Sensory properties and fatty acids profiles of fermented dry sausages made of pork meat from various breeds

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Sensory properties and fatty acids profiles of fermented dry sausages made of pork meat from various breeds

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Abstract. In this study, the parallel examinations on fatty acid profiles and sensory properties of fermented dry sausages are presented. Three types of kulen and sremska sausages were made, which varied depending on the percentage of meat and fat derived from different pig breeds: autochthonous (Swallow-Belly Mangalitsa and Moravka) and commercial (Swedish Landrace). In sausages made from meat of commercial pig breed, the highest cholesterol content was detected. However, sausage made from the Mangalitsa pork meat contained higher levels of monounsaturated fatty acid (MUFA) and unsaturated fatty acid (USFA), and lower saturated fatty acid levels (SFA). The level of polyunsaturated (PUFA) in sausage made of Landrace pork meat was significantly higher than levels in other types. These differences between fatty acids were mostly deriving by higher total n-6 PUFA content. In a sausage made of Mangulitsa meat, the values of atherogenic (IA) and thrombogenic (IT) health lipid indexes are lower. Kulen and sremska sausages made from the Mangalitsa pork meat was superior in terms of colour, odour, taste, after taste and overall acceptability. This study demonstrate that pig breed have an effect on the chemical content, fatty acids profiles and sensory properties of dry fermented sausages.

1. Introduction

In Europe, particularly in Mediterranean region, there is a special interest in autochthonous meat products produced from traditional pig breeds extensive way of breeding. Meat and meat products from local pig breeds generally have a good public acceptance and positive media image, and they are often considered to be better quality, than the meat and meat products from conventional, noble pig breeds and crossbreeds.

Mangalitsa is a typical representative fat breed of pig, for example, carcass sides are 65-70% fat and about 30-35% meat [1]. Unlike the colour of other pig meat, the Mangalitsa pig meat had a darker colour. The Mangalitsa fat was whiter and the intramuscular fat content of meat and thickness of back fat was considerably greater than fat of noble pig breeds. The lower saturated fatty acid (SFA) content and higher proportion of unsaturated fatty acid (USFA) compared with that in meat from other fat pig breeds is favorable from a human-nutrition point of view [2, 3].

Cholesterol levels in blood depend not only on dietary cholesterol, but also on the amount of fat and the fatty acid composition of the diet [4]. The influence of fat on cholesterol content can also be observed through the index of atherogenicity (IA) and thrombogenicity (IT), which includes those fatty acids that influence the change of cholesterol. Nutritionists advise a reduction in total fat intake, primarily of SFA and trans fatty acids, which are associated with an increased risk of cardiovascular disease and some kinds of cancers. Moreover, supporting that they reduce their fat intake, nutritionists force consumers to increase
their intake of polyunsaturated fatty acids (PUFA), specifically n-3 PUFA, on account of n-6 PUFA. The PUFA/SFA and n-6/n-3 PUFA ratios have, accordingly, become some of the most significant parameters in evaluating the nutritional value and healthfulness of foods [5, 6, 7].

However, inline with modern trends directed at revitalizing and advancing traditional food production processes, autochthonous meat products made from local breeds are obtaining importance. Moreover, numerous studies have been conducted to determine the characteristics of traditional and naturally fermented sausages throughout the world [8,9,10].

2. Materials and methods

2.1. Animals and Diets

In this study, thirty mature male pigs, ten of each breed, were included: Swallow-Belly Mangalitsa, Moravka and Swedish Landrace. Pigs were housed in an experimental animal house, equipped appropriately to meet the requirements of the study, at the Institute for Animal Husbandry, Zemun, Serbia. All pigs had access to green forages (pasture, clover) ad libitum, with the addition of a feed mixture based on corn and wheat. Animals were stunned, slaughtered and exsanguinated at a local slaughterhouse at 105.2 ± 5.0 kg live weight. Meat was processed 24 h after slaughter and cooling.

2.2. Sausage Formulation and Processing

The examined variants of kulen and sremska sausages (Table 1) were manufactured in a processing plant of the Institute for Animal Husbandry. Three sausages were taken from each variant for all analyses and each analysis was done in duplicate.

