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EFFECTS OF SOY PROTEIN, WHEY POWDER AND WHEAT GLUTEN ON QUALITY CHARACTERISTICS OF COOKED BEEF SAUSAGES FORMULATED WITH 5, 10 AND 20% FAT

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> ABSTRACT INTRODUCTION MATERIALS AND METHODS RESULTS AND DISCUSSION CONCLUSIONS REFERENCES

ABSTRACT

In this study the effects of fat level (5%, 10% and 20%) and soy protein (SP), whey powder (WP) or wheat gluten (WG) on binding properties, color, textural and sensory characteristics of cooked beef sausages were evaluated. Fat reduction decreased emulsion stability and water holding capacity (WHC) and resulted in higher cooking losses. Addition of SP, WP and WG increased WHC and emulsion stability (ES). The most effective additive on WHC and ES was SP. Reduced fat products were darker compared with 20% fat controls, adding fat replacers increased inside yellowness value. Fat level had no effect on shear force values, WP and WG reduced shear force values regardless of the fat level. Fat replaces resulted in no detrimental effects on sensory characteristics in low fat sausages.

Key words: sausages, whey protein, soy protein, wheat gluten, frankfurters

INTRODUCTION

Meat and meat products contain elements, which in certain circumstances and in inappropriate proportions have negative effect on human health. Some of these constituents are present in live animals for instance fat, cholesterol, residues from environmental pollution or the use of pharmaceuticals, etc. [26]. World health

organization have drawn up the following nutritional recommendations; fat should provide between 15-30% of the calories in the diet, saturated fat should not provide more than 10% of these calories and cholesterol intake should be limited to 300 mg/ day. This resulted in an increased demand for fat reduced and low-fat meat products [44].

Fat in processed meat products contributes functional and organoleptic characteristics. Reducing the fat content in low-fat frankfurter type sausages may alter meat quality, products became firmer, more rubbery, less juicy, darker in colors more costly and increased purge in vacuum package [25, 28]. To succeed in producing low-fat, healthy and palatable food products other ingredients must be chosen to replace fat. Fat replacements should contribute a minimum of calories to the product and should not be detrimental to organoleptic qualities. Most substitutes can be categorized as: leaner meats, added water, protein based substitutes, carbohydrate – based substitutes and synthetic compounds [15, 27, 42]. Fat content in processed meat products can be readily reduced through formulation with leaner meats but this increases cost and it may also lessen product quality. Reformulation with fat substitutes can cause a reduction in partial binding, darker product color, lack of beef flavor, reduced browning reactions and shorter shelf life [25, 26, 28].

Soy products (soy flours, concentrates and isolates) have been used in meat systems to improve functional properties such as water binding and textural properties [9, 39, 40, 46]. Milk proteins can act both as emulsifiers and as water and fat binders in foods [20]. Whey products have also been used as binders and extenders [12, 22, 23]. Wheat gluten is one of several vegetable proteins used as meat extenders or fillers in comminuted meat products to enhance functional characteristics and reduce cost [15, 25, 27, 28, 42]. Meat source in the production of fat reduced and low fat frankfurters could affect the quality of the products [38]. Most of the research has been carried out on the effect of fat level and various fat replacers on properties of frankfurters produced from a combination of beef and pork. Frankfurter type sausages in Turkey mostly formulated with beef and beef fat. Our objective was to determine the effects adding soy protein, whey powder or wheat gluten on processing and quality characteristics of all beef sausages with different fat levels.

MATERIALS AND METHODS

Experimental Design

Three sausage batters were prepared containing 5%, 10% and 20% beef fat. Soy protein-SP (Danpro S-760 Central Soya Inc. Denmark, 69.5% protein), whey powder-WP (Pinar Dairy Company Inc.Turkey, 13% protein, demineralized), and wheat gluten-WG (Agrostar Ltd, India, 75% protein) were added separately to these emulsions at an addition rate of 4%. In the reduced fat products, water was added to replace the fat to ensure the same protein (of meat origin) level in all formulations. Three controls without SP, WP or WG were also formulated to give a total of (3×4) 12 treatments. Two replications of the experiment were conducted each at separate times.

