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FERMENTATION OF GREEN OLIVES USING STARTER CULTURE AND QUANTITATIVE DESCRIPTIVE ANALYSIS

FERMENTAÇÃO DE AZEITONAS VERDES UTILIZANDO CULTURA STARTER E ANÁLISE DESCRITIVA QUANTITATIVA

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Resumo: Este trabalho tem como objetivo avaliar as características sensoriais, utilizando a análise descritiva quantitativa (QDA), de diferentes formulações de azeitonas verdes fermentadas por 120 dias com *Lactobacillus plantarum* ATCC 8014 e/ou culturas espontâneas (microbiota indígena). A fermentação foi realizada com solução salina em diferentes concentrações de sal (4 a 12%) e sacarose (0,1 a 0,7%). Foram avaliadas as análises químicas da solução de salmoura (pH, ácido lático e cloretos) e microbiológicas das azeitonas fermentadas. Os atributos organolépticos das azeitonas fermentadas foram realizados por painel sensorial treinado em relação aos atributos visual, olfativo, cinestésico e paladar. Os resultados mostraram que a equipe treinada conseguia se lembrar e descrever atributos consensuais. Este estudo revelou que a fermentação de azeitonas com cultura espontânea com sacarose a 0,7% e NaCl a 12% apresentou os escores mais altos em termos de aroma, odor, crocância e menores escores no sabor amargo.
Palavras-chaves: *Lactobacillus plantarum, Spontaneous,* fermentação de azeitonas, análise descritiva quantitativa.
Abstract: *This work aim to evaluate the sensory characteristics, using the quantitative descriptive analysis (QDA), of different formulations of green olives fermented per 120 days with Lactobacillus plantarum ATCC 8014 or spontaneous (indigenous microbiota) cultures. The fermentation was undertaken with brine solution in different concentrations of salt (4 to 12%) and sucrose (0.1 to 0.7%). The chemistry analysis of the brine solution (pH, lactic acid and chlorides) and microbiological of the fermented olives were evaluated. The organoleptic attributes of fermented olives were assessed by trained sensory panel in relation to the attributes visual, olfactory, kinesthetic and taste. Results showed that the trained team could remember and describe consensual attributes. This study revealed that the olives fermentation with spontaneous culture using 0.7% sucrose and 12% NaCl, presented the high scores in terms of aroma, odor, crispness and lowest scores for the taste bitter.*
Keywords: *Lactobacillus plantarum, Spontaneous, Olive Fermentation, Quantitative Descriptive Analysis.*

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1 Introduction

The reason that aims development and accept fermented foods such as olives can be attributed to the necessity of preserving, allowing the development of healthier food product with better taste, aroma and enable greater business value (CORSETTI et al., 2012).

The olives fermentation usually occurs by traditional methods, however, they increasingly being replaced by new industrial processes, with chemical pretreatment, followed by fermentation (BOSKOU; CAMPOSEO; CLODOVEO, 2015). In this context, traditional Spanish-style green olive fermentation includes an alkaline pretreatment using sodium hydroxide, before add the fruit to ferment in the brine. The fermentation occurs by spontaneously lactic cultures (Lactobacillus, Leuconostoc, Streptococcus, Enterococcus, and Pediococcus genera) and/or added cultures, which included principally the *L. plantarum* (SABATINI; MUCCIARELLA; MARSILIO, 2008)and*L. Pentosus* (BLANA et al., 2014; DE CASTRO et al., 2002; PANAGOU et al., 2008; SERVILI et al., 2006). These cultures have characteristics to minimize the fruits deterioration and accelerating the pH decrease (MEDINA et al., 2008) *L. plantarumi*s one of the most common species found in vegetables and fruit, have predominant growth, homofermentative metabolism, and tolerance to salt, acid, and polyphenols (HURTADO et al., 2012). In particular, strains of *L. plantarum* ATCC 8014 have capacity to hydrolyze oleuropein (necessary to facilitate the transformation of olives into an edible product), compound responsible for bitter taste of olives (TSAPATSARIS; KOTZEKIDOU, 2004). The hydrolyze of the oleuropein occur by the activity of β-glucosidases with the release of glucose and aglycones, which degrade in the no-bitter phenols hydroxytyrosol and elenolic acid (BIANCHI, 2003).

Microorganisms, by their metabolic activities, contribute to develop special properties such as: taste, aroma, visual appearance, texture, shelf life and safety (CORSETTI et al., 2012; ROSA et al., 2016a, 2016b). In this way, the sensory evaluation has great importance to identify, qualify and quantify the product characteristics (NASRAWI, 2009). Descriptive sensory tests are amongst the most sophisticated tools, and involve the detection and description of qualitative and quantitative sensory components of a consumer product by trained panels of judges.

