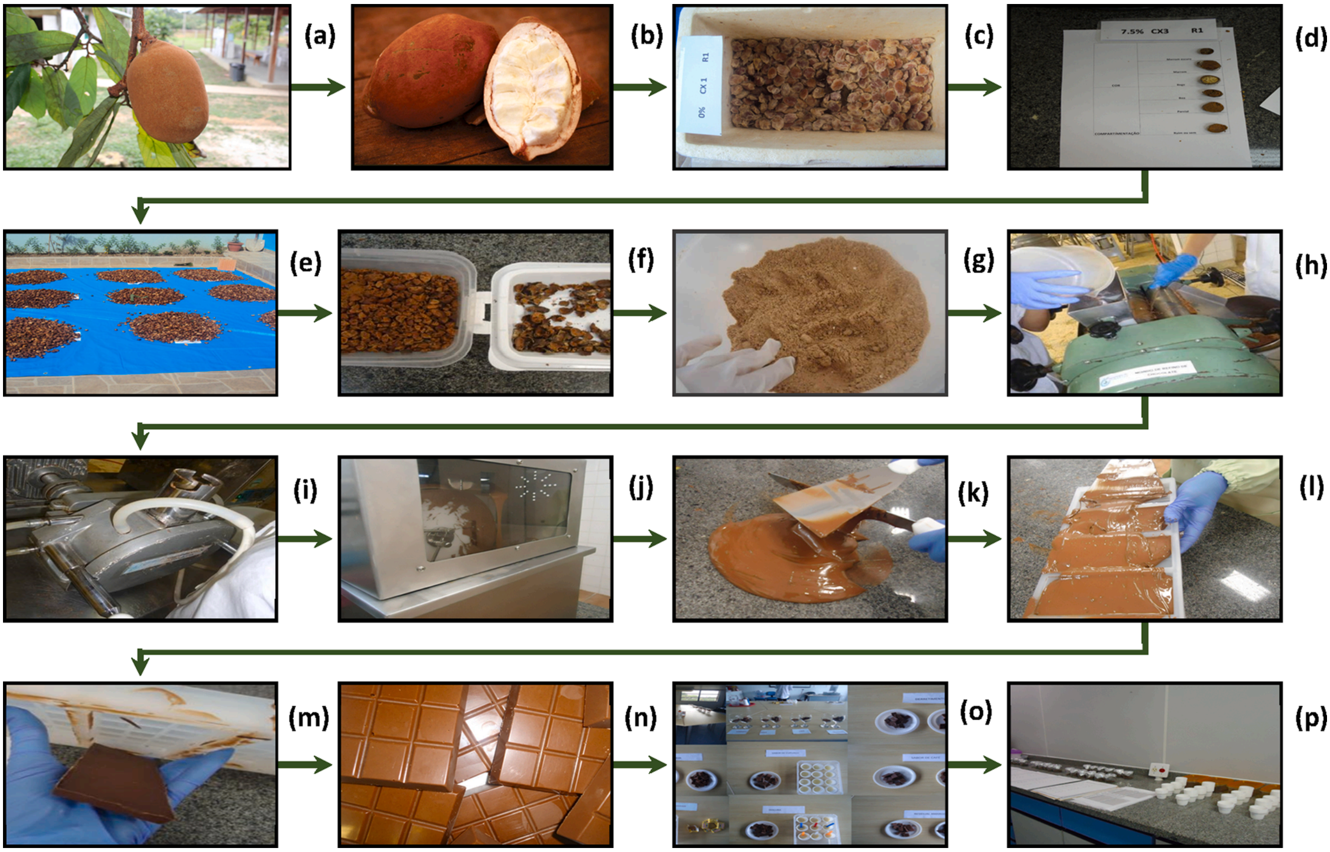

Fig. 1. (a) cupuassu fruit; (b) cupuassu seeds with pulp; (c) fermenting box with cupuassu seeds; (d) cutting test; (e) cupuassu beans drying; (f) nibs of cupuassu and shells after roasting; (g) cupuassu powder and addition of sugar; (h) refining of the mix (cupuassu and sugar); (i) addition of cupuassu butter, soy lectin, PGPR, and homogenization at 60 °C / 4 h; (j) conching; (k) tempering; (l) molding; (m) demolding; (n) cupulates bars; (o) Quantitative Descriptive Profile (QDP); (p) Sensory analysis.

2.2.1. *Consumer test*

The consumer test was performed in the sensory evaluation laboratory of the Food Technology Institute (ITAL) Campinas, São Paulo in Brazil with 60 regular consumers of dark chocolate (14 men and 46 women), most belonging to the B2 class (middle class). Definition of class is made according to the Brazilian Criterion of Economic Classification (ABEP, 2013). The definition of the number of consumers was made according to technical criteria that establish a minimum number of 30 people at random, for regular consumers of the product (Teixeira, 2009). Most recruited consumers (>50%) reported to prefer milk chocolate, and flavor was the most cited attribute.

The test was conducted in individual booths with fluorescent lighting equipped with the computational system *Compusense Five* version *5.4* for collecting and analyzing data. The samples were monadically evaluated in a single session according to a complete block design and coded with three-digit random numbers, served at room temperature in napkins. Participants were asked to sip water between samples to clean the palate.

The samples were evaluated for overall liking and especially for aroma, flavor, and texture using a nine points hedonic scale: 9 = I liked it extremely, 5 = I did not like nor disliked and 1 = I disliked extremely. Bitterness, acidity, and sweetness intensity were evaluated using a five-point just about right scale: 5 = much bitter/acid/sweet than what I like, 3 = just about right and 1 = much less bitter/acid/sweet than I like. Finally, the intent to purchase was evaluated: 5 = I would certainly buy it, 3 = maybe/maybe not and 1 = I would certainly not buy it (Meilgaard et al., 2007; Dutcosky, 2013).

