

Content of volatile fatty acids in model mortadella type sausage manufactured with substitution of meat tissue by soya grits.

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Introduction

Categories of vegetable origin meat substitutes used by meat industry greatly differ in their chemical composition mainly regarding the contents of protein, fat and carbohydrates. Soya flour, soya grits and texturates manufactured from them are carbohydrates rich meat extenders containing soluble mono and oligosaccharides, polycarbohydrates and protein-carbohydrates complexed with 10%. Using soya flour or grits which contain approx. 34 percent of carbohydrates as components of sausage mixture, much attention is required to their behaviour in final products in processes such as fermentation, which they may potentially undergo during processing and particularly during storage and distribution.

Evidence indicating presence of volatile fatty acids originated from many sources in scalded fermented sausage has been reported by several authors. /1, 3, 4, 8, 2, 5/.

In this paper we report a study on influence of the 0, 15 and 25 percent substitution of meat tissue by soya grits in model mortadella type sausage on the content of volatile fatty acids (VFA) during several steps into technological process and up to 72 hours of final product storage at 4°C.

Experimental

Preparation of model sausage.

Model mortadella type sausage was manufactured in laboratory scale. Composition of control

of sausage mixture was as follows:

scalded lean beef meat	37.5%
scalded lean pork meat	37.5%
scalded pork back fat/lard	25.5%
water in relation to meat	30.0%
NaCl " " "	0.02%
basic raw material	2.5%
spices /pepper/ in relation to all basic raw material	0.1%

Substitution of meat tissue in sausage mixture by soya grits at a level of 0, 15 and 25% was made using equations suggested by Tyszkiewicz /11/ and accordingly experimental mixtures composed as shown in Table 1.

Table 1 - Composition of model sausage mixture

Ingredients in grammes ^a	Level of meat tissue substitution %/		
	0	15	25
lean pork meat	750	637.5	562.5
lean beef meat	750	637.5	562.5
pork back fat /lard/	500	500.0	500.0
technologically added water	450	382.5	337.5
amount of meat tissue substituted	-	225.0	375.0
amount of meat substitute used/soya grits/	-	94.5	157.5
amount of water for substitute hydration	-	123.0	205.5

^aOther ingredients as per control batch of sausage in percent.

Processing and sampling procedure

Sausage mixtures were prepared in laboratory silent cutter and were stuffed to artificial casing casings 36 mm in diameter, scalded at 75°C for 25 min., chilled under running cold water for 2 min., surface was dried for 10 min. at room temperature and then the ready product was finally chilled and stored for up to 72 hrs. at 4°C. Three separate replications each of 3 model mortadella type sausage were manufactured. Control and experimental batches of sausage were processed simultaneously i.e. from the same raw material. At different stages of technological process i.e. after completion of sausage mixture preparation, after scalding and pre-chilling and after refrigerated storage at 4°C for 24, 48 and 72 hrs. samples were taken and apart from sausage mixture, after removing the casing, they were ground 3 times in laboratory meat grinder, well mixed and the mixture used immediately for analysis.

Analytical methods

Samples were analysed /in duplicate/ for dry matter and fat free dry matter, crude protein method, crude fat /ether extract/, ash content after burning the sample at 550°C according to standard methods. Amount of carbohydrates was calculated from difference according to the following equation:

$$\text{Percent of carbohydrates} = 100 - (\% \text{ protein} + \% \text{ fat} + \% \text{ water} + \% \text{ ash})$$

Quantitative determination of the sum and selected individual steam-volatile fatty acids

Isolation, separation and determination of the sum and volatile fatty acids (VFA) were accomplished by the methods described in selected sources /4,6,9/ by compilation of suggested procedures. A 40g sample was homogenized with 40ml of 80% ethyl alcohol. The homogenate was filtered on a Buchner filter under vacuum and the residue with filter paper again homogenized with 40ml of 80% ethyl alcohol and filtered. Extraction of fatty acids was repeated additionally 2x more i.e. altogether 4 times. The residue on a filter was washed with 80% ethyl alcohol, in total 200ml of the extract was collected.

