Effects of natural antioxidants in processing and stability of italian type salami during storage

Efeitos de antioxidantes naturais no processamento e estabilidade do salame italiano durante o armazenamento

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ABSTRACT

The aim of the study was to elaborate formulations of Italian type salami by adding natural antioxidant of green tea (Camellia sinensis) and to evaluate the stability during the shelf life. Formulations (F1, F2 and F3) were elaborated varying the concentrations of green tea (0.008 to 0.016%), sodium erythorbate (0.048 to 0.097%), sodium nitrite (0 to 0.012%), sodium nitrate (0 to 0.016%), curing salt (0 to 0.189%) and dehydrated glucose (0.589 to 0.959%), keeping fixed the concentrations of swine meat, bacon, water, refined salt, pepper and starter culture. To verify oxidative stability, the moisture, water activity, sodium nitrate and nitrite, pH, acidity, TBARS and product acceptance were evaluated during 120 days of storage. F1 stood out in terms of acceptance, with a higher initial and residual concentration of nitrates and nitrites up to the 80th day of storage and less lipid oxidation, that has 0.016% green tea, 0.012% sodium nitrite, 0.013% sodium nitrate, 0.081% sodium erythorbate and 0.806% dehydrated glucose in its formulation. The results indicated that the addition of green tea, associated with sodium nitrate/nitrite and sodium erythorbate reduced the formation of TBARS and did not affect the sensory quality of the product. Thus, this natural antioxidant can be easily used in Salami to improve quality and provide safer products.

Keywords: green tea, oxidation, matured meat product, shelf life.

RESUMO

O objetivo do estudo foi elaborar formulações de salame tipo italiano, adicionando antioxidante natural de chá verde (*Camellia sinensis*) e avaliar a estabilidade durante a estocagem. As formulações (F1, F2 e F3) foram elaboradas variando as concentrações de chá verde (0,008 a 0,016%), eritorbato de sódio (0,048 a 0,097%), nitrito de sódio (0 a 0,012%), nitrato de sódio (0 a 0,016%), sal de cura (0 a 0,189%) e glicose desidratada (0,589 a 0,959%), mantendo fixas as concentrações de carne suína, bacon, água, sal refinado, pimenta e cultura inicial. Para verificar a estabilidade oxidativa, foram avaliadas a umidade, atividade da água, nitrato e nitrito de sódio, pH, acidez, TBARS e aceitação do produto durante 120 dias de armazenamento. F1 se destacou em termos de aceitação, com maior concentração inicial e residual de nitratos e nitritos até o 80° dia de armazenamento e menor oxidação lipídica, que possui chá verde de 0,016%, nitrito de sódio de 0,013%, eritorbato de sódio de 0,081% e glicose desidratada de 0,806% em sua formulação. Os resultados indicaram que a adição de chá verde, associada a nitrato/nitrito de sódio e eritorbato de sódio, reduziu a fomação de TBARS e não afetou a qualidade sensorial do produto. Assim, este antioxidante natural pode ser facilmente usado no salame para melhorar a qualidade e fornecer produtos mais seguros.

Palavras-chave: chá verde, oxidação, produto carneo curado, prazo de validade.

1 INTRODUCTION

One of the main causes of meat deterioration is the oxidative processes that occur in the conversion of muscle to meat, at meat processing or during storage (Cunha et al., 2018). The oxidative stability of meat depends on various intrinsic and extrinsic factors such as the concentration of pro-oxidants/antioxidant balance, degree of unsaturation, pH, and temperature as

well as the composition of the protein and lipid fraction, which vary among different animal species (Oswell et al., 2018).

Lipid oxidation of meat products has deleterious effects on the organoleptic properties of these foodstuffs as well as the digestibility of key nutrients (<u>García-Lomillo</u> et al., 2017), causing changes in sensory (color, texture and flavor) and nutritional quality (Karakaya, et al., 2011; Shah et al., 2014; Oswell et al., 2018).

Additionally, some products that are especially susceptible to oxidative chain reactions (Jongberg et al., 2017). Consequently, it is often necessary to rely on antioxidants to prolong shelf life and preserve product quality. The use of antioxidants in food products is controlled by regulatory laws of a country or international standards (Karre et al. 2013).

