

Substitution of nitrites in a meat stuffed product with turnip (*Brassica rapa*) and partial replacement of potato flour (*Solanum tuberosum*) for mango peel flour (*Mangifera indica*) for the evaluation of the development of color and texture

Sustitución de nitritos en un producto cárnico embutido por nabo (*Brassica rapa*) y sustitución parcial de harina de papa (*Solanum tuberosum*) por harina de cáscara de mango (*Mangifera indica*) para la evaluación del desarrollo de color y textura

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Resumen

El color en alimentos cárnicos es el producto de reacciones bioquímicas entre los compuestos naturales de la carne, tales como la mioglobina, la hemoglobina y el oxígeno y la acción de agentes externos, tales como los nitratos y nitritos. Recientemente, se ha descubierto que determinados vegetales pueden ser fuentes de nitratos para la elaboración de productos cárnicos, capaces de almacenarlos en sus diversas estructuras. El objetivo de este trabajo, fue evaluar el desarrollo del color característico de las salchichas, mediante la sustitución de nitritos sintéticos por una fuente natural como el nabo, además, realizar una sustitución parcial de harina de papa por harina de cáscara de mango, para conocer su influencia en la textura final del producto. Para esto se elaboraron 3 muestras (patrón con nitrato, extracto de nabo y nabo deshidratado). La cantidad de nitritos presente se evaluó por espectrofotometría, según el método 973.31 de la *Association of Official Analytical Chemists* (AOAC), los parámetros de colorimetría se determinaron por el método CIELAB y la textura se evaluó con el método Análisis de Perfil de Textura (TPA). Se encontró que la cantidad de nitritos es inferior al patrón y a lo reportado por otras literaturas (patrón: 84,87 ppm, extracto: 9,80 ppm y deshidratado: 11,81 ppm), en cuanto a los análisis de textura y colorimetría se identificaron diferencias significativas entre el patrón y las muestras con adición de nabo. Para concluir, se debe aumentar la proporción de nabo en la adición para lograr la concentración deseada de nitritos en el producto final, la adición de harina de cáscara de mango influye en la textura final del producto. Finalmente, se recomienda realizar evaluación del contenido de nitritos en el nabo durante un

tiempo de almacenamiento determinado y aumentar la proporción de nabo en la salchicha, evaluando que las características sensoriales no se vean afectadas.

Palabras clave: nitritos; nabo; salchicha; colorimetría; textura.

Abstract

The color in meat foods is the product of biochemical reactions between the natural compounds of the meat, such as myoglobin, hemoglobin and oxygen and the action of external agents such as nitrates and nitrites. Recently it has been discovered that certain vegetables can be sources of nitrates for the production of meat products, capable of storing them in their different structures. The objective of this work was to evaluate the development of the characteristic color of the sausages substituting synthetic nitrites with a natural source like the turnip, also, to make a partial substitution of potato flour with mango peel flour to know its influence on the final texture of the product. For this, 3 samples were elaborated (Pattern with Nitral, turnip extract, and dehydrated turnip). The number of nitrites present was evaluated by spectrophotometry according to method 973.31 of the AOAC (Association of Official Analytical Chemists), the colorimetry parameters were determined by the CIELAB method and the texture was evaluated with the Texture Profile Analysis (TPA) method. It was found that the amount of nitrite is lower to the standard and the reports on other literature (pattern: 84.87 ppm, extract: 9.80 ppm and dehydrated: 11.81 ppm), in terms of texture and colorimetry analysis. They identified significant differences between the standard and the samples with the turnip addition. To conclude, the proportion of turnip in the addition must be increased to achieve the desired concentration of nitrites in the final product, the addition of mango peel flour influences the final texture of the product. Finally, it is recommended to evaluate the content of nitrites in the turnip during certain storage time and increase the proportion of turnip in the sausage, evaluating that the sensory characteristics are not affected.

Keywords: nitrites; turnip; sausage; colorimetry; texture.

