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Vegetable "*Salami*" with High Nutritional and Functional Properties

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Abstract

Vegetables are strongly recommended in human diet since their consumption is closely related to the decrease of cardiovascular diseases. Despite these advantages, many people do not like vegetables as it, as opposed to some foods that are very attractive for their sensorial characteristics but nutritionally unbalanced. Among them, '*Salami*' are typical Italian fermented products made up of pork meat and back-fat that are very appreciated for their flavour and taste but present a high content of fatty saturated acids and cholesterol.

This research aimed at the study and realization of new fermented vegetable *Salami*, with an appropriately balanced composition of nutrients and able to simulate the meat *Salami* in terms of colour and consistency. Those characteristics were obtained by choosing vegetables with good sensory and structural characteristics and optimizing the ripening processing that, for this type of *Salami*, was 18 days at 25°C and 75% RU in early days and 45% RU in the final stage of ripening. During ripening stage, lactic bacteria concentration was more than 108 CFU/g. This value represents the minimum threshold to consider a food as probiotic in vitro. The chromatic characteristics of vegetable *Salami* during ripening showed a reduction of L^* (from 50.27 to 39.02) and b^* indexes (from 44.92 to 30.59) caused by the gradual dehydration of vegetable matrix. As a matter of fact, a sudden increase of shear stress, hardness, chewiness and springiness of the slices was recorded as a function of maturation.

Keywords: Vegetable *Salami*; Extra virgin olive oil; Formulation; Rheological characteristics; Acid lactic bacteria; Ripening

Introduction

'*Salami*' is typical Italian fermented products made up of pork meat and backfat with the addition of other ingredients such as salt, pepper and garlic according to regional recipes. Commercial *Salami* usually contains 70% meat and 30% fat [1]. High content of saturated fatty acids and cholesterol in *Salami* formula is due to the use of pork back fat.

Many epidemiological studies describe the correlation between consume of this typology of products and the increase of cardiovascular diseases [2].

In order to overcome the high intake of animal fat, some researches have investigated the substitution of fat with soy proteins [3], vegetable oils [4], extra virgin olive oil [5] and carbohydrates [6]. Alternatively, the consumption of vegetables are strongly recommended in the human diet since they are rich in antioxidants, vitamins, dietary fibers and minerals that reduce the incidence of cardiovascular, metabolic and neuro vegetative diseases [7]. Many people do not like vegetables as it, as opposed to some foods that are very attractive for their sensorial characteristics but nutritionally unbalanced. Vegetables are generally consumed as fresh or processed by thermal treatments, freezing, salting or pickling. The lactic acid fermentation may be considered as a simple and valuable biotechnology for maintaining and/or improving the safety, nutritional and sensory properties of vegetables besides their shelf-life [8]. A large number of lactic acid bacteria starters are usually used for dairy and meat fermentation, while only a few cultures have been used for vegetable fermentation. Among them, *Lactobacillus plantarum* is the commercial starter most frequently used in the fermentation of cucumbers, cabbages and olives [9].

Nevertheless, on the market there are not vegetable *Salami*, therefore, for this typology of food are necessary both an innovative process and an innovative formula based on vegetables that could simulate the meat phase and the animal fat.

The aim of this research was the realization of a new product similar to *Salami*, but with high nutritional and functional properties using only vegetables both for fat and meat fraction.

Materials and Methods

Preparation of *Salami*

Different colored vegetables that reminded the chromatic characteristics of meat *Salami* were purchased in local market (Foggia, Italy): tomatoes, carrots and red peppers were chosen to simulate the red meat phase, while fennels, courgettes and artichokes simulated the fat fraction. White-green vegetables were also chosen for their structural characteristics, i.e. a good consistency due mainly to the presence of fibers.

Red-yellow vegetables were wash, cut and blanched for 3 minutes in boiling water (vegetables/water ratio 1:4) [10]. After blanching, the pH of each sample was determined. In order to improve the color and taste of the obtained cream, the following ingredients were added: potatoes puree, tomato double concentrate, extra virgin olive oil, paprika, black pepper, salt and soy lecithin as emulsifier. Finally, different percentages of xanthan gum and its blend with wheat flour or sodium caseinate were used to improve the texture of vegetable *Salami*.