Complementing the acceptance test, the perception of cupulates was assessed by the Check-All-That-Apply (CATA) (Ares et al., 2010). CATA is a method by which consumers evaluate a product and qualify it with the aid of a list of terms (descriptive, aesthetic, hedonic, emotional etc.). The choice is dependent on the individual's perception, considered appropriate to describe a product in order to find its potential attributes (Dooley et al., 2010; Dutcosky, 2013). For CATA Test, it is usually recommended to recruit consumers who have appreciation for the product to be tested or to a similar one (Meilgaard et al., 2007). According previous methodologies of analysis with chocolate using CATA test (Ares et al., 2010), and with some modifications, a list with 27 terms related to flavor was presented to the 60 consumers for free choice: sour/aggressive acidity, fruity, floral, cheese, bitterness, herb/chlorophyll, tobacco, earthy, fermented, it takes time to melt, pepper/cinnamon, vanilla, coffee, roasted nuts, caramel/molasses, sweet, citrus, it is too hard to bite, soft to the bite, firm to the bite (but not hard), granular/sandy, pleasant residual, bad residual, it melts well, it melts quickly and it melts in the mouth, smooth/creamy, pleasant acidity.

2.2.2. *Quantitative Descriptive Profile (QDP)*

In order to sensorially characterize the cupulate samples, the Quantitative Descriptive Profile (QDP) was carried out using a trained panel. For QDP, the assessors were not the same recruited for Consumer Test. The QDP was held in the sensory analysis laboratory of the Department of Food Technology, School of Food Engineering of UNICAMP - São Paulo, Brazil in the same batch of samples used to the Consumer Test.

Before QPD analysis the candidates for assessors, consumers of chocolate (42 young people with an average age of 25 years old, 60% female and 40% male), participated in a basic taste test. Twenty-one assessors with > 80% correct answers were selected to compose the QDP panel to develop the descriptive terminology of the attributes of the cupulate by means of the Kelly's Repertory Grid Method (Moskowitz,

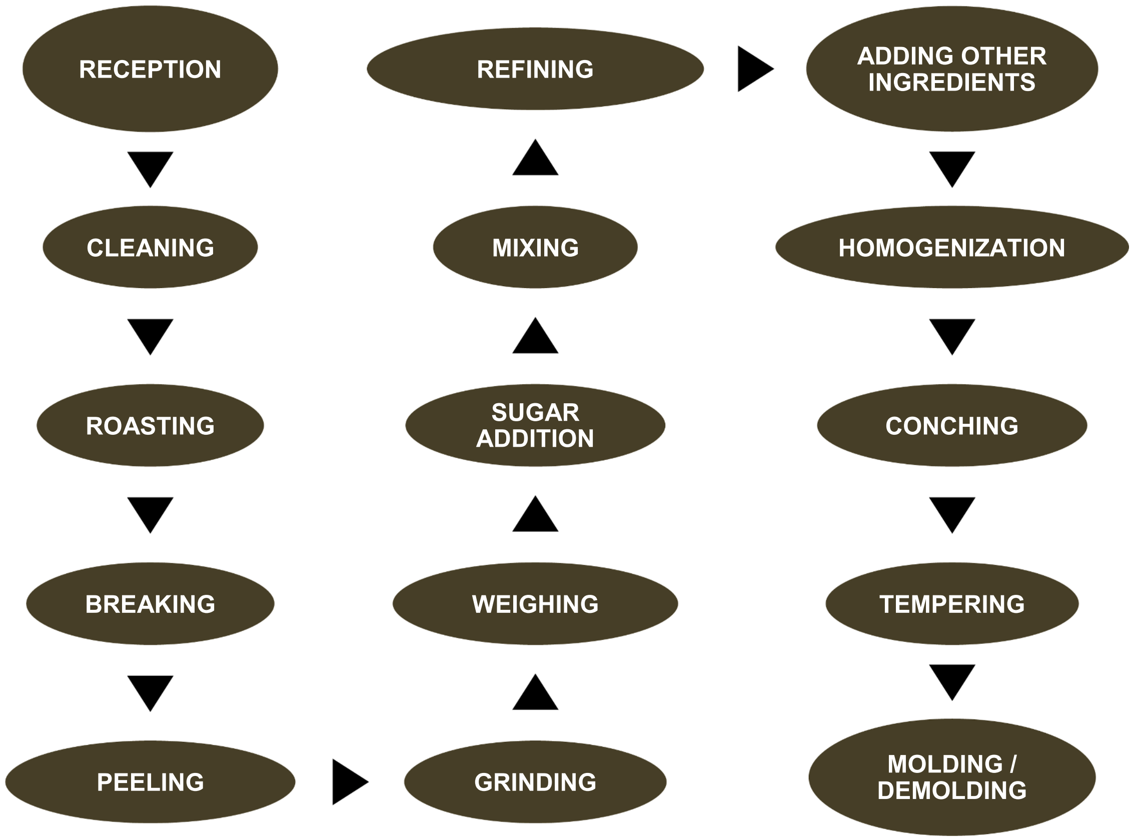

Fig. 2. Flow chart of the processing of cupuassu beans from diferents experiments to obtain cupulates.

1984). From two samples of cupulate with concentrations of 50 and 70% cupuassu, the descriptions of the terms (appearance, aroma, flavor and texture) were collected for QDP test. Discussions were held and the panel decided that attributes of aroma, basic tastes, flavor, and texture should describe the cupulates samples. Each descriptor was measured by means of a 9 cm unstructured scale anchored with intensity terms in the extremes, like weak and strong. For the final production of cupulate samples for sensory analysis, the concentration of 50% was used. The panelists were selected considering consensus judgment of the samples and their discrimination ability (Psample ≤ 0.05), and reproducibility of judgements (Prepetition ≥ 0.05). Fifteen assessors (66.6% female and 33.3% male) with the best results were selected and trained. After 4 months of training and testing, the sensory panel comprised 11 assessor for QDP panel. During the tests and QDP analysis, cupulate samples (~7 g) in small disposable cups were presented to the panelists coded with three-digit numbers. Finally, the same batch of cupulate samples used in the Consumer Test were evaluated in triplicate following a complete block design, in individual booths with fluorescent lighting and forms to fill the answers about sample attributes. The repetitions were performed for each assessor in three separate sessions to avoid sensory fatigue (a break of approximately 1 h between sessions). Mineral water and cream cracker were offered to them for cleansing the palate between tasting samples.