Table 1. The percentage of meat originating from specified pig breeds in different types of fermented dry sausages

<table>
<thead>
<tr>
<th>Pig meat different breeds</th>
<th>Fermented dry sausage types</th>
<th>Kulen</th>
<th>Sremska sausage</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>KM</td>
<td>KL</td>
<td>KMM</td>
</tr>
<tr>
<td>Mangalitsa %</td>
<td>100</td>
<td>-</td>
<td>50</td>
</tr>
<tr>
<td>Moravka %</td>
<td>-</td>
<td>-</td>
<td>50</td>
</tr>
<tr>
<td>Swedish Landrace %</td>
<td>-</td>
<td>100</td>
<td>-</td>
</tr>
</tbody>
</table>

Meat used in the production of kulen sausage contained little fat or connective tissue, being primarily meat from the leg, shoulder and some parts of neck and a firm backfat tissue, was also used. Muscle and adipose tissue (75:25) was chopped in a cutter (Seydelman K60, Germany) to achieve 10 mm granulation. After chopping, the chopped meat was transferred to a mixer and the other ingredients of the filling were added: 2.3% table salt, 0.4% saccharose, 0.3% garlic (powder), 0.3% pepper and 0.8% ground sweet and hot red paprika. The filling was then firmly stuffed into the natural pig colon casings. Smoking and maturation of sausages was carried out in the winter period. The smokehouse temperature ranged from 10 to 15º C, and humidity from 75 to 90%. Kulen sausages were smoked during the first 4 weeks. Subsequently, sausages were transferred to the ripening room at a temperature of 10 to 12º C. The ripening was carried out in a drying chamber under controlled conditions (Maurer, Germany).

The examined variants of sremska sausage were produced on the same day and in an identical manner. Meat and fat (85:15) were ground in a cutter (Seydelman K60, Germany) to 8 mm. The same amounts of ingredients were added to sausage variants: 2.3% salt, 0.011% NaNO2, 0.3% dextrose, 0.20% garlic and 0.5% sweet red paprika. The mixture was filled in pig small intestines of around 32 mm diameter. After stuffing, the sausages were hung on sticks and the ripening was carried out in a drying chamber under controlled conditions (Maurer, Germany).
2.3. Analyses

2.3.1. Chemical Analysis.
The following measurements were taken to analyse the chemical composition of the Kulen sausages after 90 days maturation: protein, water, total fat, ash, total fatty acid and cholesterol concentrations. Chemical composition was determined by the methods defined by the Association of Official Analytical Chemists [11].

2.3.2. Extraction of Total Lipids to Determine Fatty Acids.
To determine the concentration of fatty acids, total lipids were extracted by the rapid extraction method, using solvents on the Dionex ASE 200. A homogenized sample, mixed with diatomaceous earth, was extracted with a mixture of hexane and isopropanol (60 : 40 v/v) in a 33 mL extraction cell at a temperature of 100 °C and under nitrogen pressure of 10.3 MPa. The extract thus obtained was steamed in a nitrogen flow at a temperature of 50 °C until dry fat remains were obtained [12].

2.3.3. Determination of Fatty Acids.
Fatty acids as methyl esters were detected by capillary gas chromatography with a flame ionization detector. A predetermined quantity of lipid extracts, obtained by the rapid extraction method, was dissolved in tert-butyl methyl ether. Fatty acids were converted to fatty acids methyl esters (FAME) with trimethylsulphonium hydroxide, according to the SRPS EN ISO 5509:2007 method. FAMEs were analysed with the GC-FID Shimadzu 2010 device (Kyoto, Japan) on a cyanopropyl-aryl column HP-88 (column length 100, internal diameter 0.25 mm, film thickness 0.20 μm). The injected volume was 1 μL. Temperatures of the injector and detector were 250 °C and 280 °C, respectively. Nitrogen was used as a carrier gas, 1.33 mL/min, with a split ratio of 1 : 50, while hydrogen and air were used as detector gases. The temperature of the column furnace was programmed to range between 120 °C and 230 °C. The total duration of analysis was 50.5 min. Methyl esters of acids were identified according to their retention times, which were compared with those of the mixture of methyl esters of fatty acids in the standard Supelco 37 Component FAME Mix [12].