According to Stone (2012), the QDA is a sensory methodology that provides quantitative descriptions of products, based on the perceptions from a group of qualified subjects. It's a total sensory description, which taking account all the sensations perceived (visual, auditory, kinesthetic, olfactory, among others).

The present work aims to evaluate the sensory characteristics using QDA in different formulations of green olives fermented with *L. plantarum* or spontaneous (indigenous microbiota) cultures. A trained sensory panel assessed the organoleptic attributes (visual, olfactory, kinesthetic and taste) of olives fermented.

2 Material and Methods

2.1 Fruits and fermentation

Grappolo green olives were harvested in March 2012, an olive grove in Pelotas (a region situated in the south of Brazil), immediately transported to the laboratory. The

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perfect fruits were selected, following homogeneity criteria, without defect or spots on the shell, color and uniform size.

The olives were clean with water to remove any impurities and subsequently subjected to leaching with sodium hydroxide (NaOH) (1% w/v). The leaching was monitored for 7.5 h at room temperature (20-22°C), promoting the progression and diffusion to the olives pulp. To control this process, fruits were cut in the longitudinal and transverse directions with application of phenolphthalein drops until the spread around of two-thirds (2/3) of NaOH in the fruit pulp (KAILIS; HARRIS, 2019; MEDINA et al., 2008).

After alkaline treatment, the olives are washed with water. Later, 0.3% (v/v) acetic acid solution was added to the fruits for 15 min, aiming to neutralize (pH=7) the residual NaOH portion.

The brine were prepared with different concentrations of NaCl (4 and 12% w/v) and sucrose (0.1 and 0.7% w/v), for both fermentations (*L. plantarum* and spontaneous cultures), to obtain eight fermentation conditions (ROSA et al., 2016a). These percentages of salts have intentionally been used because they are more familiar to processors. These samples (treatments) are coded according to the concentration of NaCl and sucrose (%, w/v) and the fermentation ("L" for *L. plantarum* and "S" for spontaneous culture), as follow: S1 and L1 - 4% (w/v) NaCl and 0.1 % (w/v) sucrose; S2 and L2 - 12% (w/v) NaCl and 0.1% (w/v) sucrose; S3 and L3 - 4% (w/v) NaCl and 0.7% (w/v) sucrose; S4 and L4 - 12% (w/v) NaCl and 0.7% (w/v) sucrose.

Fermentation in anaerobiosis was carried out in glass reactors "kitassato" type, containing 800 g olives and 1.0 L brine solution. The reactors had lateral opening to allow the exit of carbon dioxide, with a hose immersed in distilled water. Two fermentation processes were investigated: spontaneous (indigenous microbiota of olives) and inoculated with *L. plantarum* (ATCC 8014).

In the fermentation for *L. plantarum*, pure and lyophilized strains (provided by Fiocruz) was used to prepare the inoculum, due to their ability to degrade oleuropein. In the reactivated strain was added the selective medium with *Man Rogosa and Sharpe* (MRS) broth. The broth was supplement with 4.5% of NaCl sterile solution (Sigma Aldrich, USA) (DE CASTRO et al., 2002; TSAPATSARIS; KOTZEKIDOU, 2004). The obtained solution was incubated at 35 °C overnight, until obtain a cellular mass around $1x10^9$CFU/mL. The inoculum was centrifuged at 2000 g/15min (Quimis model Q222T204), and afterwards the cellular mass was washed twice with saline solution, and resuspended in 10 mL of saline solution (0.85% w/v). The inoculum volume (10 mL) with $1x10^7$ CFU/mL were defined according to Tsapatsaris and Kotzekidou (2004) and Panagou et al. (2008). Subsequently, reactors were placed in a room, protected from the light at 18°C for 120 days, as demonstrate in Figure 1.

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Figure 1. Illustrative fermentation of green olives in glass reactors

2.2 Chemical and microbiological determinations

After 120 days of olives fermentation the following chemical characteristics were studied in the brine: pH was measured using a pHmeter (Hanna – HI 2212), titratable acidity is expressed as grams of lactic acid per 100 mL of brine and chlorides were analyzed by the Volhard method (GARCÍA; BALBUENA; FERNÁNDEZ, 1991). All the analyses were performed in triplicate.

The microbiological quality of olives at the end of the fermentation process was evaluated in relation to pathogenic *Salmonella* sp., and microorganisms indicators of microbial contamination (enumeration of thermotolerant coliforms, *Staphylococcus aureus* and sulphite reducing clostridia count), according to Brazilian legislation - Normative Instruction No. 30 (MAPA 2018).