2.3. Statistical analysis

QDP and hedonic data were submitted to Analysis of Variance (ANOVA) followed by Tukey's means comparison test ($p \leq 0.05$) using the SAS (Statistical Analysis System) software, version 9.0. For CATA, the attributes were analyzed using Cochran's Q test, Correspondence Analysis and Penalty Analysis using the Overall Liking score ®XLSTAT Pro, 2015.

3. Results and discussion

3.1. Fermentation, drying and cupulate bars processing

Fig. 1 shows an overview of pre-processing and processing of cupulates.

Pre-processing involved post-harvesting, breaking of fruits, seeds depulping, fermentation in boxes, and drying of beans in a total of 7 days in Manaus, Amazonas State, Brazil (Fig. 1). After fermentation, cupuassu beans were submitted to the cut test. The beans were considered well fermented by exhibiting darker surface and deep partitioning formation (Ramos et al., 2020). Dried beans were delivery to the University of Campinas, São Paulo, Brazil to be processed for obtaining cupulates.

Fig. 2 shows the main steps of cupuassu beans processing for cupulate production.

Upon arrival at the Faculty of Food Technology at the University of Campinas, the beans were immediately subjected to processing. Although the cupulate processing follows the same steps applied to obtain chocolate (Fig. 2), there are some differences between the raw materials used in the composition of the products that influence the sensory attributes, especially the texture. Cupuassu butter is softer, exhibiting a different melting point: cocoa butter melts at about 31 °C, while cupuassu butter melts at 33.9 °C (Cohen et al., 2003). The softness is due to the high content of monounsaturated fatty acids, especially oleic acid. Thus, the final product, in the case of cupulate, presents a smoother texture, without the firmness and characteristic snap (breaking sound) that a well-tempered chocolate has. The crystallization process in the cupulate tempering stage occurred when the mass temperature reached 32 °C. After molding and demolding, the bars presented desired texture and brightness, also indicating that the conching, and especially the tempering, was performed correctly. Cupulate bars were obtained for each experiment and then submitted to sensory analysis.

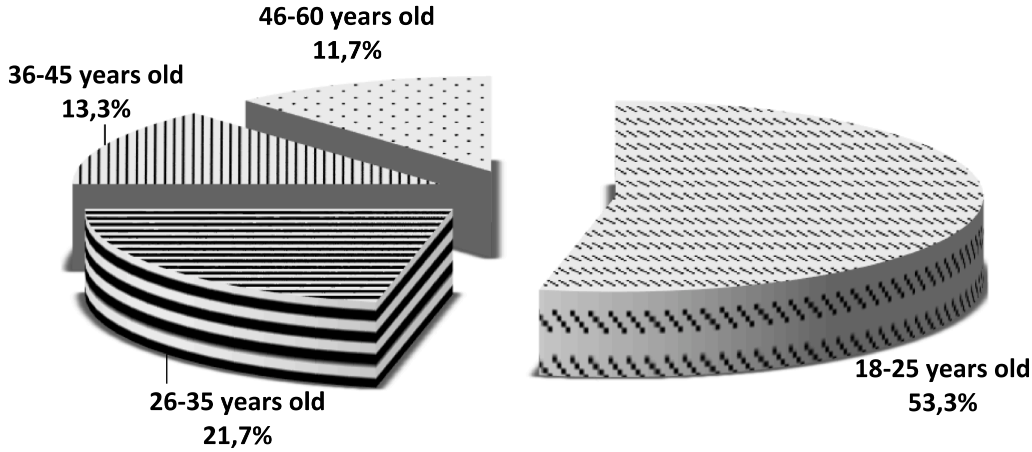

Fig. 3. Age group distribution of consumers of dark chocolate recruited to the Consumer Test.

Table 1
Overall liking scores given to the different cupulates evaluated for acceptance for the six cupulate samples evaluated during Consumer Test.

Attributes	Average scores / Sample						
	0R1	0R2	7.5R1	7.5R2	15R1	15R2	MDS
Hedonic Scale (Scale 0 – 9)							
Overall acceptance	5.8 (1.8) a	5.1 (2.0) ab	5.4 (2.1) a	4.5 (2.2) b	5.2 (2.3) ab	5.8 (2.0) a	0.73
Aroma	5.9 (1.6) ab	5.8 (1.5) ab	5.8 (1.5) ab	5.9 (1.5) ab	5.5 (1.8) b	6.1 (1.6) a	0.57
Flavor	6.0 (1.9) a	5.1 (2.0) bc	5.6 (2.1) ab	4.7 (2.3) c	5.2 (2.3) bc	5.8 (2.0) ab	0.74
Texture	6.6 (1.4) a	6.0 (1.9) bc	6.5 (1.8) ab	5.0 (2.0) d	5.8 (1.9) c	6.6 (1.6) a	0.62
Just About Right Scale (Scale 0 – 5)							
Bitterness	3.5 (0.7) a	3.6 (0.9) a	3.5 (0.9) a	3.6 (0.9) a	3.5 (0.8) a	3.3 (0.9) a	0.35
Acidity	3.1 (0.6) ab	3.4 (0.7) a	3.2 (0.6) ab	3.3 (0.7) ab	3.3 (0.7) ab	3.1 (0.6) b	0.27
Sweetness	2.6 (0.8) ab	2.3 (0.8) b	2.8 (1.0) a	2.5 (0.9) ab	2.6 (0.9) ab	2.6 (0.8) ab	0.30
Purchase intention (Scale 0 – 5)	2.9 (1.3) a	2.5 (1.2) ab	2.6 (1.2) a	2.1 (1.0) b	2.6 (1.3) a	2.9 (1.3) a	0.43

* Results expressed as mean (standard deviation).
MDS: Minimum Significant Difference (Tukey test). For each attribute (line) values followed by equal letters are not statistically different from each other to the error level of 5%.

3.2. Consumer test

Fig. 3 shows the age group distribution of consumers of dark chocolate over 18 years recruited to the Consumer Test.

In the Consumer Test, the consumers recruited evaluated aspects related to the attributes listed in Check All That Apply (CATA Test), Just About Right (JAR) scale, and purchase intent in the six samples of cupulates. Most of the volunteers (>50%) who participated in the survey were young people aged between 18 and 25 years (Fig. 3).