From the data on the fatty acid composition, the following were calculated:

1) Index of atherogenicity (IA): indicating the relationship between the sum of the main saturated fatty acids and that of the main classes of unsaturated [35, 36].
The following equation was applied:

\[ IA = \frac{[4 \times C14:0 + C16:0 + C18:0]}{[\Sigma MUFA + \Sigma PUFAs-n6 + \Sigma PUFAs-n3]} \]

2) Index of thrombogenicity (IT): showing the tendency to form clots in the blood vessels. This is defined as the relationship between the pro-thrombogenic (saturated) and the anti-thrombogenic fatty acids (MUFAs, PUFAs-n6 and PUFAs-n3), [35, 36].
The following equation was applied:

\[ IT = \frac{[C14:0 + C16:0 + C18:0]}{[0.5 \times MUFA + 0.5 \times PUFAs-n6 + 3 \times PUFAs-n3 + PUFAs-n3/PUFAs-n6]} \]

2.3.4. Determination of Cholesterol Content.
Cholesterol content was measured with a HPLC/PDA on the HPLC Waters 2695 Separations Module, with a Waters 2996 Photo Diode Array Detector, as defined by Maraschiello et al.[13].
2.3.5. Sensory Analysis.
For sensory evaluation of kulen and sremska sausages, quantitative descriptive analysis (QDA) was used. The evaluation of sensory properties of sausages were conducted by 10 selected and trained professional assessors (three males and seven females with an average age of 34.5 years) with previous experience in the evaluation of fermented dry sausages. During testing, one sample of sausage was presented at a time and the assessors were asked to rate the following nine attributes of the sausages on a numeric-descriptive scale from 1 (extremely unacceptable) to 7 (extremely acceptable): appearance, cross-section, colour intensity, odour intensity, taste, consistency, acidity, aftertaste and overall acceptability. Samples were taken from the middle of the sausages after 90 days maturation. The sausages were cut into pieces of approximately of 5 mm. These samples, individually labeled with three-digit random numbers, were left for 20 min to reach room temperature. Finally, the samples were served at room temperature on white plastic dishes. Evaluations were performed in individual sensory test booths, prepared as described in accordance with ISO 6658:2005. The assessors were given unsalted crackers and room temperature water to clean the palates and remove residual flavors at the beginning of the session and in between samples. White fluorescent light was used during tests.

2.3.6. Statistical Analysis.
Statistical analysis of experimental data was carried out by analysis of variance (ANOVA) (SAS 9.1.3 software package, 2002–2003). The differences between the different types of sausage were tested using Tukey’s test. The effect of sausage type on fatty acids, chemical content and sensory scores was determined.

3. Results and discussion
3.1. Basic Chemical Composition.
The chemical composition of the different types of kulen and sremska sausages are shown in Table 2. The lowest protein content was found in sremska sausage type SLM and the highest in the kulen type KL. Kulen and sremska sausages made from the meat of Mangalitsa and Moravka pig breeds (type KM and SM) had the lowest moisture content, plus the highest fat content, and these were significantly different in comparison with other types of sausages (P<0.001). Some studies have indicated the occurrence of lower protein content in Mangalitsa meat (m. longissimus), compared to the meat of commercial pig breeds [1, 14,15], thus partly explaining the slightly lower protein content in the kulen types KM and all types of sremska sausages in the current study. Low moisture content is typical of similar products from Greece, Hungary and Croatia [16], and is a consequence of not only drying, but also of a relatively high fat content. In our study, the ash content of the kulen and sremska sausages varied among the all types of sausages (P<0.001). The lowest ash content was found in sremska sausage type SM, and the highest in the sremska type SL. The established pH values ranged from 4.72 (sremska type SL) to 5.25 (sremska type SM) (P<0.001). At the results of studies Vuković et al.[17], beginning of the ripening process of traditional kulen, pH values range from 5.6 to 5.8, which correspond to the pH value of cooled pork meat, and subsequently they start to decline. Certainly, the sugars (fructose, glucose, sucrose) which are natural ingredients of the paprika spice, have a major impact on the decline of pH value in ripening kulen. According to literature data [18], ground paprika contains approximately 15% of sugars, whereas the total sugar content in locally-produced ground paprika is higher and amounts to approximately 25%. The lowest ash content was found in sremska sausage type SM, and the highest in the sremska type SL.