VFA were isolated by steam-distillation. 100ml of alcoholic extract was transferred to distillation flask of Parnas-Wagner apparatus and 5ml of conc. H_2SO_4 was added than distilled water to make the total volume of 150ml and 450ml of condensate was collected. The total volume of collected distillate was neutralized by titration with 0.1N NaOH in order to estimate the sum of VFA expressed thereafter as acetic acid. The collected 450ml of alkaline distillate was under reduced pressure evaporated to dryness in a rotary evaporator at 45°C. The dry sodium salts of volatile fatty acids were dissolved in several small portions of acetone and transferred in stages to test tube and acetone was evaporated to dryness at 45°C. Dry residue was then dissolved in 0.1 ml of acetone and the resulting solution was acidified by adding 0.4ml of conc. orthophosphoric acid in order to release fatty acids and the test tube was immediately stoppered and swirled for 2-3 min. After sedimentation of orthophosphoric sodium salt the free VFA dissolved in acetone were quantitatively determined using PYE UNICAM-104 gas-chromatograph. The following analytical conditions were observed: glass column length 3.2m, 10% DEGS on Chromosorb W AWMCS, 80/100 mesh, flame-ionizing detector, carrier gas - nitrogen, flow rate: 60ml/min., chart speed: 5mm/min., attenuation: 2×10^{-10} , 20×10^{-10} , column temperature: 160°C, injection volume: 1nl. Quantitation was carried out by comparison of sample peak heights with peak heights of standard mixture also subjected to steam distillation and on the basis of which standard curve was prepared.

Results and discussion.

Chemical composition changes between batches of model sausage in selected phases of manufacturing and during storage are shown in Table 2. Referring to fermentation no significant differences in the content of carbohydrates was found between control and experimental batches of sausage although the amount of carbohydrates expressed in 100g FFDM are 5.72, 7.24, and 10.62% respectively. During processing and particularly during storage in all three batches of sausage the amount of carbohydrates decreased and when calculated in 100g FFDM in batch with 25% of meat substitution, the level of carbohydrates, in relation to the amount in sausage mix, decreased significantly after 24 hrs. of storage and with 15% substitution after 48 hrs. The observation indicates that the carbohydrates undergo quite intensive fermentation even the temperature of storage was far below the optimal required for bacterial activity. The amount of carbohydrates brought into sausage mix with soya grits as meat substitute therefore be considered as not indifferent from palatability point of view because of potential undesirable acidification of the sort of sausage manufactured i.e. usually not demonstrating strong acidity.

The highest initial amount of the sum of VFA expressed as acetic acid was detected in sausage mix of control batch. The observed picture has changed after 72 hrs. of storage when the determined quantity of VFA was biggest in sausage manufactured with 25% meat tissue substitution by soya grits and amounted to 157.78 mg of acetic acid/100g FFDM while in control batch and with 15% substitution 118.78 and 138.82mg/100g FFDM respectively, although the differences found are statistically not significant, Table 4 and 5.

As can be seen from Table 3 acetic acid is the main product of fermentation and it should be considered as the main contributor to the sum of volatile acids. The amount of acetic acid determined after 72 hrs. of storage in all three batches of sausage was statistically significant in relation to the initial quantity. In sausage manufactured with 15 and 25 percent substitution the level of acetic acid increased significantly between 48 and 72 hrs. of storage and indicated intensification of fermentation processes. As can be seen from data presented in Table 3 other determined volatile fatty acids are present in incomparably smaller amounts in relation to acetic acid and it seems that their organoleptic contribution to the taste of sausage is not significant, because considering 20mg of acetic acid/100g of sausage with 25% meat substitution by soya grits after 72 hrs. of storage as a 100%, the amount of propionic acid accounts for only 5.5%, butyric 4.05%, valeric 1.8% and caproic 1.45%. However, detailed studies on this effect are required.

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Table 2 - Composition of model mortadella type sausage