3.2.1. Overall acceptance, just about right (JAR) scale, and purchase Intention.

Table 1 presents the samples' overall acceptance, like aroma, flavor, and texture, as well as the perception of bitter taste, acidity, and sweet taste on the Just About Right (JAR) scale, and purchase intention.

0R1 and 15R2 samples obtained the best acceptance, even for the aroma, flavor, and texture attributes. There was no significant difference between the bitterness intensity of the samples. Regarding the acidity, 0R1 and 15R2 samples obtained an average corresponding to "just about right" considered slightly more acidic ($p \leq 0.05$) than the ideal, while the other samples obtained intermediate averages from fermentation with greater concentration of pulp (7.5R1, 15R1 and 15R2), except 7.5R2. As for sweetness, 7.5R1 was considered just about right (Table 1). The highest score of rejection was to the flavor attribute, indicating a correlation between this attribute and purchase intention. There are few studies available in the scientific literature about sensory analysis of cupuassu and its products. Some studies indicate that this exotic fruit has technological and nutritional potential (Costa, Santos, Rodrigues, et al., 2020), also because of the pleasant and strong flavor that it presents, due to the chemical composition of the pulp, especially rich in esters (Quijano & Pino, 2007). The purchase intention presented low scores. Only samples of experiments 0R1 and 15R2 obtained the highest averages with statistical difference only for 7.5R2, with the lowest score (Table 1).

Fig. 4 shows an external preference mapping constructed to observe the descriptive profile built by the assessors and the consumers according to their preference for cupulate samples.

The preference mapping shows that the samples that underwent fixed turning (0R1, 7.5R1 and 15R1) were preferred by the consumers (>50%). The group of samples with turning R2 (according to the drop in the temperature of the fermentation mass) was preferred by less than 40% of the consumers (Fig. 4). This is an interesting result considering the global acceptance averages. Only the evaluations for the 15R2 sample were superior to the 15R1 (Table 1) while in the preference mapping obtained intermediate preference (between 40 and 50%). Preliminary studies with the seeds, during the fermentation process, indicated that the R1 condition presented a higher concentration of flavor precursor compounds in the middle of the fermentation, maintaining this level until the end (Ramos et al., 2020). This fact perhaps explains the greater preference of consumers for the samples of condition R1.

3.2.2. Check-All-That-Apply (CATA)

Table 2 shows the frequency of mentions raised by the consumers for

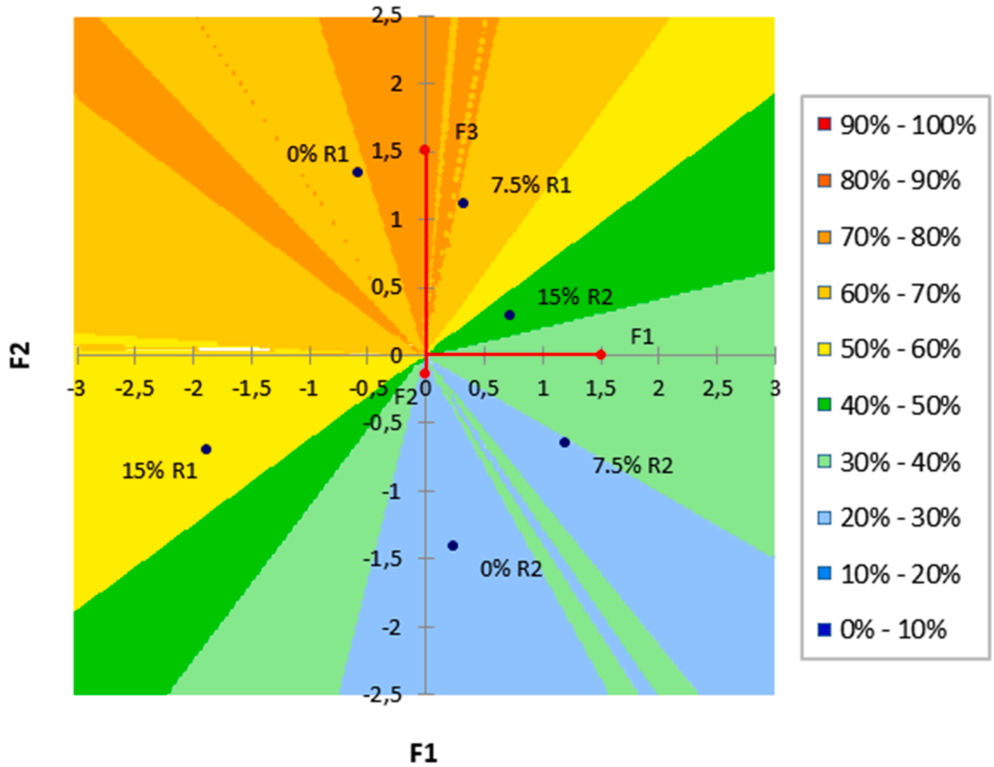

Fig. 4. External preference mapping correlating tasters (%) and their preference about cupulates samples.

Table 2
Frequency of mentions for each CATA attribute for cupulates evaluation.