White-green vegetables, that have to simulate the "discontinuous phase" i.e. the fat fraction of *Salami*, was prepared washing, cutting (0.125 cm^3), and acidifying the vegetables with 0.1% three different acids (lactic, acetic and citric acids) (vegetable/water ratio 1:4 w/w). The type and the percentage of acids were chosen to assess their acidifier ability, in order to decrease the pH up to 5.5 [11]. The analytical determinations on formula were carried out at least in triplicate.

Vegetable *Salami* were formulated with 70 % red-yellow vegetables and 30% white-green vegetables that were stuffed into synthetic casings (previously rehydrated in hot water with the addition of salt), using a pilot plant.

The obtained *Salami* were inoculated with 1013 CFU/g *Lactobacillus plantarum*.

pH determination

PH of each samples was determined using a pH meter CRISON mod. Basic 20 (Allen, Spain). The analysis was performed on 10g of samples with the addition of 10 ml of distilled water homogenized with a blender BUHLER ML 12400 (Germany) for 10 sec at 6000 rpm.

Rheological analysis

Apparent viscosity of the red-cream was determined using a rheometer Rheoscope1 equipped with a probe-type flat - plate stainless steel (6 cm diameter, 1 mm distance between the plates), and a thermostating system Thermo Electron Corporation Haake DC30. Analysis was conducted at 25°C by gradually increasing rotational stress applied from 0 to 300 s^{-1} followed by a gradual reduction from 300 to 0 s^{-1} to verify and eliminate the thixotropic effects. Experimental data were acquired and processed using the software Haake Rheowin (version 4.41.0006, Australia) and interpreted by the equation of Herschel - Bulkley (HB):

$$Herschel-Bulkley: \sigma = \sigma_0 + k.\dot{\gamma}^n$$

Where σ is the shear stress, k is the coefficient of consistency, n is the flow index and γ is the amount of deformation (share rate).

The apparent viscosity was expressed in Pascal*second (Pa*s) and the measurements were performed in duplicate. Preliminary tests showed that, in our case, γ = 1, while K and n derived by software interpolation and were replaced in the equation.

Water activity

Water activity (a_w) of samples was measured at 30°C using a dew point hygrometer (Aqualab, mod. CX-2, Decagon Devices Inc., Pullman, Washington, 99163, USA). a_w determinations were carried out in duplicate.

Colour measurements

Colour assessments were performed with a Minolta Chroma Meter CR-300 (Minolta, Osaka, Japan) by CIE Lab system (International Commission d'Eclairage). Results were expressed as L^* (brightness), a^* (redness) and b^* (yellowness). The measurements were carried out five times.

Mechanical proprieties

The texture of white-green vegetables was evaluated according to Sharoba [12] with minor modifications by Instron 3343 (Instron LTP, High Wycombe, UK) equipped with a load cell set at 500 N. Data were elaborated by Merlin software ver. 5.5 as the shear stress, that is the ratio of the force to the maximum peak and the surface subjected to stress (N/mm^2). Moreover, during ripening of vegetable *Salami*, measurements on slices of 3.5 diameter and 2 cm thickness were carried out. Data were expressed as hardness (N), chewiness (kg^*mm) and springiness (mm). Data referred to vegetable *Salami* were compared with meat *Salami*.

Starter inoculation and ripening

Vegetable *Salami* was inoculated with 1.65^*10^{13} CFU/g *Lactobacillus plantarum*. The obtained *Salami* was ripened at 25°C for eighteen days in a ventilated oven mod. TWENTY-LINE (VWR, Milan) at 75 % RU/3 days and 45% RU/15 days

Lactic acid bacteria count

Samples (10 g) were suspended in sterile 0.1% (w/v) peptone-water solution and homogenized with a Stomacher Lab-blender 400 (PBI International) for 2 min at room temperature. Mesophilic lactic acid bacteria were determined on MRS agar (Oxoid) at 30 °C for 48-72 h under anaerobiosis.

Sensory analysis

At the end of the ripening, vegetable *Salami* was judged by six trained panelists. To each judge were presented two samples: one was a vegetable preserve purchased on the market and the other was a slice of vegetable sausage at the end of ripening. Judges had to assign to each sample a numeric value from 1 (unpleasant) to 10 (excellent), ordering them according to the sensory characteristics listed on the card, with the exception of the judgment relating to off-flavors, in which a low score indicated a positive evaluation of the sample.

Statistical analysis

All experimental data were analyzed by one-way ANOVA, performed by the Stat Software (Stat soft ver. 6.0, Tulsa, OK, USA) and compared using Fisher's test. The standard deviations were calculated using Excel software (Office XP, Microsoft Corporation, USA).