In meat products, antioxidants are widely employed to prevent oxidation. The antioxidants can be of synthetic or natural origin. Are synthetic antioxidants the ascorbic acid, butylated hydroxytoluene (BHT), butylated hydroxyanisol (BHA), as well nitrites and nitrates (Formanek et al., 2001; Biswas et al., 2004; Jayathilakan et al., 2007; Pereira et al., 2015).

But the demand for natural antioxidants, especially of plant origin has increased in the recent years due to the growing concern among consumers about these synthetic antioxidants because of their potential toxicological effects (Oswell et al., 2018; Ribeiro et al., 2019). Among the plants with antioxidant properties used in meat products, the oregano (*Origanum vulgare*) (Shan et al., 2009; Prete et al., 2020), the rosemary (*Salvia rosmarinus*) (Fernandez-Lopez et al., 2003) and the sage (*Salvia officinalis*) (Mc Carthy, 2001) have components with recognized antioxidant activity such as carnosol, carnosic acid and rosmarinic acid. Studies also report the use of yerba mate (*Ilex paraguariensis*) (Terra et al., 2008; Beal et al., 2011, Coró et al., 2019), green tea (*Camellia sinensis*) and black tea (*C. sinensis*) (Rababah et al., 2011) that have flavonoids and phenolic acids in their composition.

In this sense, the main of the study was to elaborate formulations of Italian type salami by adding natural antioxidant of green tea (*C. sinensis*) and to evaluate the oxidative stability of the product during shelf life.

2 MATERIAL AND METHODS

2.1 ELABORATION OF SALAMI FORMULATIONS

Three formulations of Italian salami were elaborated in an industry located at Upper Uruguay of Rio Grande do Sul, under traditional processing conditions in accordance with current legislation (BRASIL, 2019). In the formulations (F1, F2 and F3) the concentrations of green tea

were (F1 and F2 - 0.016% and F3 - 0.008%), sodium erythorbate (F1- 0.097%, F2 - 0.081% and F3 - 0.048%), sodium nitrite (F1 and F2 - 0.012% and F3 - absent), sodium nitrate (F1 - 0.016%, F2 - 0.013% and F3 - absent), curing salt (F1 and F2 - absent and F3 - 0.189%) and dehydrated glucose (F1 - 0.959%, F2 - 0.806% and F3 - 0.589%), keeping fixed the concentrations of swine meat, bacon, water, refined salt, pepper and starter culture (Lyocarni SBM-11), which consists of *Staphylococcus xylosus, Staphylococcus carnosus*, and *Lactobacillus sakei* (anti-listeria), which in combination ensures a uniform and controlled production of fermented sausages.

The raw materials were previously weighed, and then the swine meat (7°C) was ground, with an 8 mm disc and the bacon ($\leq 5^{\circ}$ C) chopped into cubes. After, the meat and bacon were mixed in a mixer and also added the liquid and powders seasonings. After, the mass was embedded in a collagen artificial casing, previously hydrated. The pieces were hung on aluminum poles and sent to the smokers. The smoking of the pieces was carried out with natural smoke, remaining in these conditions until reaching an internal temperature of the smokehouse 38° C ± 1°C and an internal temperature of the product 36° C ± 1°C, for approximately 32 h.

The maturation and drying process was carried out in a curing room, remaining until reaching water activity ≤ 0.90 and a final pH between 4.8 and 5.6, defining the end of maturation. Maturation occurred in 28 days. After, the products were washed, removing the artificial casing, the samples were kept in a dry and cool place at room temperature, vacuum-packed and packed in secondary cardboard boxes.

2.2 CHARACTERIZATION OF FORMULATIONS AND EVALUATION OF OXIDATIVE STABILITY

For the characterization and evaluation of oxidative stability, random samples (n = 3) from each formulation were selected. The analyzes of protein and total fat were carried out after the stage of smoking and on the 1st day of shelf life, and the moisture, water activity, pH and acidity were performed on initial mass and during the shelf life of the products (20-120 days). Nitrate and nitrite were evaluates after smoking and during the shelf life of products and lipid oxidation (substances reactive to thiobarbituric acid - TBARS) during the shelf life of products (20-120 days).