Attributes	p-values	%					
		0R1	0R2	7.5R1	7.5R2	15R1	15R2
sour	0.531	5.0a	10.0a	10.0a	11.7a	11.7a	5.0a
fruity	0.762	8.3a	6.7a	8.3a	6.7a	11.7a	10.0a
floral	0.594	1.7a	1.7a	0.0a	1.7a	3.3a	3.3a
cheese	0.221	0.0a	0.0a	1.7a	0.0a	3.3a	0.0a
bitterness	0.197	**60.0ab**	**73.3b**	**63.3ab**	**58.3ab**	**63.3ab**	**55.0a**
herby	0.378	0.0a	1.7a	3.3a	5.0a	3.3a	1.7a
tobacco	0.053	13.3ab	21.7b	13.3ab	16.7ab	6.7a	16.7ab
earthy	0.001	**35.0a**	**46.7ab**	**45.0ab**	**53.3b**	**31.7a**	**31.7a**
fermented	0.917	10.0a	6.7a	10.0a	11.7a	10.0a	10.0a
takes time to melt	0.006	8.3a	20.0ab	11.7a	18.3ab	28.3b	10.0a
melt well	0.000	31.7c	23.3abc	26.7bc	8.3a	13.3ab	38.3c
pepper/cinnamon	0.677	3.3a	3.3a	1.7a	3.3a	5.0a	1.7a
vanilla	0.416	1.7a	0.0a	0.0a	0.0a	1.7a	3.3a
coffee	0.382	**33.3a**	**28.3a**	**33.3a**	**25.0a**	**36.7a**	**25.0a**
nuts	0.874	11.7a	15.0a	13.3a	16.7a	11.7a	13.3a
caramel	0.007	8.3a	10.0a	8.3a	8.3a	21.7b	10.0a
sweet	0.003	25.0bc	8.3a	26.7bc	13.3ab	28.3c	21.7abc
citrus	0.609	3.3a	5.0a	1.7a	3.3a	6.7a	1.7a
too hard to bite	0.002	3.3a	3.3a	6.7ab	18.3c	15.0bc	5.0ab
soft to bite	0.300	21.7a	21.7a	18.3a	8.3a	15.0a	20.0a
firm to the bite	0.065	**41.7ab**	**36.7ab**	**41.7ab**	**26.7a**	**40.0ab**	**51.7b**
melt quickly	0.009	18.3b	8.3ab	16.7b	5.0a	8.3ab	5.0a
smooth/creamy	0.000	16.7bc	8.3ab	11.7ab	0.0a	5.0ab	25.0c
pleasant acidity	0.176	20.0a	11.7a	15.0a	6.7a	15.0a	18.3a
sandy	0.000	23.3ab	41.7bc	16.7a	56.7c	30.0ab	15.0a
pleasant residual flavor	0.093	18.3b	5.0a	15.0ab	8.3ab	15.0ab	15.0ab
bad residual flavor	0.000	**33.3ab**	**60.0c**	**43.3bc**	**60.0c**	**35.0ab**	**23.3a**

[1] Frequency expressed on percentage of 60 evaluations.
[2]Samples' results in red indicate higher number of citations concerning to the attributes
p-value: represents the probability of the effect (or difference) observed between the samples be due to chance and not to the factors being studied. Values followed by the same letters are not statistically different from each other to the error level of 5% ($p > 0.05$).

each CATA attribute for the six cupulate samples evaluated.

Among the attributes raised by the consumers, there were fruity, floral, herby, pepper, cinnamon, nuts, caramel, and citrus flavors. All samples were characterized with higher frequency of mentions for bitterness, earthy, coffee, bad residual flavors, and texture firm to the bite. Samples 0R1, 7.5R1 and 15R1 were characterized by coffee flavor

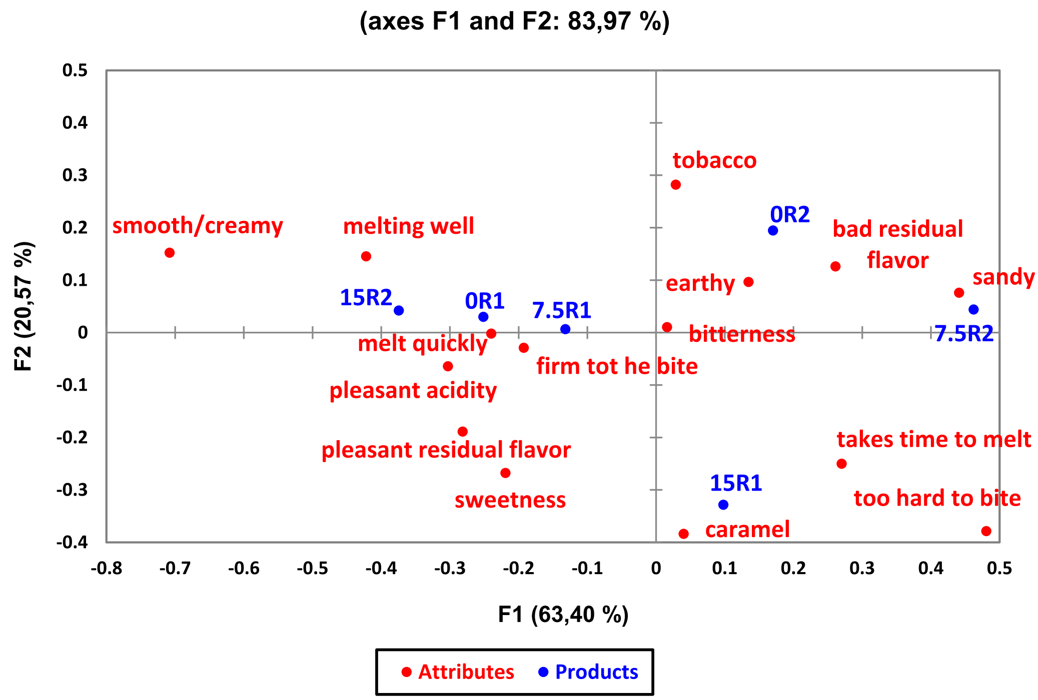

Fig. 5. Correspondence Analysis showing the attributes that consumer chose to characterize the cupulate samples.

and the perception of sweetness, and 15R1 was also characterized by caramel flavor. Samples 0R1, 7.5R1 and 15R2 were characterized by melting well and being firm to the bite, and 0R1 and 15R2 were also considered smooth/creamy with pleasant acidity. Samples 0R2, 7.5R1 and 7.5R2 were characterized by earthy flavor, whereas 0R2 also presented tobacco flavor, resulting that 0R2 and 7.5R2 had bad residual. Samples 15R1 and 15R2 obtained the highest number of mentions for fruity and floral flavors (Table 2). Those attributes are found in fine chocolate due to some compounds present in the pulp (Kadow et al., 2013; Sukha et al., 2008).

In total, samples 0R2 and 7.5R2 received the highest number of negative comments and 0R1 and 15R1 received positive comments. Despite that, bitterness was present in all samples, accounting for a reasonable rejection rate, understandable by the fact that most participating consumers have reported preference for milk chocolate. In general, the R1 scheme (fixed turning) seems to have better influenced the formation and prominence of desirable flavor compounds, as well as some texture attributes. Only sample 15R2 showed higher citations to the texture attribute (melting well, firm to the bite, smooth/creamy). Nevertheless, all samples except 7.5R2 received great mentions regarding their soft texture, an inherent characteristic of cupuassu butter perceived by the consumers (Table 2).