The naturally fermented dry sausages from Mediterranean region are generally characterised by low acidity with a final pH ranging from 5.2 to 6.4, in Italy [19,20], Greece [21], Spain [22; 23] and France [24].
Table 2. Chemical composition of different type fermented dry sausages (means ± standard error)

<table>
<thead>
<tr>
<th>Traits</th>
<th>KM</th>
<th>KL</th>
<th>KMM</th>
<th>SM</th>
<th>SL</th>
<th>SLM</th>
<th>P²</th>
</tr>
</thead>
<tbody>
<tr>
<td>Protein (%)</td>
<td>27.20±0.17a</td>
<td>35.79±0.17b</td>
<td>34.24±0.17c</td>
<td>29.16±0.17d</td>
<td>28.04±0.17e</td>
<td>23.20±0.17f</td>
<td>***</td>
</tr>
<tr>
<td>Water (%)</td>
<td>23.62±0.20a</td>
<td>37.92±0.20b</td>
<td>35.70±0.20c</td>
<td>21.67±0.20d</td>
<td>39.41±0.20e</td>
<td>33.30±0.20f</td>
<td>***</td>
</tr>
<tr>
<td>Fat (%)</td>
<td>39.30±0.22a</td>
<td>15.02±0.22b</td>
<td>21.00±0.22c</td>
<td>39.45±0.22d</td>
<td>22.00±0.22e</td>
<td>34.92±0.22f</td>
<td>***</td>
</tr>
<tr>
<td>Ash (%)</td>
<td>4.61±0.09a</td>
<td>5.25±0.09b</td>
<td>5.67±0.09c</td>
<td>4.48±0.09d</td>
<td>5.83±0.09e</td>
<td>4.86±0.09f</td>
<td>***</td>
</tr>
<tr>
<td>pH</td>
<td>5.24±0.10a</td>
<td>5.05±0.10b</td>
<td>5.03±0.10c</td>
<td>5.25±0.10d</td>
<td>4.72±0.10e</td>
<td>4.73±0.10f</td>
<td>***</td>
</tr>
<tr>
<td>F/Pratio³</td>
<td>1.45±0.01a</td>
<td>0.42±0.01b</td>
<td>0.61±0.01c</td>
<td>1.35±0.01d</td>
<td>0.78±0.01e</td>
<td>1.51±0.01f</td>
<td>***</td>
</tr>
</tbody>
</table>

1Fermented sausages type dependent on the percentage of meat from differing pig breeds
2NS- not significant (P ≥ 0.05);*: Statistical significance at the level of P<0.05;**: Statistical significance at the level of P<0.01;***: Statistical significance at the level of P<0.001;
3Fat/Protein ratio; a-e Means in the same row with different letters are significantly different (P<0.05).

3.2. Fatty Acid Composition.

The fatty acid profiles of the six different fermented dry sausages are introduced in Table 3. Palmitic acid (C16:0) was the most represented SFA, oleic acid (C18:1 n-9) and linoleic acid (C18:2 n-6) were the most represented MUFA and PUFA for the all types of sausages.