Results and discussion

Table 1 shows the pH value, chromatic characteristic and apparent viscosity of singular ingredients used in the formulation of the "red phase" of vegetable *Salami*. The obtained results have shown that vegetables such as tomato and pepper, kept a pH value close to safety (4.5) even after the blanching treatment (Table 1A) [13]. The colorimetric indexes showed the typical values of red-yellow vegetables. Carrots have presented the highest value of yellow index (47.86 ± 1.13) that can be attributed to high content of carotenoids that determine the typical yellow/orange/red color of fruits and vegetables [14]. Redness and yellowness were also very high in pepper, while, as reported in literature, the blanching treatment had amplified the brightness of all vegetables. As expected, the a_w value of all samples were not significantly different. Data related to apparent viscosity however, have pointed out that pepper had poor rheological

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characteristics (viscosity values less than 10 Pa*s) compared to tomato, carrot and potato that had a viscosity values ranged from 107 to 192.91 Pa*s. That information was very useful to determine the percentages of vegetables to use in the formula. Those percentages were chosen after preliminary determination (data not shown). The viscosity of final formula with different percentages of vegetables spices and extra virgin olive oil was 151 Pa*s (Table 1B).

The pH value of mixed vegetables was significantly influenced by the addition of concentrated tomatoes sauce, that determined a significant increase of a* and a slight decrease of brightness too.

The summary of those data permitted to choose that formula for the addition of white green vegetables.

A Blanched Vegetables	PH	aw	L*	a*	b*	Apparent Viscosity (pa*s)	K	n
Pepper	4.89 ± 0.01 a*	0.997 ± 0.002 a	40.12 ± 0.77 a	29.67 ± 0.86 a	38.92 ± 0.86 a	9.09 ^{a}	8.64	3 2
Tomato	4.49 ± 0.02 ^{b}	0.998 ± 0.001 a	39.03 ± 0.57 ^{b}	21.51 ± 0.67 ^{b}	33.14 ± 1.26 ^{b}	107 ^{b}	1427.0 0	0 2
Carrot	6.45 ± 0.03 ^{c}	0.996 ± 0.001 a	49.19 ± 0.65 ^{c}	7.91 ± 0.27 ^{c}	47.86 ± 1.13 ^{c}	160.2 ^{c}	1234.0 0	0 2
Potato	6.11 ± 0.02 ^{d}	0.998 ± 0.001 a	62.58 ± 0.40 ^{d}	-9.44 ± 0.10 ^{d}	23.67 ± 0.40 ^{d}	192.91 ^{d}	369.50	1 9
B								
Formula	4.44± 0.01	0.991 ± 0.003	42.52 ± 0.41	19.25 ± 0.73	40.56 ± 1.78	151 ± 65.05	1564.5 0	0 2

Table 1: Values of pH, water activity (a_w), brightness index (L*), red index (a*), yellow index (b*) and apparent viscosity (Pa*s) of the red-yellow vegetables just blanched (A) and the formula with vegetables, spices and extra virgin olive oil (B). (a,b,c,d the values with the same letter are not significantly different).

Table 2 showed the pH values and the colorimetric determinations of the white-green vegetables before and after the blanching treatments (with or without the addition of acids).

The acidifying-blanching carried out on white-green vegetables with the three acids evidenced that lactic acid and citric acid had a higher effectiveness on lowering of pH in all vegetable matrixes studied. PH reduction may confer some organoleptic characteristic changes such as flavor, taste, color, and texture [15,16]. Among these, the possibility of imparting a sour taste and chlorophyll degradation are the main drawbacks of acidifying treatments. The most important sensorial characteristic of acidulants is sourness but organic acids may confer other non-sour flavors as well as bitterness and astringency [17]. It may be expected acetic and citric acids have a greater congruency respectively with vinegar and citrus flavored systems rather than lactic acid that is commonly associated with milk/yogurt systems. A more deep knowledge concerning the effects of organic acids on sour taste of model systems and vegetables food are necessary to give the possibility to control the organoleptic impact on the final products [13]. As reported in literature, the color of green vegetables is mainly determined by the chlorophyll pigments that changes during blanching treatments by migration into the blanching water [18].

