DEVELOPMENT OF LOW FAT – LOW SALT PROCESSED MEAT PRODUCTS RAZVOJ MESNIH PRERAĐEVINA SA NISKIM SADRŽAJEM MASTI I SOLI

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ABSTRACT

There is a growing demand for the development of healthier meat products with reduced fat and salts. The present study was carried out to develop low fat – low salt processed meat products in a type of fermented sausages. Products were formulated with different fat contents (10 g/100g - 20 g/100g) and levels of sodium chloride (0-2 g/100g) and potassium chloride (0-1 g/100g). Potassium chloride (KCl) was used as a substitute for sodium chloride (NaCl). Physicochemical characteristics and textural attributes were assessed instrumentally, while sensory attributes were determined using Quantitative Descriptive Analysis and Principal Component Analysis. The instrumental analysis showed significant ($p < 0.05$) differences in hardness, brittleness, cohesiveness and chewiness among different sausage formulations. However, sensory analysis did not detect any textural changes due to salt substitution or fat reduction. The product with the highest fat content and KCl substitution was found to have the highest rating of perceived bitterness. The research findings identified the significant interaction between the addition of KCl and the fat content on bitterness perception.

Keywords: NaCl substitution, fat reduction, Quantitative Descriptive Analysis, Principal Component Analysis, texture analysis.

REZIME

Sve je veća potražnja za razvojem zdravijih mesnih proizvoda sa manjim udelom masnoće i soli. Ova studija sprovedena je za razvoj mesnih prerađenih proizvoda sa niskim sadržajem masti i soli u okviru fermentisanih kobasica. Proizvodi su formulisani sa različitim sadržajem masti (10 g/100 g - 20 g/100 g) i nivoima natrijum hlorida (0-2 g/100 g) i kalijum hlorida (0-1 g/100 g). Kalijum hlorid (KCl) je korišćen kao zamena za natrijum hlorid (NaCl). Fizičko-hemijske karakteristike i teksturni atributi procenjeni su instrumentalno, dok su senzorni atributi određeni pomoću kvantitativne deskriptivne analize i analize glavnih komponenti. Instrumentalna analiza pokazala je značajne ($p < 0,05$) razlike u tvrdoći, krhkosti, kohezivnosti i žvackljivosti među različitim formulacijama kobasica. Međutim, senzorna analiza nisu detektovane nikakve teksturne promene usled zamene soli ili smanjenja masti. Utvrđeno je da proizvod sa najvećim sadržajem masti i supstitucijom KCl ima najvišu ocenu osetljive gorčine. Nalazi istraživanja identifikovali su značajnu interakciju između dodavanja KCl i sadržaja masti u percepciji gorčine.

Ključne reči: Zamena NaCl, redukcija masti, kvantitativna deskriptivna analiza, analiza glavnih komponenti, analiza teksture.

INTRODUCTION

Among processed meat products, sausages are formulated with different recipes depending on locality. The main ingredients are pork and beef meat, pork backfat and sodium chloride (Georgakis, 2005). Minor ingredients may also include various herbs and spices, e.g. leek, onion, orange peel, etc. According to Ambrosiadis et al. (2004), the fat content of sausages commercially available was found to be approximately 29%, contributing significantly to the caloric content of these products. Moreover, pork backfat consists of 32% saturated fatty acids and 57 mg cholesterol per 100 grams (USDA, 2016). Saturated fatty acids and cholesterol are the main risk factors for the development of cardiovascular diseases (European Food Safety Authority, 2010). Also, dietary intake of sodium chloride, which is typically added at 2% of the meat formulation, has a positive correlation with high blood pressure and hypertension, according to several studies (Doyle & Glass, 2010; Gelabert et al. 2003; Horita et al. 2014; Ruusunen et al. 2005). However, the reduction of fat and sodium in meat products could raise important issues related to manufacturing hurdles and the quality of the final product as analyzed below.

