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# TOTAL FATTY ACIDS COMPOSITION OF RAW AND RIPE SLAVONIAN KULEN IN RELATION TO RAW MATERIAL USED <sup>1</sup>

Danijel KAROLYI<sup>2</sup>, Tomislav ČURIĆ<sup>3</sup>

#### ABSTRACT

The experiment was performed to study the changes in composition of total fatty acids during the ripening of Slavonian kulen (SK), dry-fermented sausages produced from raw material (pork meat and back fat) obtained from the feeding experiment in which toasted (T) or thermally non-treated (NT) corn-grain were used in pig's diets. All sausages were prepared using the same production technology and under the similar conditions of fermentation/ripening. Chemical analyses (content of moisture, crude protein, crude fat, ash and total fatty acid composition) were performed on the samples (n = 6) of T and NT batches taken before stuffing (raw pork/fat mixture) and on the dry-sausages (n = 6) of both groups after six months of ripening. The results indicate that T corn-fed pork/fat raw mixture did not differ in chemical composition, but had less monounsaturated (i.e., less C18:1) and more polyunsaturated (i.e., more C18:2n-6 and C18:3n-3) fatty acid composition as compared to NT raw mixture. As expected, the chemical composition throughout drying/ripening underwent a significant change; the content of moisture decreased, while the contents of protein, fat and ash increased to similar final values in both groups. On the other hand, ripening had little effect on the initial fatty acid composition; only C22:5n-3 proportion decreased with time in both groups. In conclusion, total fatty acid profile of ripe SK in this study largely reflects the fatty acid composition of raw materials used, without major changes during the six-month ripening.

Key words: meat products / dry sausages / Slavonian kulen / composition / fatty acids / ripening

# **1** INTRODUCTION

Slavonian kulen (SK) is a traditional dry-fermented sausage that originated in Slavonia region in eastern Croatia, which is greatly appreciated among local consumers and holds excellent market position. It is made from a mixture of selected and minced pork and back fat, and salt and spices such as red paprika and garlic, filled into pork blind gut (*cecum*). After stuffing, the SK is cold smoked and dried/ripened afterward for several months until the shelf-stability and typical sensory characteristics (e.g. appearance, taste and aroma) are achieved. It is known that the final characteristics of dry-fermented sausages, as well as their preservability are a result of complex physicochemical, biochemical and microbiological changes of raw meat/fat mixture that take place during the sausage processing. These modifications involve various processes such as acidification, dehydration and changes in colour and texture, along with the degradation of proteins and lipids by the action of either endogenous enzymatic systems or those of microbiological origin (Toldrá, 2002). In particular, lipids undergo a series of breakdowns which include hydrolysis and release of free fatty acids, which directly or as a substrate for further oxidative reactions and the formation of volatile compounds contribute to the aroma of the final product (Franco *et al.*, 2002). In addition, lipids are under special attention in terms of nutritional quality of sausages, as this types of products in general represent a significant

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<sup>2</sup> Univ. of Zagreb, Fac. of Agriculture, Dept. of Animal Science, Svetošimunska cesta 25, HR-10000 Zagreb, Croatia, e-mail: dkarolyi@agr.hr

<sup>3</sup> E-mail: tcuric.fax@gmail.com

Table 1: Ingredients and	d main fatty acids of the diets
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Ingredients	% of feed	
Corn	52.0	
Soybean meal (44%)	24.0	
Triticale	15.0	
Wheat bran	5.15	
Calcium carbonate	1.4	
Mono-calcium phosphate	1.2	
Sodium chloride	0.5	
DL-methionine (98%)	0.03	
L-lysine (78.8%)	0.22	
Premix <sup>1</sup>	0.5	
Fatty acids (%) <sup>2</sup>	Non-treated (NT) corn	Toasted (T) corn
C16:0	17.54	15.22
C18:0	2.36	2.99
C18:1	24.85	21.86
C18:2n-6	50.18	54.35
C18:3n-3	1.92	4.00

<sup>1</sup> Premix provided per kg of feed: vitamin A 4000 I.U., vitamin D3 500 I.U., BHT 100 mg, Cu 20 mg, Zn 50 mg.

<sup>2</sup> Determination of fatty acids, in the form of methyl esters, was performed by gas chromatography using Chrompack CP 9000 equipped with a flame ionising detector.

source of fat and saturated fatty acids in human nutrition (Valsta *et al.*, 2005).

An important quality aspect of dry sausages in general and their lipid fraction in particular is the type of raw material used. This regards to factors such as breed or production system and, in the case of products from pork in particular, the animal's diet, since fatty acid composition of pork largely reflects the composition of dietary fats (Pettigrew and Esnaola, 2001). Hence, the aim of the present work was to study the changes in composition of total fatty acids during the ripening of SK sausages produced from raw material obtained in feeding experiment in which different thermal treatments (toasted *vs.* non-treated) of corn-grain were used to alter the fatty acid composition of pigs diet.