Introduction

Sausages are stuffed meat made from meat and fat of beef, horses, pigs or poultry, mixture properly crushed, grounded and mixed, to which you can add food additives allowed, the mixture is embedded in an artificial membrane, cooked and eventually smoked (Huanca; Solís, 2010). Regarding the color in meat products, it is generated by biochemical reactions between natural meat compounds, such as myoglobin, hemoglobin and oxygen and the action of external agents, such as nitrates and nitrites (Pinzon; Hleap; Ordoñez Santos, 2015).

Currently, it has been discovered that some vegetables can be sources of nitrates for the production of meat products, capable of storing nitrate concentrations in their different structures (Gallego, 2013), including celery, lettuce, and beet that have concentrations of between 1500 and 2800 ppm of nitrates (Montiel-Flores; López-Malo; Bárcenas-Pozos, 2013), in addition to chard, cucumber, and turnip (Hernández, 2008).

Various investigations seek to generate meat products by replacing nitrites of synthetic origin with sources of plant origin. Ruiz-Capillas *et al.*, (2014) performed the replacement of nitrites with a formulation containing celery, natural dye and vitamins C and E, finding that the reformulated sausages have similar conditions to products made with commercial nitrites.

The turnip (*Brassica rapa*) Norfolk pink or violet neck, has a sour or acid taste, sweet, earthy or metallic; It generates a pungent and astringent sensation and its texture is fibrous and firm (Helland *et al.*, 2015). This is in an intermediate range of nitrite composition, being 500 to 1000 mg/Kg of nitrites (Moreno; Soto; González, 2015), however, in a study to perform the concentration, about 4800 mg/kg of nitrates and nitrites were found. Therefore, it can be implied that the turnip is one of the plant products that contain the highest concentration of nitrites (Vargas; Taborda, 2006).

On the other hand, the fiber content of the Creole mango peel ranges between 47.68 % and 59.44 % (Torres-González; Jiménez-Munguía; Bárcenas-Pozos, 2014), which is consistent with what was reported by Sánchez (2005), who establishes that the fiber content of this shell is 56.68 %, and 29.46 % corresponds to soluble dietary fiber. The Water Retention Capacity (WRC) is related to the composition of the dietary fiber, since the greater the amount of soluble dietary fiber, this property increases. Creole mango peel has a WRC of approximately 4.07 g of water/g fiber, in addition to good antioxidant activity, due to its polyphenol and carotenoid content (Cañas; Restrepo; Cortés, 2011).

The main objective of this research was to evaluate the development of the characteristic color of the products processed in a sausage, by replacing synthetic nitrites with a natural source such as turnip. Complementary to this it was intended to make a partial replacement of potato flour with mango peel flour, to know its influence, in the final texture of the product.

Materials and methods

Obtaining raw materials

The meat used to make the product was beef, pork (brazuelo), chicken, mechanically deboned (MDC) and bacon. These were obtained from a local market in the municipality of La Ceja, Antioquia. The condiments and additives used for processing, such as Regal base, accord, promax, potato flour, liquid smoke, punzo red, and oregano, were supplied by the University of Antioquia.

Preparation of the nitrite substituent

Turnip extract: the plant material source of the nitrites is the turnip variety *norfolk* pink or violet neck, came from a farm located in the Quirama village of the municipality of Carmen de Viboral, Antioquia, where the method of cultivation of turnips is organic and the type of Production is for local supply, not for commercialization. The extract was made through the slicing and sieving of the turnip, from which the juice was obtained. Due to the hardness of the vegetable, the procedure was carried out adding hot water (approximate temperatures of 60-65 °C) and a subsequent screening to eliminate the fiber that was not required for the process. Immediately, the extract was concentrated by heating and evaporation of the water added at the time of the extraction, for this, temperatures of 90-95 °C were used until the concentrated extract was obtained.