2.3 ANALYTICAL DETERMINATIONS

The moisture, pH and acidity contents were determined according to the methodology described by AOAC (2012). The levels of nitrate and nitrite were obtained following the methodology described by BRASIL (2005). The substances reactive to thiobarbituric acid (TBARS)

were performed according to the methodology described by Raharjo et al. (1992), modified by Wang et al. (2002), following recommendations by Shahidi et al. (1997). The water activity was performed on a water activity meter (AquaLab, series, 3TE).

2.4 MICROBIOLOGICAL ANALYSIS

Microbiological analyzes were carried out in the 120th, on the industry laboratory in accordance with Normative Instruction 60, of December 23, 2019 (BRASIL, 2019) for thermotolerant coliforms, positive coagulase *Staphylococcus*, *Listeria* and *Salmonella* sp.

2.5 SENSORY ANALYSIS

The study was approved by the Research Ethics Committee (URI-Erechim), registered at Brasil Plataform under number 91375418.0.0000.5351.

The sensory analysis were performed by 30 untrained judges, of both sexes, with age group of 20 to 50 years old, employees of an industry located in the Upper Uruguay of Rio Grande do Sul. The sensory evaluation of the formulations (F1, F2 and F3) of Italian type salami was performed during the shelf life (1, 20, 40, 60, 80, 100 and 120 days) using a consumer acceptance test - structured hedonic scale with 9 points (9 - I liked it a lot and 1 - I unliked it a lot) (Dutcosky, 2013). The samples were prepared by cross-sectioning the salami pieces with an average thickness of 3 mm, discarding the ends, being distributed in a balanced way in plastic plates coded with random numbers of three digits.

2.6 STATISTICAL ANALYSIS

The results (n = 3) obtained were statistically treated by analysis of variance (ANOVA), followed by the averages differences comparison by the Tukey's test, with a 95% confidence level, using the Statistica 7.0 software. For the Pearson correlation analysis and Principal Component Analysis (PCA), the XLSTAT 2020 program, Free version, was used.

3 RESULTS AND DISCUSSION

The values of water activity (aw) and moisture (Table 1) decreased (p < 0.05) during storage and differed between the formulations, where formulation F3 showing greater water activity and moisture until the end of storage, because F3 contains in its composition a higher concentration of green tea (0.024%), lower sodium erythorbate (0.048%) and dehydrated glucose (0.589%) and contains 0.189% curing salt.

Formulations F1 and F2 on the 1st day of storage (28 days of maturation) and F3 on the 20th day of storage were within the limits stipulated by Brazilian legislation (BRASIL, 2000), which defines that salami ready for consumption must present a maximum water activity of 0.92 and a maximum moisture of 35% after the maturation period.

The values of aw (0.88) on the 28th day of maturation (1 day of storage) (Table 1), are similar to those obtained by Wang et al. (2018), that obtained 0.93, 0.85 and 0.83 in samples of salami, chinese dry-cured sausage and chinese smoked-cured sausage, respectively. Settanni et al. (2020) when studying fresh swine sausage containing rosemary and green tea extracts during retail verified aw of 0.83 in 45 days of maturation. Fieira et al. (2015) found 0.894 to 0.899 in samples of Italian type salami prepared with different salts and initial cultures. A possible explanation for the aw observed in the present study, may be related to the composition of the samples (higher or lower proportions of the different components), as well as to the salami maturation site (Kunrath et al., 2017).

Alamprese et al. (2016) found for fresh swine salami moisture of 32 to 34% on the 20th day of storage and also found a linear decrease with storage. As the relative humidity in the fermentation and ripening rooms is constantly kept lower than the aw of salami, there is a difference in vapor pressure that causes the removal of moisture through the out side layers of the products. The water loss has to occur at the right speed and to be as uniform as possible in order to avoid case hardening that is negative for both safety and texture of salami. In addition, if a product is not dried at a suitable speed, the desired firmness (or loss in weight) will be obtained in longer time and every day of extended drying or ripening is very costly (Fernández et al., 2000).

Table 1. Values of water activity (aw), moisture, protein, fat, nitrate and nitrite of formulations of Italian type Salami during storage.