2.3 Sensory analysis

The samples were profiled using QDA (STONE, 2012). Responsive to all the sensory properties of a product, the QDA methodology provides a complete word description for all product's sensory properties. To ensure that product is fully explained, it was necessary select and train the panel with session's of forty-hour.

Selection procedures, training, monitoring of assessors, definition of terminologies, ideal measuring range and evaluation of results were developed according to the standards used in descriptive terms and preparation of samples for training and memory, summarized a few propositions described by international standards (MAPA 2018; COI 2011; ISO 2012), previous research studies (APONTE et al., 2010; GALÁN-SOLDEVILLA; RUIZ PÉREZ-CACHO, 2012; GONZÁLEZ et al., 2007) and adapted to the availability of reagents and local products.

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The study protocol followed the ethical principles of sensory laboratory, approved by the ethical committee of the Regional University of Erechim, under the CAAE 03152512.7.0000.5351 registration number.

Panel of judges: The panelist's recruitment (67 judges) was carried out a personal contact with questionnaire and signature of consent. Candidates for judges were excluded if not have time for train and not like olives. The selection of the best candidates was carried out by discriminative tests (triangle test for difference testing): basic tastes (sweet, salty, bitter and acid), intensity of taste, mixture of basic tastes, odor and aroma recognition. The candidates that scored at least 80% of the correct answers were selected.

Sensory evaluation of olive fruits: For descriptive terms, the judges reviewed the sensory definitions, from pre-established standards, as similarities and differences between the olives offered for visual characteristics (appearance and color), olfactory (odor and characteristic olive aroma), kinesthetic (crispness and hardness) and taste (salty, bitter and acid). Thus, after defining the characteristics of the detailed description and setting up the sensory profile.

The references identified were presented to each judge. Specific train sessions were carried out for each attribute, which was assessed using a 9 cm unstructured scale (NBR14141, 1998). The extremes were defined in terms of minimum and maximum intensity of each specific attribute.

Four samples, with triplicates, were evaluated by 10 judges trained (age range 18-25 years). Among them, six women and four man that not smoke and not present history of allergies. Tests were performed in individual cabins with white light. Samples were served in plastic cups coded with three-digit numbers random for analyzes.

2. 4 Statistical analysis

The panel leader introduced the evaluation data and verified the performance of judges with the following tests: three representative samples of each descriptor were presented to the panel, and each assessor evaluated three repetitions of three samples. The individual results of assessors and of descriptors (acid, bitter, appearance, aroma, color, crispness, hardness, odor and salty) were statistically analyzed by analysis of variance (ANOVA) and Tukey's test at 5% significance level. Analysis of variance of 2 factors (sample and repetition) was performed for each panelist with respect to each attribute and the judges (Table 1) were selected according to values of significant Fsample ($p < 0.30$) and non-significant Frepetition ($p>0.05$). After was evaluated the concordance of the judges with the group. The judges were then selected based on their ability to discriminate among different samples, repeatability and agreement with the group.

The analysis was performed used the software Statistica 5.0 (StatSoftInc®, USA) with 95% significance level. Correlations between variables were established by correlation analyses with the XLSTAT (2014.5.03) statistical software package.

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Table 1-Values of pF_{sample} (%) - (d) and $pF_{repetition}$ (%) - (R) obtained by the final training judging