# 2 MATERIAL AND METHODS

Pork and back fat used to prepare SK sausages were obtained from two groups of commercial Hypor fatteners ("Klas" d.d. Nova Gradiška) produced under the similar conditions of housing and feeding with a difference only in toasted (T) or thermally non-treated (NT) corn hybrid (Bc-462) that is used in feed composition. Ingredients and main fatty acids of diets are shown in Table 1.

All SK sausages used in the study were produced in single processing plant ("Klas" d.d. Nova Gradiška) following the similar manufacturing steps and using the same type of ingredients. Briefly, the ingredients used in the composition of the raw mixture were lean pork (selected from leg, back, shoulder and neck

parts) and firm back fat (10%), table salt (2%) and spices (1% of red hot paprika, 0.5% of red sweet paprika and 0.5% of garlic). Meat and back fat were minced (particle size 8 mm), mixed with salt and spices and stuffed in previously cleaned pork blind gut (*ceacum*). After being stuffed, the sausages were mildly smoked for 4 weeks by combustion of hardwoods (mainly beech) and than transferred to a drying/ripening chamber (12–16 °C and 65–80% of relative humidity) where they stayed until achieving the shelf-stability and sensory properties typical for ripe product at the age of 6 months.

Chemical analyses were performed on the samples

Table 2: Cl	hemical	composition	(g/kg)	of ra	w and	ripe	Slavonian	kulen	in	relation t	o raw	material	usea
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Component	Slavonian kulen									
	Raw		Ripe (6 months	Significance <sup>a</sup>						
	Т	NT	Т	NT	R	RM	RxRM			
Dry matter	$330.8\pm20.3$	$323.2\pm26.5$	$676.3 \pm 43.3$	$679.9\pm31.7$	***	ns	ns			
Moisture	$669.2\pm20.3$	$676.8\pm26.5$	$323.7\pm43.3$	$320.2\pm31.7$	***	ns	ns			
Protein	$203.2\pm5.1$	$207.2\pm6.4$	$365.3\pm32.1$	$352.3 \pm 17.2$	***	ns	ns			
Fat	$120.5\pm24.8$	$115.0\pm36.3$	$230.5\pm39.1$	$245.5\pm15.4$	***	ns	ns			
Ash	$9.4\pm0.74$	$9.8\pm0.68$	$61.5 \pm 6.5$	$59.3\pm3.5$	***	ns	ns			

Data are means  $\pm$  standard deviations of six samples; T – toasted corn-fed pork, NT – non-treated corn-fed pork; <sup>a</sup> Significance of main effects (R – ripening time, RM – raw material used) and their interaction (RxRM): \*P < 0.05; \*\*P < 0.01; \*\*\*P < 0.001; ns – not significant

(n = 6) of T and NT batches taken before stuffing (raw pork/fat mixture) and on the dry-sausages (n = 6) of both groups after six months of ripening. The moisture, ash, protein and fat content were determined by standard methods for meat and meat products group (ISO 1997, 1998, 1978 and 1973, respectively). The fatty acid composition (of total lipids) was determined by gas liquid chromatography using in situ transesterification method (Park and Goins, 1994). The content of fatty acid methyl esters (FAME) was determined using Agilent Technologies 6890 N (USA) gas chromatograph equipped with a flame ionisation detector and the Supelco Omegawax™ 320 (length 30 m, internal diameter 0.32 mm and film thickness 0.25 µm) capillary column for FAME separation. Separated FAMEs were identified by the comparison with the retention times of the FAMEs in a standard mixture (Nu-Check Prep, Inc, Elysian, USA). The same standard mixture was used to determine the response factor (Rf) for each fatty acid. The mass portion of each fatty acid in the sample was determined using the Rf and

the factor of conversion of fatty acid content from the FAME content.

Data were analysed with two-way analysis of variance by the ANOVA procedure (SAS, 2002) using a model including the main effects of raw material, ripening time and their interaction.

#### **3 RESULTS AND DISCUSSION**

Chemical composition (g/kg) of raw and ripe SK sausages in relation to raw material used is presented in Table 2.

The results indicate that raw pork/fat mixtures from batches T and NT do not differ in terms of moisture, protein, fat and ash content. As expected, the chemical composition throughout drying/ripening process underwent a significant change; the content of moisture decreased while the content of protein, fat and ash increased in a similar way in both groups. The mean values of moisture

Table 3: Fatty acid composition (g/100 g of total FA) of raw and ripe Slavonian kulen in relation to raw material used