Period		aw*	Moisture (%)*				
I CI IOU	F1	F2	F3	F1	F2	F3	
Mass	0.961 ^{aA}	0.963 ^{aA}	0.963 ^{aA}	_	_	_	
111455	(0.001)	(0.002)	(0.002)		_	_	
Smoke	0.956 ^{abAB}	0.959 ^{aAC}	0.952 ^{bB}	54.88 ^{aA}	53.16 ^{Ca}	53.35 ^{Ba}	
Smoke	(0.002)	(0.002)	(0.002)	(0.006)	(0.006)	(0.0001)	
1**	0.886 ^{Af}	0.880^{Af}	0.893 ^{Ag}	34.06 ^{Cc}	34.64 ^{Bb}	36.52 ^{Aa}	
-	(0.012)	(0.010)	(0.003)	(0.006)	(0.010)	(0.0001)	
20	0.856 ^{Cgh}	0.861 ^{Bg}	0.868^{Aj}	33.82 ^{Cc}	34.04 ^{Bc}	35.15 ^{Ac}	
20	(0.001)	(0.004)	(0.001)	(0.010)	(0.000)	(0.006)	
40	0.841 ^{abI}	0.842 ^{abH}	0.853 ^{Ak}	33.68 ^{Bd}	33.57 ^{Ce}	34.75 ^{Ae}	
40	(0.008)	(0.008)	(0.003)	(0.010)	(0.010)	(0.010)	
60	0.851 ^{bcGHI}	0.846 ^{cH}	0.867^{aJ}	32.50 ^{cG}	34.00 ^{bD}	34.48 ^{aF}	
00	(0.005)	(0.004)	(0.006)	(0.006)	(0.001)	(0.006)	
90	0.859 ^{cGH}	0.867 ^{bG}	0.878^{aHI}	32.51 ^{cG}	32.82 ^{bG}	34.77 ^{aD}	
80	(0.002)	(0.004)	(0.003)	(0.006)	(0.001)	(0.006)	
100	0.861 ^{cG}	0.861 ^{cG}	0.882 ^{aHI}	33.34 ^{bE}	33.18 ^{cF}	34.06 ^{aG}	
100	(0.002)	(0.002)	(0.002)	(0.006)	(0.001)	(0.006)	
100	0.848 ^{cHI}	0.847 ^{cH}	0.862^{aJ}	32.78 ^{bF}	32.44 ^{cH}	34.06 ^{aG}	
120	(0.001)	(0.002)	(0.001)	(0.010)	(0.010)	(0.006)	
Protein (%)			. ,	Fat (%)			
Smoke	19.74 ^{cB}	20.54 ^{aB}	19.91 ^{bB}	18.46 ^{cB}	19.52 ^{bB}	20.10 ^{aB}	
	(0.006)	(0.006)	(< 0.001)	(0.006)	(0.006)	(< 0.001)	
1**	29.91ªA	29.77 ^{bA}	27.67 ^{cA}	26.08 ^{bA}	25.68 ^{cA}	27.26 ^{aA}	
	(0.006)	(0.006)	(< 0.001)	(0.010)	(0.006)	(< 0.001	
		Nitrate (ppm)*		Nitrite (ppm)*			
a 1	137.44 ^{aA}	128.45 ^{bA}	32.48 ^{cA}	1.45 ^{cC}	0.76 ^{bD}	2.17 ^{aB}	
Smoke	(0.026)	(0.010)	(0.006)	(0.010)	(0.006)	(0.006)	
	54.89 ^{aB}	15.50 ^{bB}	5.84 ^{cbB}	1.14 ^{cE}	1.31 ^{bB}	1.63 ^{aC}	
1**	(0.006)	(0.006)	(0.006)	(0.006)	(0.010)	(0.006)	
• •	43.03 ^{aC}	8.31 ^{bC}	5.08 ^{bC}	0.94 ^{bD}	0.84 ^{aC}	1.09 ^{aD}	
20	(0.017)	(0.012)	(0.006)	(0.006)	(0.010)	(0.0001)	
4.0	20.32 ^{aD}	4.59 ^{bE}	3.67 ^{cD}	1.15 ^{cF}	0.55 ^{aF}	0.91 ^{bF}	
40	(0.006)	(0.006)	(0.021)	(0.0001)	(0.006)	(0.0001)	
	16.56 ^{aE}	4.78 ^{cD}	1.57 ^{cE}	1.74 ^{bA}	1.02 ^{cE}	1.83 ^{aA}	
60	(0.006)	(0.010)	(0.006)	(0.006)	(0.0001)	(0.0001)	
80	13.81 ^{aF}		0.00 ^{aF}	1.21 ^{aB}	0.96 ^{bA}	0.93 ^{cE}	
	(0.006)	$0.00^{\rm cF}(0.000)$	(0.000)	(0.006)	(0.001)	(0.006)	
		E	0.00^{aF}	0.00^{aG}	0.00 ^{aH}	0.00 ^{aG}	
100	$0.00^{\mathrm{aG}}(0.000)$	$0.00^{\mathrm{aF}}(0.000)$	(0.000)	(0.000)	(0.000)	(0.000)	
	0.00 ^{aG}	-E	0.00^{aF}	0.00 ^{aG}	0.00 ^{aG}	0.00 ^{aG}	
120	(0.000)	$0.00^{\mathrm{aF}}(0.000)$	(0.000)	(0.000)	(0.000)	(0.000)	
	(0.000)	1	(0.000)	(0.000)	(0.000)	(0.000)	