Descriptor	P_{level}	Assessors															
		1	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**	**11**	**12**	**13**	**14**	**15**	**16**
Appearance	F_{sample}	13.27	6.29	15.61	4.97	4.97	2.66	6.29	9.84	13.27	4.85	14.67	74.73	20.23	28.23	4.16	12.19
	$F_{repetition}$	41.51	25.79	29.65	91.82	91.82	38.45	25.79	35.71	41.51	26.22	34.88	61.88	30.5	21.02	27.42	41.23
Color	F_{sample}	22.8	8.91	20.24	11.62	11.62	39.63	8.91	29.16	22.8	15.73	22.65	20.31	18.09	9.59	12.49	0.63
	$F_{repetition}$	71.81	6.86	49.9	57.76	57.76	20.68	6.86	25.31	71.81	7.59	26.6	91.11	94.07	82.78	90.61	60.89
Odor	F_{sample}	0.22	9.76	6.94	21.42	21.42	28.1	9.76	6.56	0.22	14.36	1.22	38.46	24.07	15.78	12.96	17.68
	$F_{repetition}$	32.45	65.65	9.65	95.5	95.5	62.94	65.65	44.55	32.45	35.02	31.56	11.99	8.35	37.93	28.91	30.23
Aroma	F_{sample}	18.07	5.43	9.81	12.92	12.92	31.92	5.43	7.46	18.07	9.24	20.01	8.98	21.51	10.33	26.01	19.65
	$F_{repetition}$	77.33	12.51	92.59	68.92	68.92	28.89	12.51	68.76	77.33	61.54	63.4	7.75	14.37	45.6	25.51	6.36
Crispness	F_{sample}	57.5	28.3	75.86	5.23	5.23	11.97	30.43	5.04	23.56	2.82	22.67	1.97	10.21	3.15	4.24	24.84
	$F_{repetition}$	39.33	90.34	40.8	26.95	26.95	46.57	90.34	86.54	66.27	32.7	36.93	71.03	35.83	37.28	40.81	31.84
Hardness	F_{sample}	24.18	6.12	9.13	5.83	5.83	8.1	6.12	6.59	24.18	1.8	4.72	37.42	25.77	6.51	3.61	1.74
	$F_{repetition}$	74.28	80.31	8.68	67.7	67.7	50.01	80.31	9.1	74.98	89.81	11.11	41.96	50.22	20.06	19.62	78.28
Salty	F_{sample}	14.41	4.54	2.55	3.04	3.04	14.82	4.54	19.5	14.41	24.14	9.4	8.9	10.07	2.69	7.71	1.92
	$F_{repetition}$	13.66	56.47	1.92	50.14	50.14	23.38	56.47	59.46	13.66	41.05	70.3	7.54	9.88	30.32	96.36	18.89
Bitter	F_{sample}	66.48	0.34	25.78	3.35	3.35	10.3	0.34	0.13	6.24	0.61	6.36	4.05	16.98	1.02	4.3	1.16
	$F_{repetition}$	79.04	64.72	89.35	20.63	20.63	57.36	64.72	38.92	49.71	22.65	21.38	43.01	25.83	8434	26.83	79.58
Acid	F_{sample}	1.38	0.21	44.97	23.66	23.66	13.64	65.71	19.45	1.38	5.47	26.5	11.95	4.28	0.27	1.76	3.94
	$F_{repetition}$	51.68	15.96	4.76	36.68	36.68	15.36	89.33	11.87	51.68	12.37	93.71	60.41	16.48	24.43	20.78	21.4
Total	**D[a]**	2	0	2	0	0	2	2	0	0	0	0	3	0	0	0	0
	R[b]	0	0	2	0	0	0	0	0	0	0	0	0	0	0	0	0

D[a] = number of times that the judge not discriminate samples at a significance level (p F_{sample}< 30%);
R[b]= number of times that the judge not show repeatability of the significance level ($pF_{repetition}$> 5%);
All experiments are performed in triplicate

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3 Results and Discussion

3.1 Selection of olives formulations for sensory analysis

The brine solution of green olives fermented with spontaneous or *L. plantarum* cultures were evaluate in relation to pH, chloride and lactic acid (Table 2). All formulations showed significant difference ($p<0.05$) in the pH values. As described by (CODEX ALIMENTARIUS, 1981; COI, 2011, 2017), the final pH of naturally fermented olives must be equal or lower than 4.3. Only treatments S1, S3, S4, L2, L3 and L4 presented pH values less than 4.3, and thus complied with those standards.

Table 2-Values of pH, lactic acid and chlorides of green olives fermented using spontaneous (S) or *L. Plantarum* (L) cultures with different concentrations of sucrose and NaCl in the brine solution

Formulation	NaCl (%)	Sucrose (%)	pH	Latic acid (g 100/mL)	Chloride (%)
S1	4	0.1	4.22^{e}±0.02	$0.30^{c,d,e}$ ±0.01	$4.80^{d,e}$ ±0.01
S2	12	0.1	5.19^{a} ±0.02	0.18^{f} ±0.01	$11.01^{a,b}$ ±0.01
S3	4	0.7	4.43^{c} ±0.02	0.25^{e} ±0.551	$4.47^{d,e}$ ±0.28
S4	12	0.7	3.33^{h} ±0.02	$0.34^{b,c}$ ±0.01	$10.43^{b,c}$ ±0.01
L1	4	0.1	4.81^{b} ±0.01	0.25^{e} ±0.01	3.89^{f} ±0.01
L2	12	0.1	4.25^{d} ±0.01	$0.30^{c,d}$ ±0.66	$11.01^{a,b}$ ±0.01
L3	4	0.7	3.99^{f} ±0.01	$0.31^{b,c}$ ±0.33	$4.26^{e,f}$ ±0.01
L4	12	0.7	3.91^{g} ±0.03	0.39^{a} ±0.01	$10.79^{a,b,c}$±0.01

Means (± standard deviations) followed by different letters on columns represents significant difference at 5% level (Tukey's test).All experiments are performed in triplicate

The lactic acid in the brine solution in the formulations S1, S4, L2, L3 and L4 maintained the levels equal or above the minimum recommended by Consejo Oleícola Internacional (COI, 2011) which is 0.3% of free acidity expressed as lactic acid (g/100mL). In addition, was observed a significant difference ($p<0.05$) of the higher lactic acid value of (L4) in relation to the other formulations. For the chlorides is possible to observe that the formulations with 12% of NaCl (S2, S4, L2, and L4) do not present significant difference ($p<0.05$) among them.