Fig. 5 shows the Correspondence Analysis with attributes that consumers chose to characterize the cupulate samples with the attributes from Check All That Apply (CATA) analysis.

The Correspondence Analysis showed that 15R1 had a stronger caramel flavor than the other samples. 15R2 was perceived as firm to the bite and creamy. 0R1 and 7.5R1 melt quickly than 15R1 and 7.5R2. The pleasant aftertaste of 0R1 was considered more intense than that of 0R2. In this analysis, it is evident that the 15R2, 0R1 and 7.5R1 samples were characterized with the positive attributes, while 0R2 and 7.5R2 were characterized with the negative attributes. Except for the fact that 15R1 was considered to have a stronger caramel taste, a positive attribute (Fig. 5). These results indicate that different schemes of turning in addition to pulp concentration can influence the formation of flavor compounds. In fact, cupuassu with higher concentration of pulp favored more the development of bacteria than that of yeasts (Ramos et al., 2020). Both communities of microorganisms present in the fermentation environment can produce precursor compounds that give the product different sensory characteristics, depending on the condition of fermentation (Ayad et al., 2001; Crafack et al., 2014; Ramos et al., 2016).

Fig. 6 shows the penalty analysis for each sample, demonstrating the mean impact on Overall Acceptance.

The attributes of the earthy and bad residual taste were negative consensus for all samples. In fact, these attributes reflect an inherent characteristic of the cupulate. Because of that, the pre-processing practices, mainly the fermentation step, can improve the raw material to highlight the positive sensory aspects of the cupulate and reduce the perception of negative attributes, such as earthy flavor. In the samples 0R1, 7.5R1, and 15R1, positive attributes (pleasant acidity, sweet taste, coffee flavor) that mask negative attributes stood out (Fig. 6).

3.3. *Quantitative Descriptive Profile (QDP)*

Table 3 shows the descriptor terms and references defined for the sensory profile evaluation of the six cupulates samples.

The panel of eleven assessors performed the QDP analysis about the six samples of cupulate based on terms descriptions (appearance, aroma, flavor, and texture) collected for QDP test. The panel defined the attributes aroma, basic tastes, flavor, and texture to describe the cupulate samples, along with a brief description. Most of the reference material was compound by cupuassu products (pulp, liquor) to facilitate the characterization of samples. Each descriptor was measured by means of a 9 cm unstructured scale anchored with intensity terms in the extremes like weak and strong.

The average scores of the three repetitions assigned by the assessors in relation to the attributes is shown in Table 4.

In the sensory evaluation by QDP for cupuassu flavor, coffee flavor and texture-melting attributes, there was no significant difference between samples ($p \geq 0.05$) (Table 4). Sample 15R2 presented the highest levels for residual bitterness; and 15R1 for fruity aroma, sweet taste, and caramel aroma. The amount of pulp present in 15R1 and 15R2 may have influenced the formation of flavor compounds, reflecting the characteristics of attributes in those samples due to the different turning schemes applied to them. Only samples 15R1 and 15R2 presented