Dehydrated turnip: the turnip was washed and disinfected, subsequently cut into sheets for dehydration, which was done in a UNOX® convection oven, reference *Rossella* at a temperature of 90 °C for 90 min. After the dehydration process, the particle size was reduced in a NUTRIBULLET brand food processor, Magic reference. The flour was stored in a polythene bag until it was used.

Mango Peel Flour

Mango peels were obtained (purchased from a local market in the municipality of La Ceja del Tambo, Antioquia), which were washed and disinfected. They were subsequently dehydrated at 80 °C for 120 min in a UNOX® convection oven, reference *Rossella*. After the dehydration time was over, the particle size was reduced in a NUTRIBULLET brand food processor, Magic reference. The flour was stored in a polythene bag until it was used.

Preparation of sausage type stuffed meat

To begin, the reception of the raw materials was done taking into account the physicochemical analyzes, mainly the pH and temperature of these. In addition, the organoleptic characteristics of each one were taken into account; also, the weight of the raw material was verified with their respective removal of unwanted parts (veins, large portions of fat, among others). Subsequently, the weighing of the ingredients took place according to what was established in the product formulator.

Next, the grinding process of the meat and fat raw material took place, where a 4.5 mm diameter disc was used. Special care was taken to maintain a temperature below or equal to 12 °C, and so that the fat raw material was not mixed with the meat to not affect subsequent processes. Next, the cutting process was performed, adding the ingredients in order, the temperature (below 10 °C) and the processing time (maximum 8 min). Once this process was finished, the product was still stuffed in a 22-gauge cellulose tubular, using a manual stuffer with a maximum capacity of 1 kg/pothole. Subsequently, the portioning of the sausage with an approximately equal length between them was performed. The heat treatment was carried out in two sections. The first, in dry cooking using a smoker oven until the internal temperature of the product, was maintained between 50-55 °C, ensuring a color stabilization. After this process, the second section of the heat treatment was passed with a wet hot water firing, which did not exceed a temperature of 80 °C, ensuring that the product reached an internal temperature of 72 °C so that the complete cooking of the same. To finish the heat treatment, the product was brought to a temperature between 0 and 4 °C, preventing it from coming into contact with the cooling water to ensure its safety. Finally, the product was vacuum packed, ensuring a shelf life of 30 days without being opened.

As for the formulation of sausages made with turnip, the following percentages were used: 3 % turnip extract, 2 % dehydrated turnip, and for the standard 0.1 % nitrating (nitral) salt was used.

Nitrite Determination

For the three treatments developed, the nitrite content was determined, according to the *Association of Official Analytical Chemists (AOAC)* method 973.31. 10 to 12 g of the finely crushed sample was weighed on a Sartorius CP2245 analytical scale, depositing it in a 100 mL *beaker*. 40 mL of deionized water was added without exceeding a temperature of 80 °C. The sample was transferred to a 500 mL flask. The *beaker* containing the sample was washed with successive portions of hot water and the washings were added to the 500 mL flask; subsequently, hot distilled water was added until approximately 300 mL was adjusted, the samples were taken to a steam bath for 2 h with occasional stirring. Then they cooled to room temperature. In addition, it was packed with deionized water, filtered and an aliquot of the 25 mL filtrate was taken. Then, with a volumetric pipette, a 50 mL flask was taken and 2.5 mL of sulfanilamide ($C_6H_8N_2O_2S$) was added with the burette and the ingredients were mixed. Then, it was allowed to stand for 5 min, after the time 2.5 mL of ferrochrome T black ($C_{20}H_{12}N_3O_7SNa$) with burette was added, mixed and capped with deionized water. It was then allowed to rest for 15 min and the absorbance was read on a Hewlett Packard brand spectrophotometer, *Agilent* reference series 8453 UV-VIS at a wavelength of 540 nm.

Texture Profile Analysis

It was developed in a texture meter brand StableMicro Systems mod. TAXT 2i with an initial force load cell of 30 kg and was performed with a speed before the test of 1 mm/s. During the test, the speed of 2 mm/s was used and after the test speed of 5 mm/s. The time for the performance of each test was 5 s with a compression percentage of 40 %. The parameters analyzed were hardness, chewiness, and rubberiness. These analyses were performed on the three samples prepared standard, extract and dehydrated.