* Mean ± standard deviation followed by the same lowercase letters in lines and uppercase in columns do not differ statistically at 5 % level (Tukey's test). **28 days of maturation

In meat products, the protein content has several roles that determine the yield, quality, structure and sensory attributes (Olivo; Shimokomaki, 2006). According to the results (Table 1).

1), there is a small variation in the protein content from 19.74 to 20.54% after smoking, and at 28 days of maturation values from 27.67 to 29.91%. Kunrath et al. (2017) when studying the application and evaluation of propolis as a natural antioxidant in Italian type salami verified values that varied from 28.86 to 32.78% 28 days after production, values similar to those obtained in this study.

Lipid is considered an important constituent of this meat product, as it gives the products juiciness, flavor and aroma (Olivo; Shimokomaki, 2006). The formulations showed a fat level of 18.46 to 20.10% after smoking, and at 28 days of maturation, values of 25.68 to 27.26% (Table 1). These variations are considered normal in this type of product, due to the presence of ingredients such as bacon and the distribution of these in the mass of the product part, which was also verified by Zanardi et al. (2004) and Settanni et al. (2020). The formulations after maturation are in accordance with current legislation (BRAZIL, 2000) which fixes for proteins the minimum of 25% and for fat maximum of 32%.

As shown in Table 1, it can be seen that for nitrate there was a significant difference (p <0.05) between the formulations after smoking and on the 1st day of storage (corresponding to 28 days of maturation). F1 showed a significant difference (p <0.05) from the other formulations until the 80th day of storage, and the values obtained for such formulation are higher when compared with F2 and F3. On the 100th day of storage, it appears that there is no significant difference (p > 0.05) between the formulations, presenting residual zero for nitrate. However, for nitrite was observed a significant difference (p <0.05) between the formulations up to the 80th day of storage and on the 100th and 120th the samples show residual zero for nitrite.

Nitrite is responsible for color development and also for antimicrobial action. In fermented meat, nitrite is reduced to nitric oxide through the action of the enzyme nitrite reductase and when the pH of the medium is between 5.6 and 6.2 (Ritter, 2016; Gaziano et al., 2007). In the process of curing meat products, the levels of nitrite and nitrate tend to decrease after some time of storage, because both nitrite and nitrate are converted into other substances.

The pH values (Table 2) of salami changed during the maturation period, showing a higher value at the beginning of manufacture (6.02 to 5.81). The decrease of pH until 28 days of maturation probably occurred due to the presence of *Lactobacillus* in the initial culture added to the formulation, obtaining a pH between 5.14 to 5.10. Marangoni; Moura (2011) studied Italian type salami with addition of essential oil *Coriandrum Sativum* L. as an antioxidant activity and found pH values 5.10 to 5.29, close to those observed in the present study. Italian type salami is evaluated with the addition of *C. Sativum* L. essential oil as an antioxidant activity.