The levels of PUFA in fermented dry sausages made from the meat of Mangalitsa pig breed was significantly lower (P<0.001) than levels in other types of fermented dry sausages. These differences were mainly produced by lower total n-3 PUFA content (P<0.01), and also by lower levels of total n-6 PUFA (P<0.001). These led to lower n-6/n-3 ratios in fermented dry sausages type KM (16.96) and SM (14.38). In spite of that though, the n-6/n-3 ratio of unsaturated fatty acids in other types of sausages were between 25 and 37. In separate trials, Hoz [25] and Valenciaet al.[26], both found, in their control groups of fermented dry sausages, lower ratios of n-6/n-3 fatty acids (12.05 and 13.86, respectively), compared to our findings. MUFA values ranged from 43.49 to 52.80 percent. The sausage types SM and KM, made from the meat of Mangalitsa pigs, contained higher levels of MUFA (P<0.001) than the other types of sausages. Additionally, oleic acid (C18:1 cis-9), cis-vaccenic acid, (C18:1 cis-11) and palmitic acid (C16:1) levels in these types of sausages were considerably higher than in the other types. Kulen and sremska sausages type KM and SM had higher USFA (P<0.01) and lower SFA (P<0.001) levels. Overall, the USFA was definitely significantly higher in types of sausages made from the meat of Mangalitsa pig breeds.
Table 3. Fatty acid composition (%), cholesterol content (mg/100g), Index of atherogenicity (IA) and Index of thrombogenicity (IT) of different fermented dry sremska sausages.

<table>
<thead>
<tr>
<th>Traits</th>
<th>Fermented dry sausages³</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>KM</td>
</tr>
<tr>
<td>C14:0</td>
<td>1.21±0.05</td>
</tr>
<tr>
<td>C16:0</td>
<td>26.27±0.09a</td>
</tr>
<tr>
<td>C16:1</td>
<td>3.87±0.07a</td>
</tr>
<tr>
<td>C17:0</td>
<td>0.31±0.03</td>
</tr>
<tr>
<td>C18:0</td>
<td>11.25±0.08a</td>
</tr>
<tr>
<td>C18:1c-9</td>
<td>42.73±0.14a</td>
</tr>
<tr>
<td>C18:1c-11</td>
<td>4.38±0.07a</td>
</tr>
<tr>
<td>C18:2n-6</td>
<td>6.37±0.10a</td>
</tr>
<tr>
<td>C18:3n-6</td>
<td>ND</td>
</tr>
<tr>
<td>C18:3n-3</td>
<td>0.39±0.03</td>
</tr>
<tr>
<td>C20:0</td>
<td>0.17±0.02</td>
</tr>
<tr>
<td>C20:1</td>
<td>0.85±0.03</td>
</tr>
<tr>
<td>C20:2</td>
<td>0.63±0.07</td>
</tr>
<tr>
<td>C20:3n-6</td>
<td>1.33±0.06a</td>
</tr>
<tr>
<td>C20:3n-3</td>
<td>0.08±0.02</td>
</tr>
<tr>
<td>C22:1/C20:4</td>
<td>0.14±0.02a</td>
</tr>
<tr>
<td>SFA</td>
<td>39.22±0.17a</td>
</tr>
<tr>
<td>MUFA</td>
<td>51.97±0.20a</td>
</tr>
<tr>
<td>PUFA</td>
<td>8.80±0.16a</td>
</tr>
<tr>
<td>USFA</td>
<td>60.78±0.31a</td>
</tr>
<tr>
<td>MUFA/PUFA</td>
<td>5.94±0.09a</td>
</tr>
<tr>
<td>MUFA/SFA</td>
<td>1.33±0.01a</td>
</tr>
<tr>
<td>PUFA/SFA</td>
<td>0.22±0.00c</td>
</tr>
<tr>
<td>Total n-3</td>
<td>0.47±0.03</td>
</tr>
<tr>
<td>Total n-6</td>
<td>7.70±0.12a</td>
</tr>
<tr>
<td>n-6/n-3</td>
<td>16.96±1.98a</td>
</tr>
<tr>
<td>Cholesterol</td>
<td>50.16±0.20a</td>
</tr>
<tr>
<td>IA</td>
<td>0.70±0.00a</td>
</tr>
<tr>
<td>IT</td>
<td>1.24±0.01a</td>
</tr>
</tbody>
</table>

Values are means and standard error (mean ± SE).

¹Fermented sausage type dependent on the percentage of meat from differing pig breeds (Table 1)

²**NS- not significant (P ≥ 0.05);***: Statistical significance at the level of P < 0.05;***: Statistical significance at the level of P < 0.001;***: Statistical significance at the level of P < 0.001;***: Statistical significance at the level of P < 0.001;

Means in the same row with different letters are significantly different (P<0.05).