CIELAB method colorimetry analysis

This test was performed on a DIGI-EYE device manufactured by VERIVIDE, using a D-65 illuminant. These analyses were performed on the three standard samples (sausage made with nitral), extract and dehydrated (replacing the nitral with turnip) both on the surface of the sausage and on the map (internal part) of it.

Data analysis

The data obtained in the different analyzes were recorded and evaluated using the Microsoft Office Excel 2016 software.

Results and Discussion

Determination of nitrites in sausages

According to the method used, the results obtained for the nitrite content present in the three sausage samples evaluated are listed in Table 1:

Table 1.

Average nitrate content in the three sausage samples

Sample	Concentration (ppm)
Pattern	84,87
Extract	9,80
Dehydrated	11,81

Source: self-made.

According to Resolution 4125 of 1991 of the Ministry of Health and Social Protection of Colombia, the maximum allowable limit of nitrites in food is 200 ppm. The values obtained were lower than this reference since the highest concentration obtained (84.87 ppm) was in the standard sample, which was prepared from the addition of nitrites of synthetic origin (nitral curative salt). The other samples, which were made from a natural source of nitrates, *B. Rapa*, presented values below the standard with the amount added reported in other research Vargas and Taborda (2006). This may be mainly due to the fact that nitrates are originally found in the natural source (*B. Rapa*), which, due to the bacterial activity of the meat, is degraded to nitrites. The latter are the ones that exert the action of color development in the product and, for this reason, if during the process there is not a long time for the transformation of nitrates to nitrites, there will be no adequate color development.

According to the theoretical formulation of the product, the results obtained experimentally from the standard are lower, because the additive (Nitral) had a prolonged storage time, which means that the reaction between nitral and meat does not occur in an expected way.

Texture analysis

The parameters evaluated in the texture profile analysis (TPA) are presented in Table 2 and are plotted in Figure 1.

Table 2.

Texture analysis data (TPA) in the three sausage samples

Sample	Hardness (g)	Rubberiness (g)	Chewiness (g)
Pattern	4254,3458±453,08	3177,2846±289,86	2966,664±265,89
Extract	3968,7652±302,67	2813,2356±222,03	2551,369±176,42
Dehydrated	3797,4338±386,07	2211,8170±185,72	2554,433±164,24

Source: self-made.

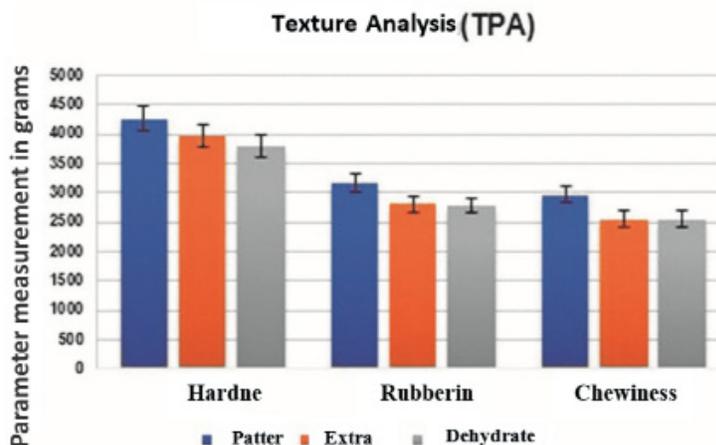


Figure 1. Texture parameters evaluated (Hardness, rubberiness, and chewiness)

Source: self-made.

Using Figure 1, it was possible to determine that there are significant differences between the analyzed samples and the standard (using 95 % confidence), however, between the sausage samples with turnip extract and dried turnip, there is no significant difference. This is due to the fact that in both samples a natural source was used to obtain the nitrites and for the standard sample a synthetic source was used, which has greater activity and stability in the medium.