In relation of the microbiological analysis, the *Salmonella* sp. was investigated due to the potential of food hazard. None of the analyzed formulations presented *Salmonella* sp. The absence of coliforms is a good indicator of the appropriate sanitary conditions of the food. Thermotolerant coliforms were detected in the formulations S1 and S3, with higher counts of 10^2 UFC/g. These samples should be considered unacceptable and improper for consumption. The Brazilian legislation, not establish limits for sulphite reducing clostridia. As some treatments contained 4% NaCl, and this condition can allow undesired growth of these microorganisms, the analyses were performed in olives samples and the results no showed viable colonies. Moreover, the counts of *S. aureus* in all samples were lower than the limit (5 x 10^2 CFU/g) established by the RDC No.12 (ANVISA, 2001).

In this way, considering the criteria of: pH lower than 4.3, lactic acid content above of 0.3%, according to Anvisa (2001), and also the results of microbiological analyses, visual and

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olfactory aspects, the formulations S1 and S3 did not attends the microbiological criteria, and S2, S3 and L1 did not meet the prerequisites of pH and lactic acid. Thus, these formulations were excluded of sensory analysis and the formulations chosen for further analysis were L2, L3, L4 and S4.

3.2 Sensory profile of the olives samples

For the sensory panel initially 67 judges participated in the recruitment and selection stages, and 16 judges developed a descriptive terminology (visual and descriptive analysis), olfactory (odor and characteristic olive aroma), kinesthetic (crispness and hardness) and taste (salty, bitter and acid) defining the similarities and differences among the sample, and 10 judges were considered trained for the QDA of fermented olives.

Variability between samples and attributes was observed comparing the sensory profiles of fermented olive (Table 3 and Fig. 2). The formulation L3 differs significantly ($p < 0.05$) considering the crispness and salty attributes. Moreover, the hardness in the formulation L3 did not differ from L2, but differed of the formulations L4 and S4. The formulation L3 showed higher average score for bitterness and lower for the crispness, hardness and acid, when compared with other formulations. These differences can be related to the low concentration of NaCl used in these formulations, because the salty taste can be suppressed bitter taste due to the use of lower concentrations of salt. In addition, in a previous study (ROSA et al., 2016a) verified that *L. plantarum* showed high counts, reaching fast adaptation at the beginning of fermentation (9 days), different of the spontaneous culture that require 20 days.