Fat affects the dehydration and ageing processes during sausage production by allowing gradual dehydration, limiting the

weight losses and preventing case hardening phenomena on these products (Gallego et al. 2014; Muguerza et al. 2002, Olivares et al. 2010; Ventanas et al. 2010; Yim et al. 2016). More specifically, fat, due to its chemical structure, acts also as a solvent - carrier of volatile aromatic lipophilic compounds, through lipolysis and lipid oxidation (Olivares et al. 2011; Zanardi et al., 2004). Moreover, fat is a major determinant of the textural attributes of fermented meat products by increasing juiciness, lubricity and tenderness, as measured both by instrumental and sensory analysis (Gallego et al. 2014; Muguerza et al. 2002). Muguerza et al. (2002) have suggested that fat reduction up to 15-20%, with respect to control formulation, can be achieved without major odor and taste changes to the final products.

NaCl substitution by other salts or ingredients has been proposed for a variety of meat products (Corral et al. 2016; Gimeno et al. 2001; Gelabert et al. 2003). However, NaCl substitution can lead to several physicochemical and sensorial changes of the final product (Doyle & Glass, 2010), because of its multiple functionalities. Sausages containing NaCl exhibit higher ionic strength compared to substitutes like KCl (Gimeno et al. 2001), which enhances the extraction and solubilization of the actino-myosin protein complex – a function that contributes to the development of desirable textural properties (Corral et al. 2016; Puolanne & Petronen, 2013). Salt also enhances flavor

intensity and taste perception by consumers, through lipid oxidation (Love & Pearson, 1974; Osinchak et al., 1992; Quintanilla et al., 1996; Ventanas et al. 2010). Thereby, it could be assumed that the ionic strength of the added salt used can have a direct effect on a product's flavor and taste. However, the substitution of NaCl by KCl results in the development of bitter and metallic taste notes in meat products (Perisic et al. 2013), when the substitution was conducted above 40% (Horita et al., 2011).

Although fat reduction and NaCl replacement by KCl in processed meat products have been investigated by different scientists over time (Jin et al. 2018; Muguerza et al. 2002), there is little information in the literature regarding the interaction between fat reduction and NaCl substitution by KCl and its impact on physicochemical, textural and sensorial characteristics of processed meat products. The objective of this study was to investigate the combinatory effect of the use of KCl as a NaCl substitute and fat reduction on the textural and sensorial attributes of processed meat products.

MATERIALS AND METHODS

Materials

Fresh boneless lean pork cuts from ham, beef cuts from the lean shoulder and fresh pork backfat were obtained from the local market (Thessaloniki, Greece) and were transferred to the laboratory within 30 min. Pork and beef cuts (in a 1:1 ratio) were trimmed of adhering tissues and visible connective tissue and they were minced with the backfat through a 12 mm grinding plate. All meat and pork backfat were obtained on the day of sausage production. Fresh leek was chopped into 2-5 mm pieces beforehand. All herbs and spices were in dry form.

Methods

Processed meat product formulation

A typical recipe from Northern Greece was used for the formulation of the sausages. The products were formulated with different fat content (added backfat at 10% - 20%) and also a different level of NaCl substitution with KCl (0% - 50%), based on a 2X2 full factorial design. In Table 1, the different formulations prepared are presented. The sample formulations are denoted as RFN for the regular fat formula with NaCl, RFK for the regular fat formula with KCl, LFN for the low fat with NaCl and LFK for the low-fat formula with KCl.

The sausages were produced as follows: immediately after grinding and mixing of meat and backfat, the salt and other ingredients were added into the meat mixture and were kneaded thoroughly. The mixtures were stuffed into natural casings from pork small intestines. The raw products were placed in a temperature and humidity-controlled room (20 °C, 75-60% RH) for seven days to allow for partial dehydration and fermentation of the sausages to take place. This is the typical duration of fermentation – dehydration in the artisanal production of processed meat products like sausages. After this period, the sausages are usually consumed within a few days, since they are not heat-processed, they are not fully fermented, and they contain no added nitrites/nitrates. After the seven-day fermentation, the sausages were vacuum packaged (to avoid further dehydration of the samples) and were stored in a refrigerator at 4 °C.