By comparing the hardness of the standard sausage with the other two samples evaluated, it can be identified that it has a greater hardness ($4254.35 \text{ kg m s}^{-2}$), which indicates that it is required to perform a greater force of chewing the product to obtain a deformation similar to that which would be obtained in the sausages to which the substitution was made. These reported values of $3968.77 \text{ kg m s}^{-2}$ for the sausage with the extract and $3797.43 \text{ kg m s}^{-2}$ for the sausage with the dried turnip. This difference in hardness is mainly due to the addition of mango peel flour in sausages with turnip extract and dried turnip, which has a greater water retention capacity, compared to the potato flour that was used for preparation of the standard sausage, which generates an increase in the moisture content in the final product, resulting in a decrease in the hardness of the sausage.

Gumsiness is defined as the multiplication of hardness and cohesiveness. In Figure 1 it can be shown that this behaved in a similar way to hardness since it showed a decrease in the rubberiness of the samples with the replacement of nitrites ($2813.24 \text{ kg m s}^{-2}$ for sausage with extract and $2211.82 \text{ kg m s}^{-2}$ for dehydrated sausage) in relation to the pattern used ($3177.29 \text{ kg m s}^{-2}$), this difference being significant for both cases. However, there is no significant difference between the studied samples. The rubberiness is directly related to the state of the meat (Arnau, 2011), when it is too lean, the final product has a hardness and a higher rubberiness, than when you have a product with higher fat content, favoring the product emulsion, resulting in a lower hardness and rubberiness.

The chewiness of a processed meat product analyzed by the TPA method is defined as the multiplication of hardness, cohesiveness, and elasticity, this relationship being directly proportional. Therefore, an identical behavior to the aforementioned cases was obtained, since if the hardness decreases, as is the case of the two samples analyzed, there will be a decrease in the chewiness of the product. The chewiness is a variable that confirms the final behavior of the product since this is the combination of different factors thrown by the TPA texture analysis.

Color analysis by CIELAB

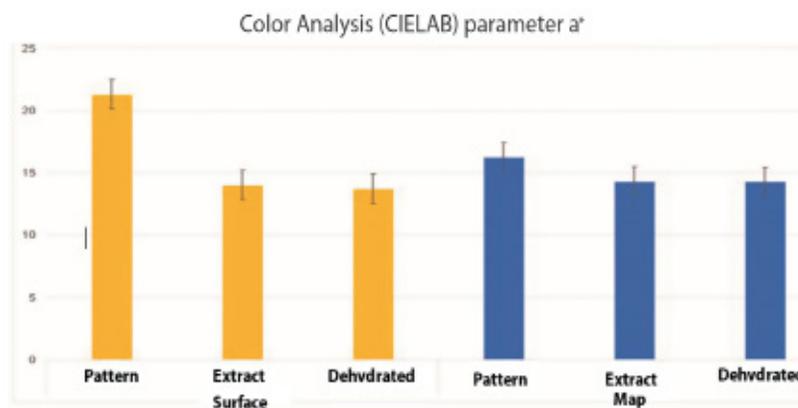
The color analysis for the three samples was carried out by the CIELAB method, in which the coordinates a* and b* (from green to red and from blue to yellow, respectively), brightness (L*), chroma (C) and tone (h). The latter obtained by direct reading from the colorimeter. According to these aspects evaluated, the significant difference that may exist between treatments can be determined. Table 3 shows the data obtained and Figure 2 shows the factors evaluated.

Table 3.