In general, the formulations shown an increase in pH until the 40^{th} day of storage and in the period of 40 to 80^{th} days there is a decrease in pH, and an increase in acidity, which may be due to the production of organic acids (Terra et al., 2006). However, in the period of 100 and 120^{th} of storage there is an increase (p <0.05) in the pH for F3 and F2.

The decline in the pH value during the first days of ripening (fermentation) is very important due to the inhibition of undesired bacteria, rate of conversion of colour, and formation of desired flavour in dry-fermented sausages (Bozkurt, 2006). Furthermore, such acidification has positive technological aspects, including the faster drying and improved texture as a result of protein denaturation and coagulation and increased reddening through the formation of nitric oxide and nitrosyl myoglobin (Ammor; Mayo, 2007; Wang et al., 2018). Acidification promotes a decrease in pH until reaching the isoelectric point of proteins, release of water in sausages, thus reducing water activity and promoting properties such as feasibility (Pinto et al., 2001).

F3 showed higher (p < 0.05) values for TBARS after the 40th day of storage, which had lower levels of green tea (0.008%), sodium erythorbate (0.048%) and glucose (0.589%) and 0.189% curing salt. The formulations F1 and F2 with the addition of 0.016% of green tea showed a significant reduction in oxidative activity compared to formulation F3, demonstrating the effectiveness of green tea against oxidative damage.

Period _	рН			Acidity (g/100g)			TBARS (mg malonaldehyde/kg)		
	F1	F2	F3	F1	F2	F3	F1	F2	F3
Mass	5.99 ^{aA}	6.02 ^{aA}	5.81 ^{bA}		-	-	-	-	-
	(0.085)	(0.031)	(0.010)	-					
After	4.96 ^{bDE}	5.05 ^{aCE}	5.12 ^{aDE}	10.41 ^{bH}	10.00 ^{cH}	14.43 ^{aH}		-	-
smoke	(0.015)	(0.017)	(0.046)	(0.006)	(0.000)	(0.006)	-		
1**	5.14 ^{aD}	5.10 ^{bCE}	5.11 ^{abD}	19.00 ^{bG}	19.21 ^{aF}	15.22 ^{cG}	0.034 ^{cE}	0.049 ^{aE}	0.043 ^b
	(0.040)	(0.012)	(0.012)	(0.006)	(0.006)	(0.000)	(0.004)	(0.005)	(0.005
20	5.37 ^{abC}	5.31 ^{bB}	5.55 ^{aBC}	20.93 ^{aF}	18.29 ^{cG}	20.00 ^{bF}	0.075 ^{cC}	0.097 ^{aC}	0.082 ^b
	(0.076)	(0.026)	(0.140)	(0.006)	(0.006)	(0.006)	(0.004)	(0.002)	(0.002
40	5.64 ^{aB}	5.50 ^{bB}	5.66 ^{aAB}	33.85 ^{aA}	26.87 ^{bD}	23.84 ^{cD}	0.062 ^{cD}	0.081 ^{bD}	0.105ª
	(0.096)	(0.035)	(0.021)	(0.006)	(0.010)	(0.010)	(0.008)	(0.016)	(0.016
(0)	5.39 ^{aC}	5.38 ^{aB}	5.39 ^{aC}	23.38 ^{aE}	21.67 ^{cE}	21.77 ^{bE}	0.120 ^{bA}	0.122 ^{bA}	0.140 ^a
60	(0.036)	(0.055)	(0.045)	(0.010)	(0.012)	(0.006)	(0.010)	(0.02)	(0.009
80	4.75 ^{cH}	4.96 ^{bCDE}	5.09 ^{aDHI}	27.49 ^{aD}	27.35 ^{bC}	25.91 ^{cC}	0.102 ^{bB}	0.104 ^{bB}	0.124ª
80	(0.006)	(0.020)	(0.012)	(0.006)	(0.006)	(0.006)	(0.004)	(0.028)	(0.065

Table 2. Values of pH, acidity and TBARS of formulations of Italian type Salami during storage.