A				
Fresh vegetables	**pH**	**L***	**a***	**b***
Fennel	5.82 ± 0.006 ^{A}	84.10 ± 0.90 ^{A}	-3.26 ± 0.59 ^{A}	10.48 ± 2.74 ^{A}
Courgette	6.34 ± 0.006 ^{B}	87.01 ± 1.24 ^{A}	-4.01 ± 3.40 ^{A}	27.16 ± 1.98 ^{B}
Artichoke	6.05 ± 0.010 C	77.29 ± 10.71 ^{B}	-4.01 ± 3.40 ^{A}	25.71 ± 5.12 ^{B}
B **Blanched vegetables**				
Fennel	6.06 ± 0.034 A	85.07 ± 0.56 a	-4.31 ± 0.61 A	9.10 ± 1.47 a
Courgette	6.53 ± 0.009 ^{B}	88.41 ± 1.04 ^{b}	-4.81 ± 2.00 A	24.17 ± 1.88 ^{b}
Artichoke	7.20 ± 0,016 C	79.13 ± 4.71 ^{c}	-4.52 ± 2.11 A	23.61 ± 3.22 ^{b}
C **Blanched-acidified vegetables (0.1% acid)**				
Fennel				
Acetic acid	5.86 ± 0.016^{A}	69.35 ± 4.26^{a}	-3.45 ± 0.53^{A}	9.53 ± 2.03^{a}
Lactic acid	5.31 ± 0.017^{B}	66.78 ± 4.51^{a}	-3.00 ± 1.08^{A}	5.62 ± 1.32^{b}
Citric acid	5.31 ± 0.017^{B}	67.67 ± 3.11^{a}	-2,50 ± 0,36^{A}	4.83 ± 0.85^{b}
Courgette				
Acetic acid	6.53 ± 0012^{A}	78.51 ± 1.52^{B}	-10.38 ± 0.76^{B}	46.01 ± 1.91^{C}
Lactic acid	6.33 ±0.005^{B}	71.29 ±3.94^{CD}	-12.21 ±0.10^{C}	39.95 ±1.14^{D}
Citric acid	5.70 ±0.019^{C}	71.96 ±0.67^{C,D}	-10.26 ±0.32^{B}	35.35 ±3.16^{e}
Artichoke				
Acetic acid	6.66 ±0.012^{A}	65.10 ±9.98^{e,f}	-6.73 ±2.95^{D}	31.10 ±4.54^{f}
Lactic acid	5.69 ±0.014^{B}	74.87 ±2.53^{f}	-4.84 ±1.29^{D}	22.00 ±5.12^{g}
Citric acid	5.71 ±0.037^{B}	58.18 ±5.04^{e,f}	-4.26 ±0.71^{D}	25.55 ±6.00,^{f,g}

Table 2: Values of PH, brightness index (L*), red index (a*) and yellow index (b*) of white-green vegetables blanched with and without acidification. (a,b,c,d,e,f,g and A,B,C,D the values with the same letter are not significantly different).

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Results related to brightness index (L*) as affected by different blanching treatments showed an initial increase of L* values. Some authors attributed that behaviour to air removal around the fine hairs on the surface of the plant and to the expulsion of air between the cells of vegetable tissues [19]. The presence of different acids in hot water, reduced drastically L* index: that is probably due to the high penetration of acidified water into the cells.

Figure 1 reports the values of shear stress (N/mm^2) in blanched and acidified vegetables. After acidifying-blanching treatment, vegetables did not excessively lose their texture. In particular, samples that preserved the maxim consistency were those blanched with lactic acid. In view of that, they were used to simulate the "fat fraction" of *Salami*.

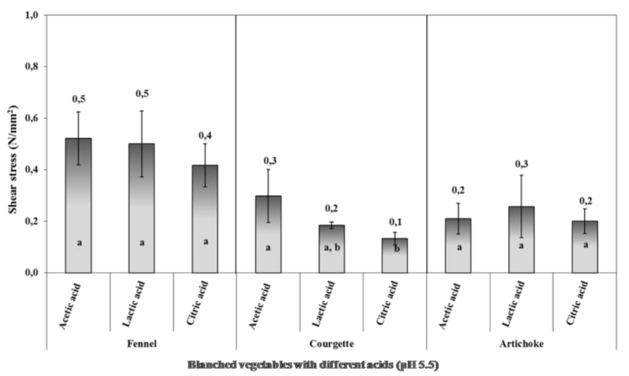

Figure 1: Shear stress of blanched vegetables acidified with different acids.