Average color analysis data by CIELAB method for the three samples made inside (map) and on the outside (surface)

SURFACE					
Treatment	L	a	b	C	h
Pattern	56,49±0,48	21,30±0,27	17,96±0,26	27,87±0,15	40,14±0,71
Extract	56,32±0,35	14,03±0,33	17,90±0,19	22,74±0,22	51,93±0,79
Dehydrated	56,82±0,14	13,72±0,46	18,18±0,24	22,78±0,27	52,97±1,09
MAP					
Treatment	L	A	B	C	H
Pattern	64,31±0,27	16,24±0,29	14,41±0,09	21,71±0,24	41,59±0,45
Extract	61,86±0,36	14,33±0,20	15,27±0,18	20,94±0,25	46,84±0,22
Dehydrated	61,53±0,23	14,28±0,30	15,39±0,15	21,00±0,14	47,14±0,84

Source: self-made.



a)

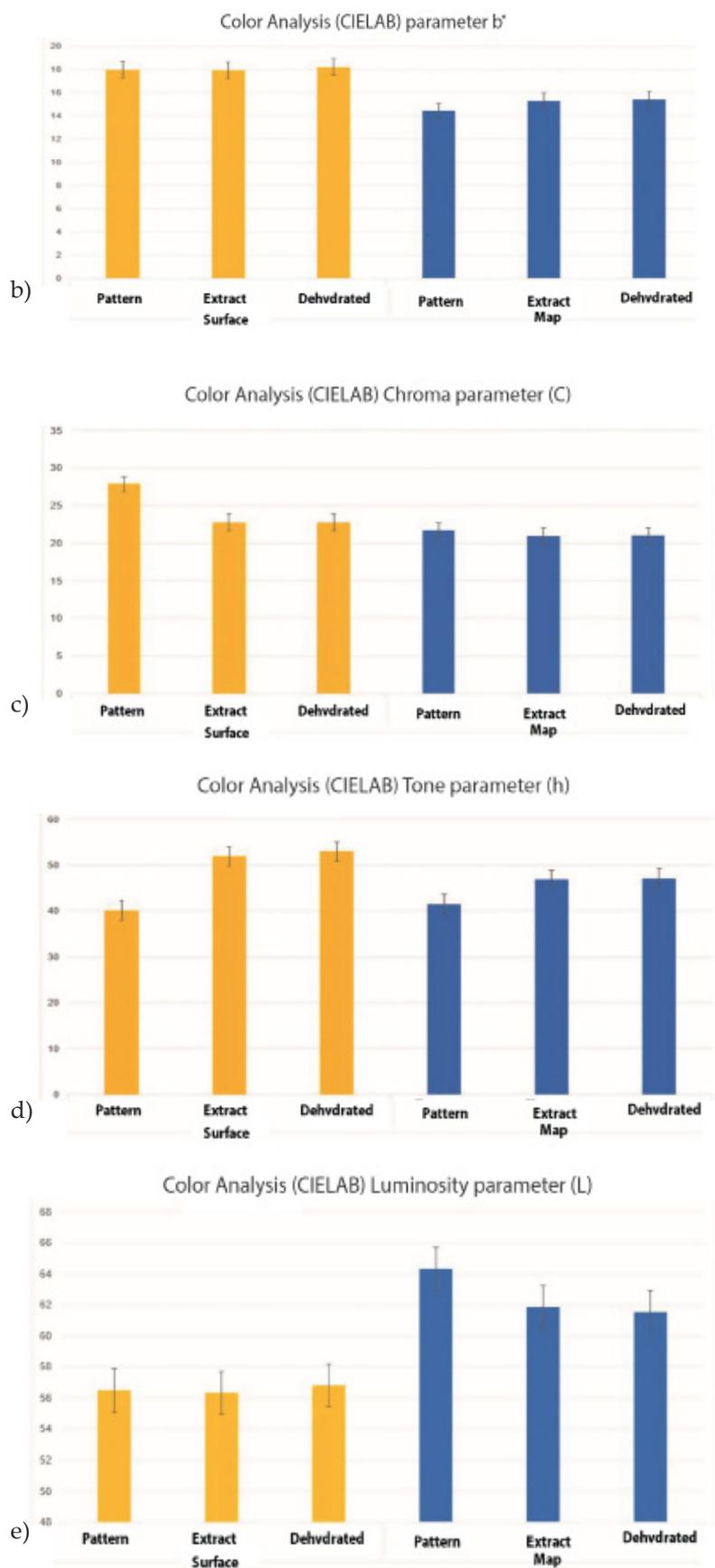


Figure 2. Analysis of color in the sausage samples for both the surface and the map
Source: self-made.