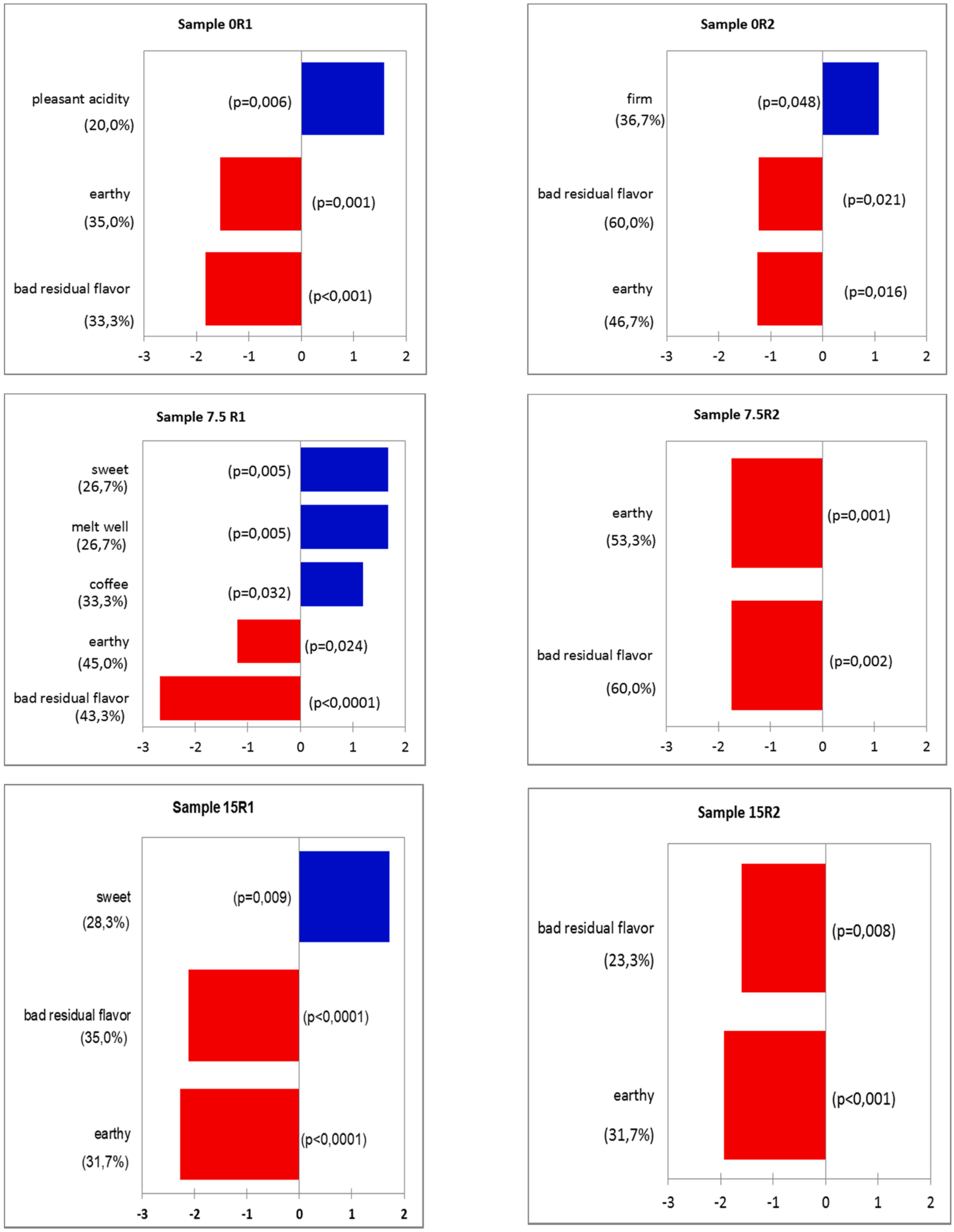

Fig. 6. Penalty Analysis - Mean impact on Overall Acceptance for all samples (0R1, 0R2, 7.5R1, 7.5R2, 15R1 and 15R2).

significant differences ($p \leq 0.05$) regarding the following attributes: fruity aroma, sweet taste, and residual bitterness. The turning scheme seems to have influenced the experiments with the higher amount of pulp for the formation of contrasting flavor compounds between them. The three cupulate samples obtained with R2 turning showed lower intensities of fruity aroma and sweet taste. Conversely, these samples showed slightly higher residual bitterness.

Fig. 7 shows Principal component analysis (PCA) on the attributes of the six cupulate samples by QDP tasters.

The principal component analysis (PCA) showed that samples 15R1 and 15R2 differed significantly in relation to roasted, caramel and fruity aroma. Sample 15R2 was more characterized by the coffee flavor and roasted aroma, while sample 15R1 was more characterized by the sweet taste and fruity aroma (Fig. 7). These results for samples 15R1 and 15R2 corroborate the those obtained in the consumer test, in which the R1 turning scheme during fermentation showed the best result for preference. Also, in the CATA analysis, the attribute caramel flavor was cited more times for sample 15R1, when compared to sample 15R2. The QDP results, in addition to characterizing the samples, also supported the understanding of consumers' perceptions about the cupulates.

3.4. Consumer Test × Quantitative Descriptive Profile (QDP)

In the Consumer Test, through the Preference Mapping, the cupulate

Table 3
Attributes, respective descriptor terms and references used for the sensory profile evaluation of cupulate samples.

DESCRIPTOR	DEFINITIONS	REFERENCES
AROMA		
Roasted	Aromatic score associated with roasted cupuassu.	**Weak:** Cupuassu liquor **Strong:** Cupulate whose liquor has gone through over-roasting
Caramel	Intensity of characteristic caramel aroma.	**Weak:** Cupuassu liquor **Strong:** Cupulate with an increase of 2% aroma identical to natural caramel (Citromax).
Coffee	Typical or characteristic aroma of coffee.	**Weak:** Cupuassu liquor **Strong:** Cupulate with an increase of 4% soluble coffee powder (Nescafé – Nestlé®)
Fruity	Aromatic score associated with yellow fruits	**Weak:** Cupuassu liquor **Strong:** Cupulate with an increase of 0.05% aroma identical to natural orange (Citromax)
BASIC TASTES AND FLAVOR		
Sweetness	Taste associated with sucrose.	**Weak:** Cupuassu liquor **Strong:** Condensed milk (Nestlé®)
Bitterness residual	Bitterness taste remaining in the mouth after ingestion.	**Weak:** Milk chocolate (Lacta – Mondeléz) **Strong:** Cupuassu liquor
Cupuassu flavor	Characteristic flavor of cupuassu.	**Weak:** Milk chocolate (Lacta – Mondeléz) **Strong:** Cupuassu pulp
Coffee flavor	Characteristic coffee flavor.	**Weak:** Cupulate with 50% of liquor **Strong:** Cupulate with an increase of 2% soluble coffee powder (Nescafé – Nestlé®)
TEXTURE		
Hardness	Force required for the first bite of the sample.	**Weak:** Milk chocolate (Lacta – Mondeléz) **Strong:** Compound (fancy chocolate) milk (Bel)
Melting	Characteristic associated with the texture and melting of samples at melting point close to tongue temperature.	**Weak:** Compound (fancy chocolate) or milk (Bel) **Strong:** Milk chocolate (Lacta – Mondeléz)

Table 4
Average scores given by the tasters to the attributes in the Quantitative Descriptive Profile (QDP) of the six cupulate samples.

ATTRIBUTE	0R1	0R2	7.5R1	7.5R2	15R1	15R2
Roasted aroma	3.0 ± 1.8^{b}	3.4 ± 2.1^{ab}	4.3 ± 1.9^{a}	4.3 ± 1.8^{a}	3.4 ± 2.3^{ab}	4.2 ± 2.1^{ab}
Caramel aroma	2.4 ± 1.8^{b}	2.7 ± 1.8^{ab}	2.6 ± 1.7^{ab}	2.7 ± 2.1^{ab}	3.4 ± 2.2^{a}	2.9 ± 2.1^{ab}
Coffee aroma	2.2 ± 1.6^{a}	2.1 ± 1.7^{a}	2.6 ± 1.8^{a}	3.0 ± 1.8^{a}	2.6 ± 1.9^{a}	2.7 ± 1.8^{a}
Fruity aroma	1.5 ± 1.4^{b}	1.7 ± 1.6^{ab}	$2.0 \pm 1.8^{a,b}$	1.4 ± 1.2^{b}	$2.4 \pm 2,0^{a}$	1.5 ± 1.3^{b}
Sweet taste	$4.8 \pm 2.3^{a,b}$	$3.9 \pm 2.1^{b,c}$	4.4 ± 2.3^{b}	4.3 ± 2.2^{b}	5.5 ± 2.0^{a}	3.3 ± 2.1^{c}
Residual bitterness	$3.0 \pm 2.0^{a,b}$	3.8 ± 2.3^{a}	3.3 ± 2.2^{a}	3.8 ± 2.0^{a}	2.1 ± 1.6^{b}	3.9 ± 2.1^{a}
Cupuassu flavor	2.7 ± 1.7^{a}	3.1 ± 1.7^{a}	3.1 ± 1.8^{a}	3.4 ± 1.9^{a}	2.9 ± 1.7^{a}	2.8 ± 1.8^{a}
Coffee flavor	3.2 ± 1.8^{a}	3.1 ± 2.1^{a}	3.3 ± 2.0^{a}	3.4 ± 1.8^{a}	2.5 ± 2.0^{a}	3.2 ± 2.0^{a}
Texture: hardness	$5.3 \pm 1.9^{a,b}$	5.8 ± 1.9^{a}	3.1 ± 2.0^{c}	$4.8 \pm 2.0^{a,b}$	$5.0 \pm 1.8^{a,b}$	4.5 ± 1.8^{b}
Texture: melting	4.6 ± 1.9^{a}	3.6 ± 2.1^{a}	4.2 ± 2.4^{a}	3.7 ± 2.3^{a}	4.3 ± 2.0^{a}	4.0 ± 2.2^{a}