Two batches of sausages were prepared, on different days, using different meat and other raw materials, in order to have two complete replicates. All analyses were performed in triplicate within three days after the completion of the fermentation process.

Table 1. Sausages formulations

Ingredients (g/100g sausage)	RFN*	RFK	LFN	LFK
Pork and beef meat	57.3	57.3	67.3	67.3
Pork backfat	20	20	10	10
Sodium chloride	2	1	2	1
Potassium chloride	0	1	0	1
Fresh leek	20	20	20	20
Cumin	0.15	0.15	0.15	0.15
Oregano	0.4	0.4	0.4	0.4
White pepper	0.15	0.15	0.15	0.15

*RFN: regular fat, NaCl formula; RFK: regular fat, KCl formula; LFN: low fat, NaCl formula; LFK: low fat, KCl formula.

Physicochemical Analysis

The weight loss of the fresh sausages during the seven-day fermentation and dehydration period was measured gravimetrically. The pH of the sausages was measured using a Hanna H221 pH-meter (Hanna Instruments BV, IJsselstein, The Netherlands). Moisture content was determined by air drying ~10 g sausage samples at 105°C for 24 h in an air oven equipped with a ventilator (AOAC, 1990). Fat content was measured using the Soxhlet method (AOAC, 1990). Protein content was measured using the Kjeldahl method a nitrogen-to-protein conversion factor of 6.25 was used for the calculation of % protein (AOAC, 1990).

Texture Profile Analysis (TPA)

A universal TA-XT2i texture analyzer (Godalmyng, Surrey, UK) equipped with a 25 kg load cell was used to perform instrumental texture profile analysis (Biliaderis, 1992). The analysis was performed nine days after sausage production. For each treatment, two sausages were selected randomly, cut as cylindrical sections, and grilled in an electric hotplate for 6 minutes to an internal temperature of 75 °C. Two cm sections of cooked sausages were used for the texture profile analysis. Two consecutive compressions with an interval of 5s took place. The cylindrical samples were compressed at 75% of their initial height using a P75 probe (75 mm diameter) moving at a velocity of 1.0 mm s⁻¹. All measurements were conducted at a temperature of 20-22 °C. The data were processed with the Texture Expert, v.1.22 software (Stable Micro Systems Ltd., Godalmyng, UK). Six measurements were taken from each treatment. A force-time curve was plotted for each measurement and the following texture parameters were determined: hardness, brittleness, springiness, chewiness, resilience and cohesiveness.

Sensory Evaluation

Prior to each sensory evaluation session, sausages were cut into 2 cm-thick cylindrical slices and then cooked in a grill pan for 6 min (3 min on each side) to an internal temperature of 75°C. An appropriate cooking time was selected so that samples maintained their shape and juiciness. Post-cooking, all slices were kept in the oven at around 40 °C until tested (maximum 20 min to prevent dehydration).

A semi-trained sensory panel consisting of thirteen assessors participated in the quantitative descriptive analysis (QDA). All the assessors were either staff or students of the Department of Food Science and Technology, Aristotle University of Thessaloniki, and had previous experience in sensory evaluation of related products. Sensory evaluation was conducted in four consecutive sessions; during the first session, the panelists were asked to generate sensory descriptors to characterize the sausage samples. Subsequently, a discussion led by the panel leader followed and the panelists developed a consensus vocabulary in which sensory characteristics of all samples were described and defined. During the following training session on a separate day, panelists decided on the references and the anchors to be used for all the descriptive terms, while this session allowed panelists to get more familiar with the sensory attributes. In the next sessions, panelists individually rated samples in duplicate on two separate days, in individual booths under artificial daylight and at ambient temperature. Samples coded with three-digit random codes were presented at around 40°C and in balanced order to average out any possible sequential effect (bias) in the group data. A non-structured 15-cm scoring scale was used, where 0 was labeled as either “absence” or “very weak” and 15 meant as the highest intensity of the respective descriptor. Water and crackers were given between each sample evaluation.