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100	5.01 ^{cDEF}	5.13 ^{bBC}	5.27 ^{aCDH}	30.64 ^{aB}	28.78 ^{cB}	28.83 ^{bA}	0.117 ^{bA}	0.121 ^{bA}	0.136 ^{aC}
	(0.012)	(0.026)	(0.006)	(0.006)	(0.006)	(0.006)	(0.012)	(0.018)	(0.026)
120	5.11 ^{cD}	6.04 ^{aA}	5.51 ^{bBC}	30.27 ^{aC}	29.89 ^{bA}	28.65 ^{cB}	0.103 ^{bB}	0.104 ^{bB}	0.145 ^{aA}
	(0.110)	(0.190)	(0.070)	(0.006)	(0.006)	(0.006)	(0.008)	(0.010)	(0.014)

* Mean \pm standard deviation followed by the same lowercase letters in lines and uppercase in columns do not differ statistically at 5% level (Tukey's test). **28 days of maturation

There are reports in the literature that natural antioxidants reduce the formation of TBARS more than BHT (Tang et al., 2001; Bozkurt, 2006). Finding the natural equivalent of synthetic antioxidants is important for human health because some synthetic antioxidants have carcinogenic activity and their usage in the food industry is more than the natural antioxidants (Bozkurt, 2006).

Marangoni; Moura (2011) with analysis of TBARS indicated that the coriander oil was more effective than BHT as an antioxidant in Italian type salami, where the use of only coriander oil was more efficient than in conjunction with BHT, presenting a significant effect greater than the BHT in relation to the amount of malonaldehyde present in the sample.

Terra et al. (2003), evaluated the action of different concentrations (0.5% and 1.0%) of yerba mate antioxidant and synthetic antioxidant (BHA) (0.025%) in improving the quality of Italian type salami and found that 0.5% of yerba mate extract and BHA showed the best results. Then to use 0.5% of yerba mate extract can be used as a substitute for the antioxidant BHA in Italian type salami.

The antioxidant capacity of these extracts is largely due to the presence of phenolic compounds of various classes that are inherent to the plants (Jiang; Xiong, 2016). Phenolic compounds function predominantly as primary antioxidants via hydrogen atom donation, and quench reactive free radicals. The delocalization of the radical species in the phenol significantly reduces the reactivity of the ensuing radical compound (Liebler et al., 1990). In addition to scavenging free radicals, plant phenolics can also function as secondary antioxidants and chelate metal ions (Estévez; Heinonen, 2010). The antioxidative property of green tea is due to the presence of catechins, apicatechins, epicatechin gallate, epigallocatechin, and epigallocatechin gallate (Higdon; Frei, 2003; Bozkurt, 2006; Nimse; Pal, 2015; Oswell et al., 2018; Schilling et al., 2018).

The results of the microbiological analyzes presented results for Thermotolerant Coliforms (<1.0 x 10 CFU/g), positive coagulase *Staphyococcus* (Absence/25g), *Salmonella* (Absence/25g) and Listeria (Absence/25g) in the formulations F1, F2 and F3, stored at room temperature for 120 days, and are on the standars of they Normative Instruction 60, of December 23, 2019 (Brazil, 2019), thus being suitable for human consumption. Consequently, the effective combined factors such as low pH, reduced aw, nitrate or nitrite addition has contributed to the preservation of cured meat

products (Rocelle et al., 1996), associated with the quality of raw material and the Good Practices of Manufacturing adopted during processing.

Table 4 and Figure 1 shows Pearson's correlation and principal components analysis (PCA) of physical-chemical and sensory variables of the formulations (F1, F2 and F3) in 0, 20, 40, 60, 80, 100 and 120th day of storage, respectively.

In Figure 1 the variables are represented as vectors, the longer the vector, the better the explanation of the variability between the variables. The first (PC1) and second (PC2) dimensions explained 64.04% of the total variance. The main component 1 (PC1) accounted for 36.23%, while the main component 2 (PC2) accounted for 27.81%. There is discrimination between the formulations of Salami, mainly F3 in relation to F1 and F2. F3 is the closest to the variables of moisture and aw at 0, 20, 40, 60 and 80th day of storage. F2 had the largest TBARS from 40 days of storage. F1 stood out in terms of acceptance (Table 3, Figure 1), with a higher initial and residual concentration of nitrate and nitrite on the 80th day of storage, because presenting in its formulation 0.016% green tea, 0.012% sodium nitrite and 0.013% sodium nitrate and also 0.081% sodium erythorbate and 0.806% dehydrated glucose, which positively influenced the sensory characteristics and oxidative stability of the product during storage.