Figure 2 shows the values of shear stress of the different formula of vegetable *Salami*. The formulation containing xanthan gum and wheat flour has shown shear stress values very similar to meat *Salami*. Those data are confirmed by hardness values that show how the formula containing xanthan gum and wheat flour shows mechanical properties similar to meat *Salami* (Figure 3 and 4) except for springiness that did not show any significant difference (data not shown).

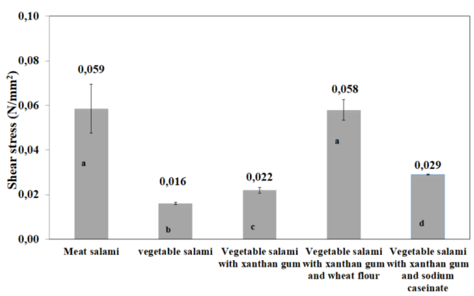

Figure 2: Shear stress values (N/mm^2) of different formulas of vegetable *Salami*.

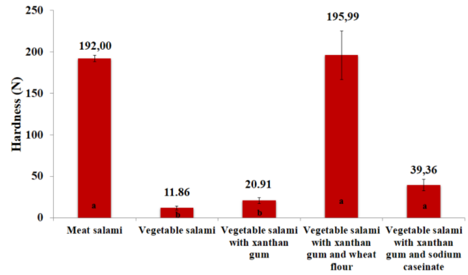

Figure 3: Hardness values (N) of different formulas of vegetable *Salami*.

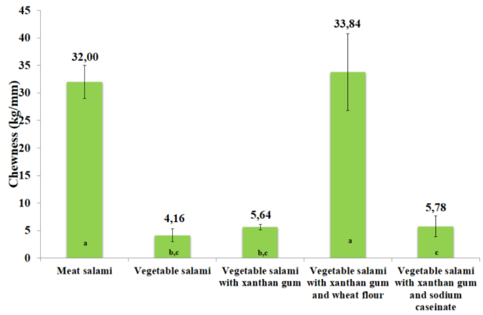

Figure 4: Chewiness values (kg/mm) of different formulas of vegetable *Salami*.

That formula was chosen to inoculate the selected microbial starter that allowed conducing the fermentation in vegetable matrix up to a pH of safety. The obtained vegetable *Salami* were ripened for eighteen days at 25°C. Again, the pH, aw, L*, a*, b*, shear stress and mechanical properties of vegetable *Salami* were monitored during ripening stages.

Figure 5 shows the pH variation as a function of ripening time. After four days of ripening, the pH reaches a value below 4.5 that could be considered safe.

The chromatic characteristics of vegetable *Salami* during ripening have shown a reduction of L* (from 50.27 to 39.02) and b* indexes (from 44.92 to 30.59), while the a* index did not show any significant change. That behaviour could be mainly attributed to the gradual dehydration of vegetable matrix rather than oxidation of pigments present in red fraction.

Concerning the a_w, a gradual reduction was observed up to 15 days of ripening, after that the aw values remain constant at about 0.9 (Figure 6).

Regarding the mechanical properties of vegetable *Salami*, a sudden increase of shear stress force was recorded as a function of maturation up to eighteenth day of ripening (Figure 7). In addition, the hardness, the chewiness and springiness of the slices subjected to compression analysis showed the same trend (Figures 8-10). Those data permitted

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to assert that it would be appropriate to extend the ripening of this kind of products up to eighteen days.

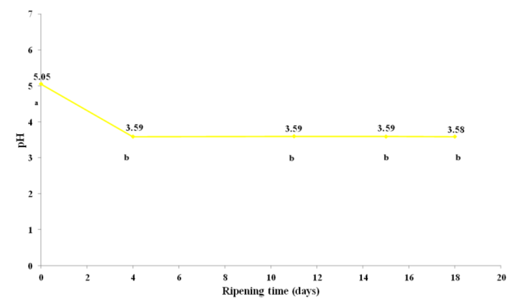

Figure 5: Trend of pH values as a function of ripening time.

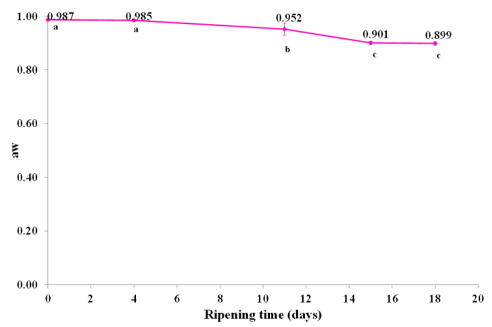

Figure 6: Trend of aw values as a function of ripening time.