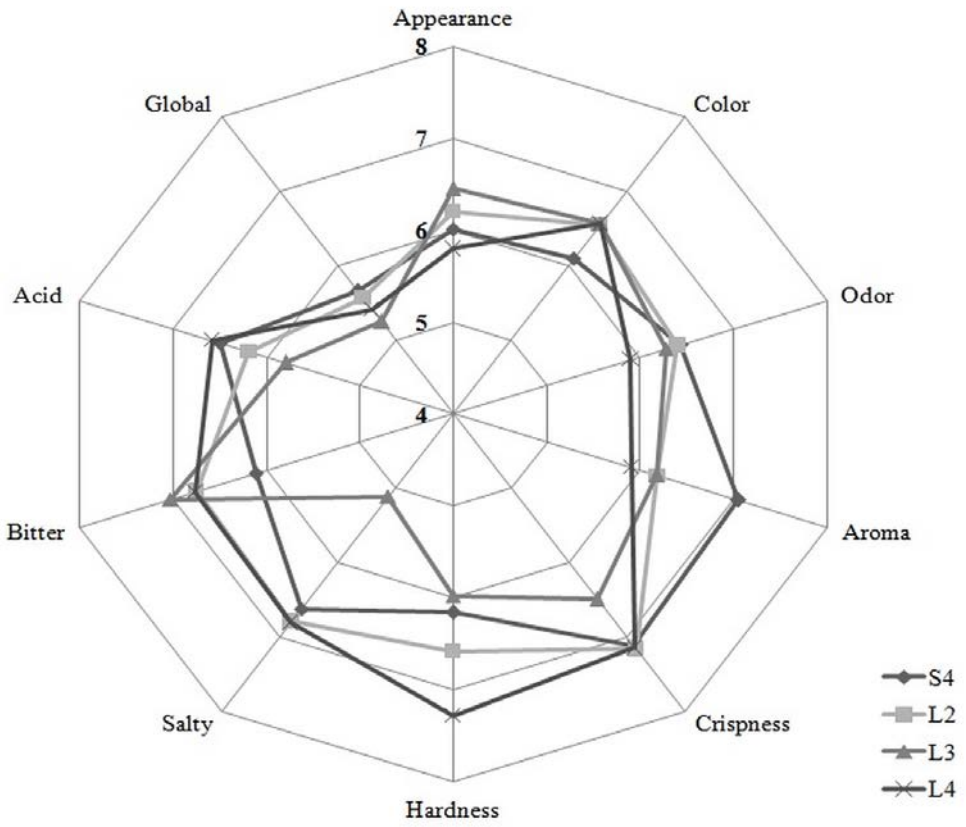

Figure 2. Sensory characteristics in fermented green olive

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Table 3-Features, profile obtained using QDA for olives fermented by the Spanish method

Attributes	Formulations			
	L2	L3	L4	S4
Acid	$6.17^{a} \pm 0.000$	$5.79^{a} \pm 0.110$	$6.57^{a} \pm 0.096$	$6.49^{a} \pm 0.035^{a}$
Bitter	$6.75^{a} \pm 0.170$	$7.02^{a} \pm 0.055$	$6.76^{a} \pm 0.020$	$6.41^{a} \pm 0.075$
Appearance	$6.16^{a} \pm 0.050$	$6.45^{a} \pm 0.165$	$5.80^{a} \pm 0.315$	$6.01^{a} \pm 0.065$
Aroma	$6.15^{a} \pm 0.190$	$6.18^{a} \pm 0.030$	$5.91^{a} \pm 0.125$	$6.36^{a} \pm 0.375$
Color	$6.54^{a} \pm 0.080$	$6.56^{a} \pm 0.195$	$6.55^{a} \pm 0.010$	$6.09^{a} \pm 0.205$
Crispness	$7.15^{a} \pm 0.050$	$6.50^{b} \pm 0.150$	$7.15^{a} \pm 0.020$	$7.32^{a} \pm 0.065$
Hardness	$6.61^{ab} \pm 0.045$	$5.99^{b} \pm 0.185$	$7.29^{a} \pm 0.040$	$6.95^{a} \pm 0.290$
Odor	$6.39^{a} \pm 0.065$	$6.28^{ab} \pm 0.220$	$5.90^{b} \pm 0.200$	$6.44^{a} \pm 0.215$
Salty	$6.79^{a} \pm 0.100$	$5.12^{b} \pm 0.210$	$6.82^{a} \pm 0.265$	$6.84^{a} \pm 0.070$

Means (± standard deviations) followed by different letters in same line are significantly different at 5% level (Tukey's test).All experiments are performed in triplicate

Moreover, central cognitive effects occur when different taste stimuli are mixed together and the perceived intensity of one or more stimuli is diminished by the perception of the others (KEAST, 2008). The bitter sensation depends on the presence of substances that come from the fruit, which are mainly polyphenols, this might therefore be more intense in preparations, which the debittering is incomplete (LANZA, 2013). For odor, the formulation L4 was significantly different from L2 and S4 but equal to L3, obtaining the lower score.

Formulation S4 (Fig. 2 and Table 3) showed the larger scores in relation to aroma, odor, crispness but lower for bitterness and color. These attributes are crucial for the acceptance or rejection of table olives, and must maintain the level of bitterness and fruitiness (KAILIS; HARRIS, 2019). The formulations L2, L3, L4 and S4 did not differ significantly at 5% level in terms of the acid, bitter, appearance, aroma and color attributes. Thus, the formulation S4 (12% w/v NaCl and 0.7% m/v sucrose) was the most accepted by the attributes evaluation of the judges.

Agglomerative hierarchical cluster analysis indicated that attributes were clustered into three different segments as illustrated in the dendrogram (Fig. 3 a).

The dendogram shows the high proximity between the bitter, salty and acid attributes, and between aroma, color, appearance, hardness, crispness and odor. Thus, to improve the presentation of the results has conducted two analyzes of major components using the multivariate analysis between variables and factors.

Principal component analysis (Fig. 3b and c) showed that the first and second principal components explained, respectively, 58.94 and 54.40% of the observed variance. It was possible to observe that the samples are clearly separated into different groups. The means is a segregation of the samples according to their organoleptic characteristics, thus confirming the general homogeneity of the samples analyzed.