Dave of storage	Formulations						
Days of storage	F1	F2	F3				
0	6.83 ^{cA}	6.60 ^{aB}	6.57 ^{aB}				
20	6.72 ^{dA}	6.52 ^{bB}	6.47 ^{bB}				
40	7.19 ^{aA}	6.20 ^{aB}	6.33 ^{cB}				
60	7.12 ^{aA}	6.09 ^{dC}	6.40 ^{bB}				
80	7.13 ^{aA}	6.44 ^{cB}	6.31 ^{cB}				
100	6.93 ^{bA}	6.00^{dB}	6.16^{dB}				

Table 3. Average score of tasters for the acceptability of formulations of Italian type Salami during storage.

* Mean \pm standard deviation followed by the same lowercase letters in columns and uppercase in lines do not differ statistically at 5% level (Tukey's test).

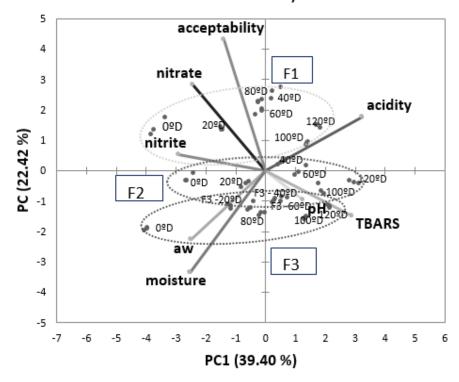
Variables	Nitrite	Nitrate	рН	aw	Moisture	Acidity	TBARS	Accep.
Nitrite	1							
Nitrate	0.366	1						
pH	-0.128	-0.072	1					
aw	0.228	0.141	-0.455	1				
Moisture	0.387	0.014	0.013	0.588	1			
Acidity	-0.635	-0.364	0.115	-0.507	-0.650	1		
TBARS	-0.372	-0.636	0.065	-0.261	-0.347	0.442	1	
Accep.	0.385	0.474	-0.369	-0.068	-0.212	0.048	-0.383	1

Table 4. Pearson correlation matrix for the variables nitrite, nitrate, pH, aw, moisture, acidity, TBARS and acceptability (accep.) of salami formulations on 0, 20, 40, 60, 80, 100 e 120th day of storage.

Values in bold the correlation > 0.47, significance level =0.05.

The values obtained by Pearson's correlation (Table 4) confirm the relationship between the parameters observed in PCA (Figure 1), where the nitrite concentration was negatively correlated (p < 0.05) with acidity (r = -0.635), indicating that by decreasing the nitrite content, the acidity of the product increases.

Figure 1. Principal Component Analysis (PCA) of salami formulations on 0, 20, 40, 60, 80, 100 e 120th day of storage.



PC1 e PC2: 61.81 %)

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Nitrate also is negatively correlated (r = -0.636) with TBARS and weakly correlated with acceptability (r = 0.474), indicating that by decreasing nitrate there is a tendency to increase oxidation and decrease acceptability. In addition, there is a positive correlation between aw and moisture (r = 0.588), and acidity is negatively correlated with nitrate (r = -635), aw (r = -0.507) and moisture (r = -0.650).

Thus, the results obtained indicated that the addition of green tea, associated with sodium nitrate/nitrite and sodium erythorbate reduces the formation of maloaldehydes, maintaining the oxidative stability and acceptance of the product during storage.

4 CONCLUSIONS

The natural antioxidant (green tea) was used in Italian Type Salami and its effect on safety and quality was investigated during the maturation periods. The addition of green tea, associated with sodium nitrate / nitrite and sodium erythorbate significantly reduced the formation of TBARS and did not affect the sensory quality of the product. Thus, this natural antioxidant can be easily used in Salami to improve quality and provide safer products.

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COMPLIANCE WITH ETHICAL STANDARDS

The authors declare no potential conflicts of interest.

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