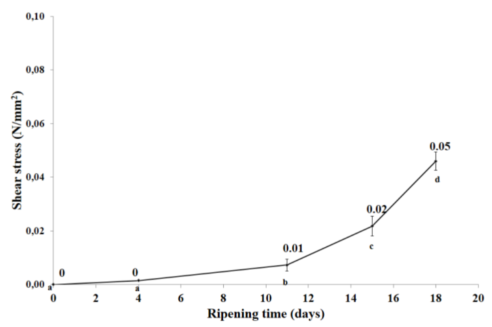

Figure 7: Trend of shear stress (N/mm) values as a function of ripening time

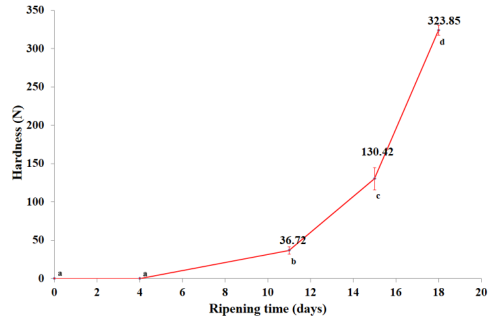

Figure 8: Trend of hardness (N) values as a function of ripening time

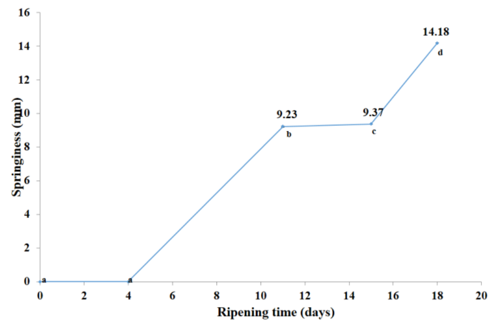

Figure 9: Trend of chewiness value (Kg/mm) as a function of ripening time

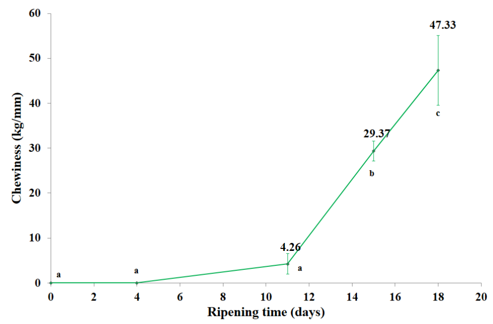

Figure 10: Trend of springiness (mm) values as a function of ripening time

The concentration of lactic acid bacteria at the end of ripening time was 10^8 CFU/g vs 10^{13} CFU/g initial inoculum. This value represents the minimum threshold to consider a food as probiotic *in vitro.*

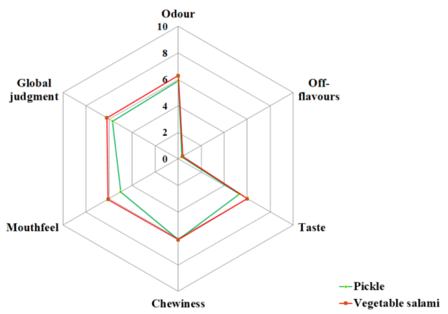

Figure 11: Sensory characteristics of vegetable *Salami* at the end of ripening compared with a commercial pickle

Figure 11 shows the judgments emerged from the comparison of vegetable *Salami* with a common pickle. Samples of vegetable *Salami* showed the highest score for all the considered characteristics. Finally, Figure 12 showed a sample of vegetable *Salami* at the end of ripening stage.

Figure 12: picture of a vegetable *Salami* at the end of ripening

Conclusion

Results of this research have allowed developing an innovative formula that is able to simulate the meat *Salami* in terms of colour and consistency. Moreover, the optimization of processing for this type of *Salami* should provide a ripening of 18 days at 25°C and 75% RU in early days and 45 % RU in the final stage of ripening in order to obtain a product with high sensorial characteristics.

Concerning the functional properties, we can assert that vegetable *Salami* could considered as probiotic food *in vitro* since the lactic bacteria concentration was more than 10^8 CFU/g at the end of ripening time.

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