	Slavonian kulen									
Fatty acid	Raw		Ripe (6 months)		Significance <sup>a</sup>					
	Т	NT	Т	NT	R	RM	RxRM			
C14:0	$1.23\pm0.08$	$1.23\pm0.10$	$1.22\pm0.09$	$1.19\pm0.08$	ns	ns	ns			
C16:0	$21.54\pm0.75$	$21.41\pm0.95$	$21.96 \pm 0.74$	$21.88 \pm 0.95$	ns	ns	ns			
C16:1	$2.26\pm0.30$	$2.25\pm0.20$	$2.27\pm0.36$	$2.10\pm0.47$	ns	ns	ns			
C18:0	$10.99 \pm 1.21$	$11.05\pm0.76$	$10.93 \pm 1.15$	$10.95\pm0.72$	ns	ns	ns			
C18:1	$40.58\pm0.61$	$42.13\pm2.07$	$40.38\pm0.47$	$42.28 \pm 1.51$	ns	**	ns			
C18:2n-6	$17.37 \pm 1.35$	$16.02 \pm 1.61$	$17.15 \pm 1.13$	$15.75 \pm 1.61$	ns	*	ns			
C18:3n-3	$1.10\pm0.07$	$0.66\pm0.11$	$1.21\pm0.05$	$0.73\pm0.17$	ns	***	ns			
C20:0	$0.18\pm0.03$	$0.20\pm0.02$	$0.18\pm0.02$	$0.19\pm0.02$	ns	ns	ns			
C20:1	$0.89\pm0.13$	$0.94 \pm 0.09$	$0.83\pm0.13$	$0.89\pm0.12$	ns	ns	ns			
C20:2n-6	$0.79\pm0.11$	$0.74\pm0.07$	$0.79\pm0.09$	$0.74\pm0.07$	ns	ns	ns			
C20:3n-6	$0.18\pm0.02$	$0.19\pm0.03$	$0.17\pm0.02$	$0.17\pm0.03$	ns	ns	ns			
C20:4n-6	$0.97\pm0.20$	$1.15\pm0.41$	$0.86\pm0.16$	$0.88\pm0.29$	ns	ns	ns			
C20:3n-3	$0.18\pm0.04$	$0.10\pm0.02$	$0.16\pm0.01$	$0.11\pm0.03$	ns	***	ns			
C22:4n-6	$0.21\pm0.03$	$0.26\pm0.07$	$0.20\pm0.03$	$0.23\pm0.07$	ns	ns	ns			
C22:5n-3	$0.40\pm0.06$	$0.36\pm0.10$	$0.17\pm0.02$	$0.15\pm0.03$	***	ns	ns			
Σ MUFA	$43.91 \pm 0.64$	$45.48 \pm 2.02$	$43.99\pm0.55$	$45.93 \pm 1.71$	ns	**	ns			
$\Sigma$ PUFA	$21.34 \pm 1.70$	$19.64 \pm 2.04$	$20.94 \pm 1.38$	$19.02\pm2.07$	ns	*	ns			
$\Sigma$ SFA	$34.75 \pm 1.77$	$34.88 \pm 1.60$	$35.07 \pm 1.64$	$35.05 \pm 1.52$	ns	ns	ns			
Σ n-3	$1.74\pm0.12$	$1.18\pm0.14$	$1.57\pm0.07$	$1.15 \pm 0.23$	ns	***	ns			
Σ n-6	$19.60 \pm 1.60$	$18.46 \pm 1.94$	$19.43 \pm 1.29$	$17.87 \pm 1.94$	ns	ns	ns			

Data are means  $\pm$  standard deviations of six samples; T – toasted corn-fed pork, NT – non-treated corn-fed pork; <sup>a</sup> Significance of main effects (R – ripening time, RM – raw material used) and their interaction (RxRM): \*P < 0.05; \*\*P < 0.01; \*\*\*P < 0.001; ns – not significant

in the SK observed in this study were somewhat lower than those (36–38%) reported for kulen from households (Vuković *et al.*, 2011; Karolyi, 2011). With regard to protein and fat content, the observed values were within the limits of characteristics specific to traditional SK production (Karolyi, 2011).

Table 3 shows the total FA composition of raw and ripe SK made from T and NT corn-fed pork.

It may be observed that T corn-fed pork/fat raw mixture had less monounsaturated (i.e., less C18:1) and more polyunsaturated (i.e., more C18:2n-6, C18:3n-3 and C20:3n-3) fatty acid composition as compared to NT raw mixture. These differences in fatty acid profile between T and NT batches can be attributed to the difference in fatty acid composition of diets (Table 1), as in pigs the fatty acid deposition in body fats largely reflects dietary fatty acid composition and therefore, can be influenced by dietary means (Pettigrew and Esnoala, 2001). Unlike the chemical composition, drying/ripening had a little effect on total fatty acids composition; only C22:5n-3 proportion decreased with time in both groups, while for the rest of fatty acids the initial proportions persisted until the end of ripening. The lack of changes in total fatty acids composition during ripening has also been described in other traditional pork sausages, e.g., from Italy (Moretti et al., 2004) or Spain (Franco et al., 2002). On the other hand, the formation of free fatty acids, a phenomenon that is typical for the ripening of dry-fermented sausages, is not considered in this study and therefore remains to be investigated in SK.

# 4 CONCLUSIONS

It can be concluded that total fatty acid profile of ripe SK in this study largely reflects the fatty acid composition of raw materials used, without major changes after the six-month drying/ripening. This finding can be useful when planning strategies for developing healthier pork and pork products such as SK, by modification of its fatty acid composition.

#### 5 ACKNOWLEDGEMENT

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