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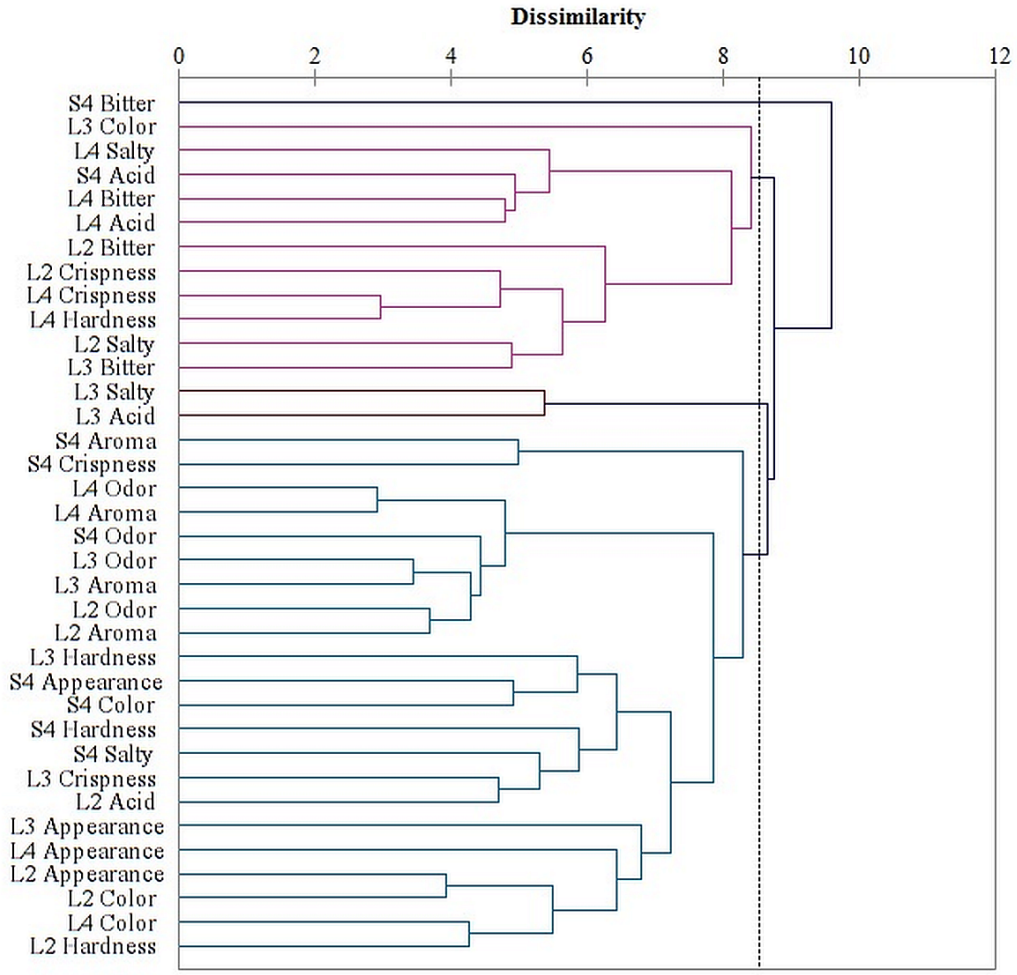
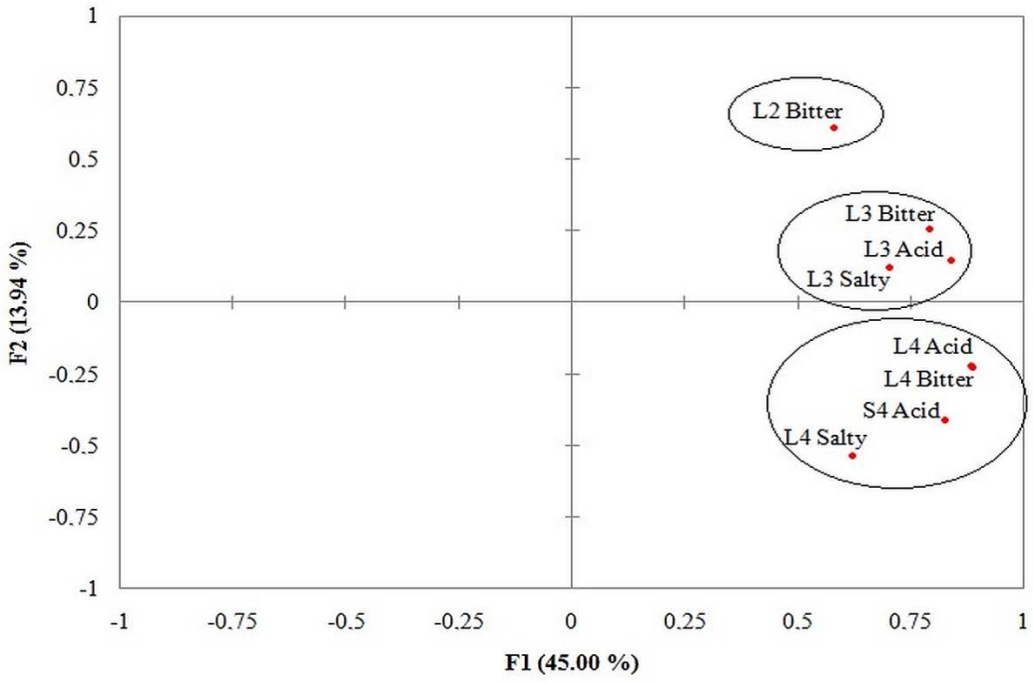