The PUFA/SFA ratio is, nowadays, recommended to be above 0.4 to 0.5 in order to prevent both an excess of saturated fatty acids with a negative effect on low density lipoprotein (LDL) cholesterol plasmatic level, and an excess of PUFA, some of them being precursors of powerful clotting agents and...
also being involved in the etiology of some cancers [28]. In our study, the PUFA/SFA ratio was determined to be the lowest in sausages type KM and SM (0.22 and 0.23), with none of the levels in the sausages exceeding 0.40. Baggio and Bragagnolo [29] ascertained that PUFA/SFA for Italian type salami was 0.4 at the end of production.

3.3. Cholesterol Content.
The cholesterol content in kulen and sremska sausages at the conclusion of the production process ranged from 50.16 mg/100 g (KM) to 66.00 mg/100 g (KMM), with significant differences between the samples (P<0.001). Based on the obtained results (Table 3), we can conclude that sausages made from the mixed meat and fat of Mangalitsa and Moravka indigenous pig breeds contained the highest percentage of cholesterol, while the lowest cholesterol level was seen in sausages produced from the Mangalitsa pig breed.

For salami Milano, Zanardi et al.[30] established cholesterol contents ranging from 94.8 to 110.5 mg/100g. Baggio and Bragagnolo [29] for Italian type salami found the cholesterol content ranged from 48 to 57 mg/100g. Pleadin et al.[31] concluded that the average cholesterol content of industrially fermented sausages was 58.48 to 105.24 mg/100g, until that of homemade fermented sausages was up to 75.07 mg/100g.

3.4. Sensory Properties.
The results of sensory analyses by professional trained assessors are presented in figures 1 and 2. Kulen type KMM and sremska type SLM were awarded the lowest marks and so had the least acceptable appearance, while the kulen type KL and sremska type SL was rated the most acceptable. The cross-section of kulen sausage made from the meat of Mangalitsa pigs was assessed as the poorest. Sremska sausage type SL was the most consistent sausage produced. Product colour was correlated with the colour of the meat used in production. The meat of Mangalitsa pigs was darker than the meat of Swedish Landrace and Moravka; therefore, sausages made from the meat of Mangalitsa breed were assessed as too dark, and received a somewhat lower grade than the other sausage types. Odour was the sensory indicator most affected by the pig breed. The most typical and the best sausages were made from the meat of Mangalitsa breed. The odour of this sausage type was rich and very pronounced, and received a much higher grade than the other kind of sausages. The taste of all sausage types were similar and graded from 5.08 to 6.08, while after taste was graded from 5.17 to 5.33. Professional evaluators gave sremska sausage type SM the highest marks for both taste and after taste. The overall sensory acceptability scores of the examined products showed some differences. Sremska sausage type SM was of the highest stable quality. On the other hand, the quality properties of sausage made from Moravka pork meat were significantly poorer. The current study has shown that the Sremska sausages type SM was good sensory quality, which was in line with Ortiz-Somovilla et al. [32] and Moretti et al. [33]. Overall, the professional assessors found sremska sausage type SM, was the most acceptable. Sausages made only from the meat of Mangalitsa breed (type KM and SM) had better sensory characteristics, thus confirming the work of Radman et al. [34], who found that some pig breeds are suitable for the production of dry fermented pork sausages.

Fig 1. Sensory properties of different types of kulen fermented dry sausage rated by professional assessors (scale test rating)
Fig 2. Sensory properties of different types of sremska fermented dry sausage rated by professional assessors (scale test rating)

4. Conclusion
The results of this study demonstrated that pig breed have an effect on the sensory and chemical characteristics of fermented dry sausages. According to the results of the present research, it is possible to produce, with the appropriate combination of meat and fat from autochthonous pig breeds alone, kulen and sremska dry fermented sausages, with a respectable chemical content, a favourable and reasonably healthful fatty acid composition and with sensory qualities acceptable to discerning consumers. Provided market possibility exist for kulen and sremska sausages, these results should contribute to encouraging the sustainable breeding of the Moravka and Mangalitsa pigs.

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