	Percent of meat tissue substitution by soya grits														
	0					15					25				
	Phases of manufacturing process and duration of storage														
	I	II	III	IV	V	I	II	III	IV	V	I	II	III	IV	V
Moisture %	35,9	37,8	38,4	39,1	42,2	36,4	38,4	39,1	40,4	42,1	38,1	38,9	39,4	40,8	44,8
Protein %	18,9	19,7	20,3	20,8	22,7	18,9	19,6	20,3	20,9	22,1	19,5	19,9	20,1	20,8	23,1
Fat %	13,7	14,8	15,0	15,3	16,8	13,9	15,0	15,4	16,1	17,0	14,1	14,9	15,1	15,8	17,4
Carbohydrates %	2,3	2,2	2,2	2,3	2,4	2,4	2,2	2,3	2,5	2,6	2,7	2,4	2,7	2,7	2,9
FFDM = 100%	17,0	18,1	18,1	18,3	19,5	17,5	18,8	18,8	19,5	20,0	18,6	19,0	19,3	20,0	21,7
Protein / FFDM %	80,5	81,6	83,1	83,7	86,0	79,1	79,7	82,0	82,2	85,1	76,0	78,2	78,2	79,1	80,4
Fat / FFDM %	13,8	12,3	12,1	12,6	12,5	13,7	11,9	12,5	12,8	13,1	14,6	12,8	13,8	13,4	13,6
Carbohydrates / FFDM % as difference	5,71	6,08	4,81	3,67	1,54	7,24	8,37	5,49	5,01	1,75	9,31	9,03	7,96	7,50	5,95

I = sausage mix, II = sausage after scalding and chilling, III, IV, V = 24, 48, 72 hours of storage, respectively.
 Presented figures are means of three separate productions.

Table 3 - Volatile fatty acids in model mortadella type sausage

Acids	Percent of meat tissue substitution by soya grits														
	0					15					25				
	Phases of manufacturing process and duration of storage														
	I	II	III	IV	V	I	II	III	IV	V	I	II	III	IV	V
Acetic	17,1	16,3	17,1	16,1	23,2	16,3	15,6	17,8	19,2	27,7	14,1	14,1	14,1	19,7	33,3
S.D.	2,3	2,7	1,7	3,1	3,1	3,4	2,1	2,5	2,3	3,8	3,8	3,8	3,5	3,0	6,9
Propionic	100,6	90,4	94,7	87,8	118,7	91,3	84,3	94,7	98,0	138,8	75,4	74,0	73,0	98,1	153,8
S.D.	10,3	9,3	6,9	11,8	14,7	12,9	12,3	13,2	12,0	17,0	20,0	18,9	17,9	10,4	35,3
Butyric	7,6	7,2	6,8	7,9	11,2	5,1	8,8	8,8	10,3	16,5	5,9	7,1	9,2	9,5	20,0
S.D.	1,5	1,5	1,8	2,7	1,5	1,0	2,5	0,7	2,4	1,8	2,5	2,1	1,1	1,5	4,5
Valeric	44,4	39,7	37,4	43,0	57,5	28,8	47,3	47,2	52,9	82,3	31,8	37,3	47,5	47,8	92,6
S.D.	7,2	4,6	8,0	13,5	8,9	8,2	12,0	3,8	12,1	6,4	14,6	11,5	7,0	10,9	23,5
Caproic	0,5	0,2	0,5	0,3	0,5	0,1	0,5	0,4	0,6	0,7	0,2	0,5	0,4	0,5	1,1
S.D.	0,2	0,1	0,2	0,2	0,1	0,0	0,2	0,1	0,4	0,2	0,1	0,3	0,2	0,2	0,4
Heptanoic	2,8	1,3	2,7	1,8	2,5	0,8	2,4	2,3	2,9	3,5	1,4	2,4	2,0	2,7	5,1
S.D.	1,3	0,5	1,2	0,8	0,3	0,2	0,9	0,7	1,9	1,1	0,6	1,7	0,8	1,1	1,9
Octanoic	0,3	0,3	0,4	0,3	0,4	0,2	0,3	0,3	0,3	0,5	0,2	0,2	0,3	0,5	0,8
S.D.	0,0	0,1	0,1	0,2	0,1	0,0	0,1	0,1	0,2	0,3	0,1	0,1	0,2	0,3	0,3
Nonanoic	2,0	1,9	2,1	1,9	1,9	1,2	1,7	1,6	1,7	2,5	0,9	1,1	1,5	2,7	3,7
S.D.	0,1	0,3	0,5	1,0	0,3	0,3	0,5	0,4	1,1	1,3	0,3	0,5	1,3	1,4	1,4
Decanoic	0,4	0,3	0,4	0,5	0,3	0,3	0,3	0,3	0,3	0,5	0,2	0,2	0,3	0,3	0,4
S.D.	0,2	0,2	0,2	0,2	0,0	0,0	0,0	0,1	0,2	0,1	0,1	0,1	0,1	0,1	0,1
Undecanoic	2,2	1,8	2,2	2,7	1,8	1,3	1,4	1,7	1,7	2,5	1,0	1,1	1,4	1,7	1,7
S.D.	1,3	0,9	0,8	0,8	0,2	0,3	0,3	0,5	0,8	0,6	0,4	0,5	0,5	0,5	0,6
Dodecanoic	0,3	0,2	0,3	0,2	0,2	0,2	0,2	0,3	0,3	0,3	0,2	0,2	0,2	0,3	0,3
S.D.	0,0	0,1	0,1	0,1	0,1	0,0	0,0	0,0	0,1	0,0	0,0	0,1	0,1	0,0	0,0
Tridecanoic	1,7	1,1	1,9	1,3	1,0	1,1	1,1	1,6	1,7	1,3	1,3	1,1	1,1	1,7	1,4
S.D.	0,2	0,5	0,4	0,8	0,4	0,1	0,2	0,2	0,1	0,1	0,2	0,4	0,3	0,2	0,3