Statistical Analysis

Physicochemical analysis, texture profile and color analysis data were statistically analyzed with the univariate general linear model of SPSSTM with fat content and salt type and their interaction as fixed effects. For the sensory attributes, a two-way analysis of variance (ANOVA) was applied and Fischer's LSD post hoc test was performed to identify significant differences ($p < 0.05$). Subsequently, the mean sensory ratings were used to carry out a principal component analysis (SENPAQ software; Qi Statistics, Ruscombe, UK).

RESULTS AND DISCUSSION

Physicochemical Characteristics

The progression of the dehydration of the sausages during the 7-day period is presented in Figure 1. The RFN samples (the typical “control” formulation) exhibited lower weight loss compared to the other treatments ($p < 0.05$). No statistically significant differences were measured among the other three treatments ($p > 0.05$). The pH values and the moisture, fat and protein contents of the sausages on the seventh day (end of the fermentation process) are presented in Table 2.

As expected, based on the initial formulation, the low-fat treatments had higher levels of moisture and a lower fat content. Even though the pH values did not vary to a great extent from a practical standpoint (5.46 to 5.63), the differences were statistically significant ($p < 0.05$).

Fig 1. Weight loss of the different formulations of sausages during the 7-day fermentation period. Different superscripts indicate statistical differences ($p < 0.05$);

(RFN: regular fat NaCl, RFK: regular fat KCl, LFN: low fat NaCl, LFK: low fat KCl).

Texture Profile Analysis (TPA)

In Table 3, the results of instrumental texture profile analysis are presented; low-fat products (LFN, LFK) exhibited greater values for hardness, brittleness and chewiness with respect to their regular fat counterparts. Thereby, it can be assumed, that fat content largely affects juiciness and, in this sense, it is an effective tenderizing factor. Similar results have been also reported by other researchers (Muguerza et al., 2002). Moreover, sensorial and textural hardness, chewiness and brittleness were affected not only by fat reduction, but also by salt substitution, and their interaction. Specifically, it was observed, that sensorial and textural hardness, chewiness and brittleness exhibit greater values as fat decreases, especially for the formulations in which NaCl was substituted by KCl. This could be explained by the fact that NaCl substitution by KCl above 40% can bring about lower moisture contents in processed meat products like sausages compared to control samples (Gelabert et al. 2003).

Table 2. Physicochemical properties of the different formulations of raw sausages after a 7-day fermentation period (mean $\pm$ st. deviation).

	RFN*	RFK	LFN	LFK
Moisture (%)	50.69 $\pm$ 0.41 ^b	50.57 $\pm$ 0.21 ^b	53.15 $\pm$ 0.44 ^{aa}	51.63 $\pm$ 0.50 ^{ab}
Protein (%)	17.47 $\pm$ 0.81 ^a	16.59 $\pm$ 0.05 ^a	18.72 $\pm$ 0.49 ^a	17.59 $\pm$ 1.28 ^a
Fat (%)	31.45 $\pm$ 0.40 ^a	31.95 $\pm$ 0.33 ^a	21.19 $\pm$ 2.09 ^b	20.87 $\pm$ 1.95 ^b
pH	5.63 $\pm$ 0.01 ^a	5.46 $\pm$ 0.01 ^c	5.56 $\pm$ 0.02 ^{ab}	5.55 $\pm$ 0.03 ^b

^{a,b,c} Means with different superscripts in the same row, are statistically different ($p < 0.05$)

*RFN: regular fat, NaCl formula; RFK: regular fat, KCl formula; LFN: low fat, NaCl formula; LFK: low fat, KCl formula.