(a)



(b)

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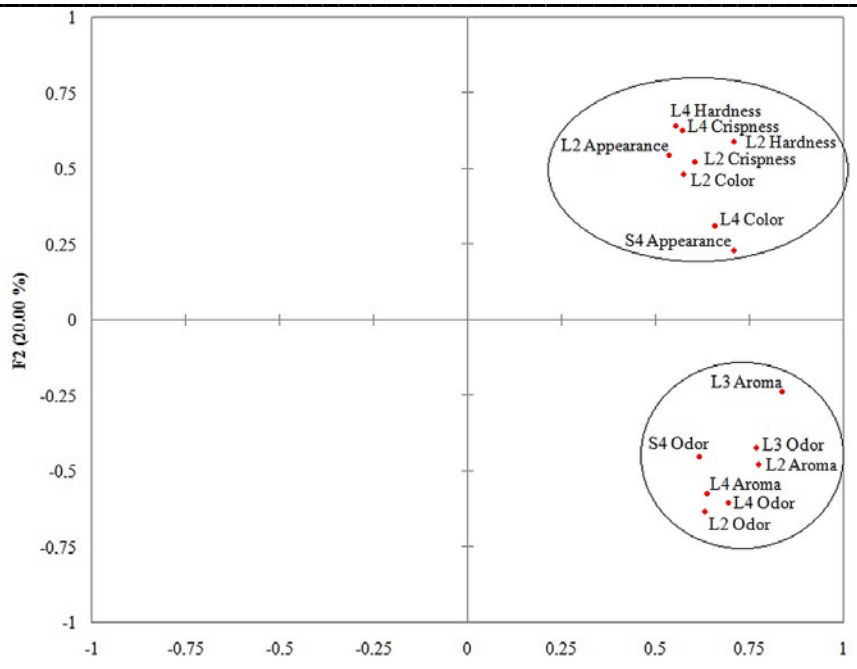

(c)

Figure 3. Hierarchical cluster analysis dendrogram for sensory attributes into groups of similar perceptions of fermented green olives. Dotted line denotes level of dissimilarity along which the three segments (clusters) were selected (a), PCA on sensory bitter, salty and acid (b) and PCA on sensory attributes of hardness, crispness, color, odor, and appearance (c).

Also is possible to observe that the trained team could remember and describe consensual attributes, according of the homogeneity of numerical values and low values of standard deviation.

Some authors provided a broader perspective to the use starter cultures in fermentation process; ensure safe and best sensory characteristics. However, the presence of spontaneous species result in special sensory character to the olives, which can be inhibited by the use of starter culture (APONTE et al., 2010; RANDAZZO et al., 2012, 2014; ZAGO et al., 2013). These results corroborate with those of the present study, where the formulation S4 (12% w/v NaCl, 0.7% w/v sucrose and fermentation with spontaneous culture) was scored as the better formulation by sensorial analysis.

Mourad and Nour-Eddine (2006), observed in naturally fermented green olives, in the first fermentation stage (15 days), wherein was characterized by the growth of heterogeneous microorganisms, including aerobic bacteria, coliforms, staphylococci, lactic acid bacteria, lactobacilli, enterococci, psychrotrophs, lipolytic bacteria and yeasts, and after 60 days lactic acid bacteria and yeast were the predominate population in relation to the initial heterogeneous bacterial flora. Ruiz-Barba and Jiménez Díaz (2012) cited that spite of some available commercial inoculums, the use of starter cultures not have a habitual practice in the olive processing industry. However, it is possible to obtain more effective starter cultures with detailed selection of strains and relevant technological characteristics.

4 Conclusion

After the training steps, 10 tasters were considered trained to evaluate the QDA of fermented olives for attributes visual characteristics (appearance and color), olfactory (odor and characteristic olive aroma), kinesthetic (crispness and hardness) and taste (salty, bitter and acid).

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The olives fermentation with spontaneous culture using 0.7% sucrose and 12% NaCl, presented the high scores in terms of aroma, odor, crispness and lowest scores for the taste bitter, and the formulation with *L. plantarum* using 4% (w/v) NaCl and 0.7% (w/v) sucrose showed higher average score for bitterness and lower for the crispness, hardness and acid. Demonstrating that both fermentation processes have significant advantages in the sensorial characteristics of fermented olives.

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