The a^* parameter (Figure 2a), which includes the area between red and green, showed significant differences between the samples evaluated and the pattern for both the surface and the map. This parameter is important in meat products since they are mostly red or similar. In the case of the tests carried out, the difference between the standard and the samples has a direct relationship with the nitrate content, which was initially determined, since these are the ones that contribute the development of the color to the processed meat products. Although the difference in nitrate concentration between the samples with turnip addition and the standard was wide (70 ppm of difference) visually, when performing the colorimetric analysis it is not evidenced with the same intensity, although on the surface there is a greater variation than on the map since the development of the second skin appears on the surface of the sausages, which implies a greater intensity of color on the surface than on the map.

According to a study by Amensour *et al.*, (2010), they explain that in meat products the coordinate a^* is influenced by various technological factors (hot or cold emulsion, type of chopped, among others.) Moreover, composition (ratio lean fraction/fat fraction, among others). On the other hand, the luminosity is related to the free water on the surface, the fat content, the connective tissue, the content and the type of heme pigments, among others.

Regarding the decrease in the coordinate a^* as the nitrite content was reduced when substituting for turnip extract or dehydrated turnip, it can be explained by the reduction of nitrite to nitric oxide (NO) and its consequent reaction with myoglobin to form nitrosomyoglobin that is stabilized by treatment because the less nitrites, the less nitrosomyoglobin-forming reactions. These results coincide with those obtained by Deda; Bloukas and Fista (2007), who worked with meat and pork sausages added with sodium nitrite and tomato paste.

Regarding the evaluation of the color parameter b^* , this followed a behavior similar to that studied in parameter a^* , presenting a greater variation between the pattern and the samples, which may be related to the process of formation of the second skin in the sausage, in which there is a greater development of the yellow color, due to the contribution of this color by both the turnip dehydrated and in extract. This effect is not evidenced in the same way as with the addition of nitral (Deda *et al.*, 2007).

On the other hand, in the colorimetric evaluation of the sausage samples, it was observed that the luminosity value (L^*) does not represent significant differences, that is, taking into account $\alpha < 0.05$; this happens for both the surface and the map.

The chroma coordinate (C) represents the degree of color saturation in the samples. In the case of the surface, variation was presented despite not being significant; however, this difference does not exist on the map, which may be related to the reasons stated above for the other coordinates.

Finally, in the coordinate h, where the information about the sausage tone is reflected, an inverse behavior was presented to the others, since the samples with turnip addition reflect values superior to the pattern, this being a significant difference for both surface as for the map.

The differences in color between sausages from various studies are also related to the color characteristics of the main ingredients used in their preparation (Isaza; Restrepo; López, 2012).

Conclusions

From the results obtained, it can be concluded that, according to the nitrite analysis, the concentration of turnip both dehydrated and in extract must be in a greater proportion to that used in the current study if obtaining a greater development of color is desired.

The partial substitution of potato flour with mango peel flour decreases the hardness, rubberiness, and chewiness in the sausage because the latter presents a higher CRA.

With the concentration of natural source nitrites used, it was possible to obtain a product with colors of red hue, characteristic of meat products.

Greetings

Professor Nelly Ospina de Barreneche, a tenured professor at the University of Antioquia, Mr. Hernán Darío Villa Montoya, of Industrias Zenú S.A. To Mr. Edwin Arcila, technician of the mobile plants of the University of Antioquia. To Professor Rosario Echeverri, coordinator of the Laboratory of Physicochemical Analysis of Food at the University of Antioquia, and Mr. Andrés Román, coordinator of Laboratories University of Antioquia Sectional Oriente.

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