Table 3. Effect of fat content and salt type on textural characteristics of sausages (TPA test).

Sample	Brittleness	Hardness	Cohesiveness	Springiness	Chewiness	Resilience
RFN*	154.55 ^b	174.05 ^b	0.29 ^a	1.08 ^a	54.39 ^b	0.27 ^a
RFK	178.16 ^{ab}	205.75 ^b	0.25 ^b	1.00 ^a	52.66 ^b	0.25 ^a
LFN	194.34 ^a	276.81 ^a	0.23 ^b	1.00 ^a	64.64 ^b	0.25 ^a
LFK	216.11 ^a	327.36 ^a	0.27 ^{ab}	1.00 ^a	90.43 ^a	0.27 ^a

^{abcd} Least-squares means that do not share a letter are significantly different ($P < 0.05$) for each textural attribute.

*RFN: regular fat, NaCl formula; RFK: regular fat, KCl formula; LFN: low fat, NaCl formula; LFK: low fat, KCl formula.

Sensory evaluation

Table 5 lists the sensory descriptors and their definitions as developed by the quantitative descriptive analysis (QDA) panel. The results for the intensity of the sensory attributes are shown in Figure 2; each descriptor bears a prefix denoting the perceived sensation, olfactory (O_), gustative (T_), tactile (TAC_), textural (Tex_) and aftertaste (AT_). The sensory panel evaluated cooked samples and did not detect significant color differences that would lead them to develop sensory vocabulary regarding the appearance of sausage samples. Cooking probably minimized any color differences among samples. Analysis of variance revealed that 4 out of 15 sensory attributes differed significantly among samples. In terms of orthonasal perception, only one attribute, “O_Ripened”, was found to vary significantly with the different product formulations. As it can be seen in Figure 2, the LFN sample was scored with the highest ripeness odor, followed by LFK, while samples containing 20% fat were perceived as having a lower intensity of ripeness. Similar results were also obtained when the samples were retronasally evaluated; sausage samples with low-fat content and irrespective of the salt type used were perceived as more ripened. When comparing samples containing KCl (RFK, LFK) the bitterness intensity was found higher in the case of the samples with regular fat content. Strong correlations between the perceived bitterness and the use of KCl as a NaCl substitute have been reported (Gelabert et al. 2003). Likewise, the sensory panel identified and retronasally perceived bitterness in the sausages, with the sample RFK receiving the highest rating. Furthermore, samples of higher fat content exhibited greater bitterness intensity. This observation is well-established from previous studies with regard to the effect of fat content on sausages (Ruusunen & Puolanne, 2005); i.e. when fat content increases by replacing meat with fat on an equal weight basis, the perceived saltiness of sausages increases. Hence, in the current study, it is believed that a similar effect on the perceived bitterness of sausages is caused by fat content. Specifically, with the increase of fat content and the reduction of water, there is a higher concentration of the salts in the aqueous phase resulting in a saltier and bitter perception of the sample.

In PCA, the two principal components explained 93.7% of the total variance (Figure 3). PC1 accounted for 74.4% of the variance and distinguished the samples according to fat content, placing the low-fat products on the left quadrants and those with regular fat on the right quadrants. PC2 accounted for 19.3% of the variance, incapable of lucid discrimination between samples. Taking into account how the sensory variables are scattered on the plot, it can be pointed out that RFN is negatively correlated with all the sensory descriptors, indicative that sample RFN is perceived as the most balanced between samples (which is expected as this is the typical sausage formulation). The sample LFN was strongly correlated with ripened and fatty sensory properties while the sample RFK stood out from the remaining samples due to its bitterness. When NaCl is replaced with KCl in fermented sausages or dry-cured pork loins, bitterness appears to be the most relevant sensory trait detected. Moreover, the latter observation of the PCA plot further strengthens our hypothesis that the detected differences in the retronasal perception of bitterness by the sensory panel are amplified by a high-fat level in processed meat products like sausages.