Values are expressed as Mean (SD). Equal letters in the same line indicates no significant difference between the sample ($p > 0.05$) (Tukey's test).

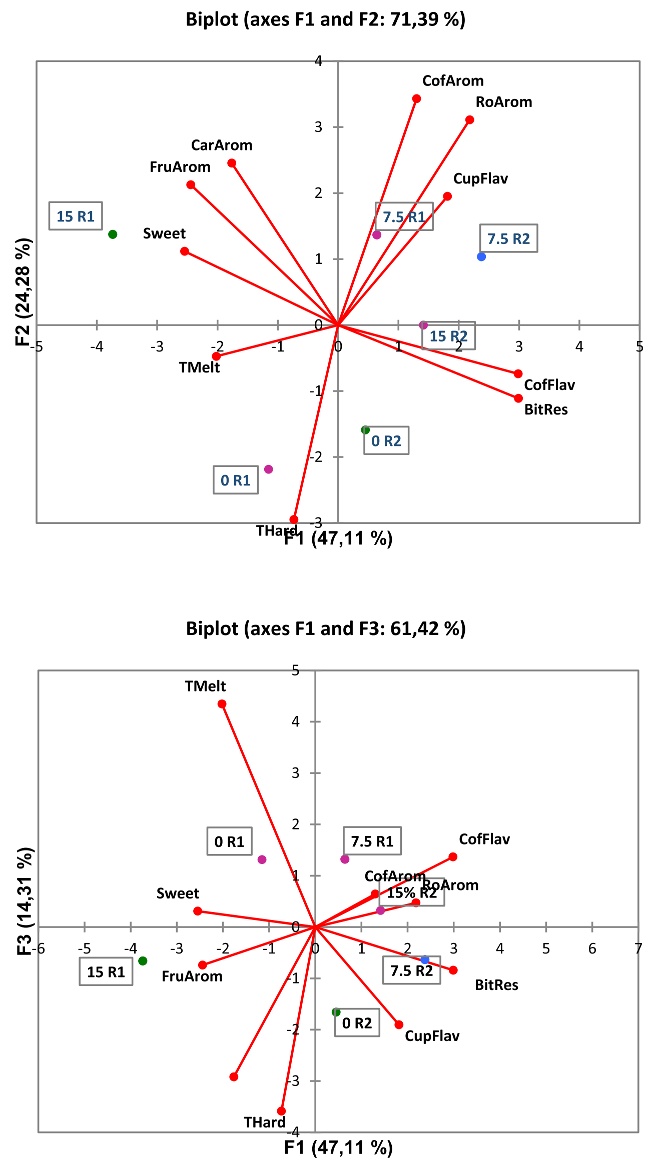

Fig. 7. Principal component analysis (PCA) on the attributes of the six cupulate samples by QDP tasters: Roasted Aroma (RoArom), Caramel Aroma (CarArom), Coffee Aroma (CofArom), Fruity Aroma (FruArom), Sweetness (Sweet), Bitterness Residual ((BitRes), Cupuassu Flavor (CupFlav), Coffee Flavor (CofFlav), Texture Hard (THard), Texture Melting (TMelt).

samples 0R1, 7.5R1 and 15R1 were preferred. In the penalty test, these same samples showed positive attributes that mask the negative attributes. This result may indicate that the R1 type (fixed) scheme of turning applied to the experiments during fermentation contributes to the development of flavor compounds with positive attributes. Establishing a comparison between the tests applied using untrained consumers in CATA and consumers trained in QDP, the results point to some sensory characteristics in common, noticed by the two groups. The cupulate samples produced from seeds with a higher amount of pulp (15R1 and 15R2) had a higher number of positive mentions, for the fruity and floral flavors. This characteristic was also perceived in the QDP in the same samples by the trained panelists, although the perception of intensity was higher for the 15R1 sample. These results reinforce the importance of maintaining a minimum amount of pulp in the seeds during fermentation to ensure the formation of important flavor compounds. However, further studies are still needed to reach an ideal fermentation pattern that involves both the form of turning to be applied and the ideal minimum amount of pulp during the process.

4. Conclusion

The present study demonstrated that cupuassu seeds, still considered a by-product of cupuassu fruit processing, have a potential application to produce a chocolate-like product (cupulate), if post-harvest steps, especially fermentation, are optimized. In this research, tests performed with trained panel and consumers showed their perceptions about cupulates, with peculiar sensory characteristics. Regarding negative attributes, the earthy taste and bad residual taste stood out as the main reasons for rejection of the product by the consumer. However, the results demonstrated that the cupulate has contrasting sensory attributes and reinforced the importance of maintaining a certain amount of pulp in order to highlight the desirable attributes and minimize the perception of the negative ones.

The higher presence of pulp impacted positively on the sensory attributes of cupulate, thus opening the perspective of maintaining some concentration of pulp during fermentation to obtain a product with acceptable sensory characteristics. The two sensory evaluation methods applied, using trained panel (to characterize the samples) and consumers (to express their preference and attributes) were useful to identify the attributes in common among the samples evaluated and the best fermentation conditions to obtain products with desirable sensory characteristics.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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