Expressed as mg of acetic acid / 100 g sausage c = mg/100g sausage d = mg/100g FFDM

Table 4 - Statistical analysis of VFA versus phases of manufacturing and storage

Phases	Sum of VFA ¹			Acetic			Propionic			Butyric			Valeric			Caproic		
	Batches of model sausage																	
	AA1	BB1	CC1	AA1	BB1	CC1	AA1	BB1	CC1	AA1	BB1	CC1	AA1	BB1	CC1	AA1	BB1	CC1
I-II	-	-	-	-	-	-	-	+	+	-	-	-	-	-	-	-	-	
I-III	-	-	-	-	+	+	-	-	-	+	+	-	-	-	-	-	-	
I-IV	-	-	-	-	-	+	+	-	-	-	-	-	-	-	-	-	-	
I-V	-	+	+	+	+	+	-	+	+	+	+	-	-	+	-	-	-	
II-III	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
II-IV	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
II-V	+	+	+	+	+	+	+	+	+	-	-	+	+	-	+	-	-	
III-IV	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
III-V	+	-	+	+	+	+	+	+	+	-	-	+	-	-	-	-	-	
IV-V	-	+	+	+	+	+	-	-	-	+	-	-	-	-	-	-	-	

I = sausage mix, II = scalded, chilled sausage, III, IV, V = 24, 48, 72 hours of storage

A = control sausage mg/100g B = 15% meat substitution mg/100g
 A1= " " mg/100g FFDM B1= " " " mg/100g FFDM

C = 25% meat substitution mg/100g
 C1= " " " mg/100g FFDM

Volatile fatty acids expressed as mg of acetic acid

Table 5 - Statistical analysis of VFA during phases of manufacturing and storage versus experimental batches of sausage

VFA	Comparable batches of sausage	Phases of manufacturing and periods of storage									
		I	Ia	II	IIa	III	IIIa	IV	IVa	V	Va
Sum of VFA	A - B	-	-	-	-	-	-	-	-	-	-
	A - C	-	-	-	-	-	-	-	-	-	-
	B - C	-	-	-	-	-	-	-	-	-	-
Acetic acid	A - B	-	-	-	-	-	-	-	-	+	+
	A - C	-	-	-	-	-	-	-	-	+	-
	B - C	-	-	-	-	-	-	-	-	-	-
Propionic acid	A - B	+	+	-	-	-	-	-	-	-	-
	A - C	-	-	-	-	-	-	-	-	-	-
	B - C	-	-	-	-	-	-	-	-	-	-
Butyric acid	A - B	+	+	-	-	-	-	-	-	-	-
	A - C	+	+	-	-	-	-	-	-	-	-
	B - C	-	-	-	-	-	-	-	-	-	-
Valeric acid	A - B	-	-	-	-	-	-	-	-	-	-
	A - C	-	-	-	-	-	-	-	-	-	-
	B - C	-	-	-	-	-	-	-	-	-	-
Caproic acid	A - B	+	+	-	-	-	-	-	-	-	-
	A - C	-	-	-	-	-	-	-	-	-	-
	B - C	-	-	-	-	-	-	-	-	-	-

I = mg/100g mince

II = mg/100g of sausage

III = mg/100g sausage 24 h. stor.

Ia = mg/100g FFDM of mince

IIa = mg/100g FFDM of sausage

IIIa = mg/100g FFDM saus. 24h. stor.

IV, V respectively 48 and 72h. stor.
 IVa, Va " " " " " "

A = Control batch

B = 15% meat tissue substitution

C = 25% meat tissue substitution