It is well recognized that ripening conditions, raw meat type and ingredients have a considerable impact on the sensory quality of sausages as induced by the various physical, biochemical and microbiological transformations that occur during the ripening process (Villani et al. 2007). Similarly, sausages possess very complex sensory attributes due to their product formulation (various raw ingredients) and varying

processing conditions employed. Different production strategies have been adopted to reduce fat and salt content in dry fermented sausages resulting in developing products with different organoleptic properties. For example, it has been found that a 50% fat reduction (from 20% to 10% total fat in the formulation) largely affects the external appearance and flavor intensity of the fermented sausages (Liaros et al. 2009). One robust finding that emerged from the current study is that the effect of fat content possessed a greater influence on the sensory perception of sausages when compared to the effect of salt type. This might be explained by the addition of various spices and herbs (leek, cumin, oregano, pepper), also used in the current product formulation, which behave similarly to salt as flavor enhancers and mask salt reduction. As a result, the perceived saltiness did not vary significantly between samples. Fat is also known to greatly affect the texture, i.e. hardness, mouthfeel perception (Oliveras et al. 2010) and provides lubrication. In general, reduced-fat foods are evaluated by consumers as having inferior sensory properties to regular-fat products. However, Tobin et al. (2012) have shown that fat and salt can be successfully reduced, within limits, in the case of burgers and frankfurters without major sensorial changes. It is worth noting here, that the observed differences in hardness values (in the present study), as determined by instrumental analysis, were not large enough to be perceived by the sensory panel. Thus, even a fat reduction by 50% did not seem to affect the sensory hardness of the sausages as perceived by the sensory panel when comparing reduced fat and normal fat products.

Table 5. Attributes and definitions of the consensus sensory lexicon developed by the assessors

Attributes	Definition
Odor attributes	
Spicy	Characteristic odor generated by the presence of spicy ingredients
Leek	The typical odor of leek
Meaty	The characteristic odor of cooked meat, generated mainly from the presence of glutamine
Ripened	Pleasant odor developed by dry-cured meat products
Taste and flavor	
Saltiness	Basic taste sensation elicited by NaCl
Bitterness	Bitter taste sensation elicited by KCl
Ripened	Pleasant flavor characteristic of dry-fermented sausages
Fattiness	Characteristic lubricity and juicy sensation after fat consumption
Spicy	Taste sensation elicited by spice seasonings
Leek	The characteristic taste sensation of leek
Meaty	A typical sensation when eating cooked minced meat mainly generated from volatile peptides and glutamine
Tactile attributes	
Hardness	Degree of easiness displayed when trying to compress the sausage sample with hands
Texture attributes	
Hardness	The force required to compress a chunk of sausage between the molar teeth to complete breakdown
Aftertaste	
Bitterness	The sensation of bitter taste remaining in the palate after the consumption of sausage
Tongue effect	Characteristic oral sensation after consumption of fermented meat products

Fig 2. Effect of fat reduction and NaCl substitution by KCl on sensory attributes. (RFN: regular fat NaCl, RFK: regular fat KCl, LFN: low fat NaCl, LFK: low fat KCl); different letters above the bars represent significant differences ($p < 0.05$, Fisher's LSD)

Fig 3. Principal Component Analysis (PCA) plot of sensory analysis factors on sausages as affected by fat content and salt type; (RFN: regular fat NaCl, RFK: regular fat KCl, LFN: low fat NaCl, LFK: low fat KCl).

CONCLUSIONS

Sausages made with 50% reduced fat, (~ 33% reduced calories), and NaCl substitution by KCl at a 50% level, can be produced without unacceptable sensory defects. The addition of various spices and herbs used in the developed product formulation behaved similarly to salt as flavor enhancers and masked effectively salt reduction. Overall, the findings of the

present study indicated that any adverse sensorial attributes associated with fat and salt reduction in the fortified products could be alleviated by proper product formulation (e.g. use of seasonings).

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