Sausage Production

Lean beef and beef fat were obtained from a local meat processor. Raw meat had 78.4% moisture, 19.3% protein, 1.6% fat and pH was recorded 5.56. The meat and fat were frozen, separately in polyethylene bags and kept frozen for up to 3 weeks at –18°C. Partly thawed lean beef was ground through a 3 mm plate. After that lean meat was mixed with curing ingredients, (2.5% salt, 0.25% sucrose, 0.3% nitrite, 0.15% polyphosphate, 0.05% ascorbic acid) and half the ice/water and chopped for 1 min in silent cutter. After that beef fat, seasonings, 4% corn starch and fat replacer (SP,WP or WG at a level of 4%) together with the reminder ice/ water added and the batter was chopped at high speed for 4 min. The batter temperature did not exceed 13°C. Immediately after chopping the batter of each formulation was stuffed into natural sausage casings (salted sheep intestines). Sausages were hand linked at 15 cm intervals and smoked in smoking chamber (drying at 40°C for 1 hr, smoking at 40°C for 30 min) and heat processed in water (85°C) to an internal temperature of 73°C. The sausages were showered for 2 min and chilled at 4°C for 24 hours. After chilling the samples were vacuum packaged and stored in cooler at 4°C until analysis.

Proximate analyses

Immediately after processing for each raw sausage batter and cooked sausage; pH [37], moisture [37], fat [16] and protein [45] were determined.

Water Holding Capacity

WHC of batter was determined using the procedure of Hughes et al. [21] with a modification. Samples (10 g) were placed in glass jars and heated for 60 min in a water bath (90°C). After heating, the samples were removed, cooled to room temperature, wrapped in a cotton cheesecloth, and centrifuged in 10 ml polycarbonate tubes (containing absorbent cotton wool) for 15 min at 4000 rpm at 4°C. The cheesecloth was removed and the sample weights were recorded. The following equation was used to calculate WHC:

%WHC = $1-T/M \times 100 = 1-B-A/M \times 100$

T = total fluid loss during heating and centrifugation

B = weight of sample before heating

A = weight of sample after heating and centrifugation

M = total water content in the sample

Emulsion stability

Emulsion stability was measured using the procedure of Hughes et al. [20] with a modification. Approximately 25 g (exact weight recorded) of the raw emulsion was placed in a centrifuge tube and centrifuged for 15 min at 6000 rpm after that heated in a water bath for 60 min at 70°C and then centrifuged again for 20 min at 6000 rpm. The pellet was removed and weighed and the supernatant was poured into pre-weighed crucibles and dried overnight at 100°C. The volume of total expressible fluid (TEF) and the percentage fat was calculated as follows: TEF = (weight of centrifuge tube and sample) - (weight of centrifuge tube and pellet)

%TEF = TEF/ sample weight \times 100,

% Fat = (weight of crucible + dried supernatant) – (weight of empty crucible) / TEF \times 100

Cooking Loss

Weight of sausages before and after cooking was recorded and the cooking loss was expressed as a percentage difference between the raw and cooked weights.

Color Measurement

Color measurements were performed 1^{st} week of storage. Minolta 508d spectrophotometer was used to evaluate lightness (L*), redness (a*) and yellowness (b*). Outside and inside sausage color were evaluated. Two sausages per treatment were used and four measurements were taken form each sausage.

Shear Force

Shear force analysis was performed on the Instron Universal Testing Machine (Model 1140). Shear force as peak force was determined by shearing a 2.5 cm height and 2 cm diameter core sample with a Warner Bratzler blade mounted in the Instron. 10 kg load cell was used at a speed of 100 mm/ min. Shear force (kg) was expressed maximum force to shear the sample.

Data were recorded as the average of 6 samples per treatment.

Sensory Evaluation

Sensory evaluation was conducted the 1st week of storage by a six-member trained panel. The panelists were chosen on the basis of previous experience in evaluating sausages. The following attributes were evaluated on a 3 point or 5 point scale: appearance, flavor and texture. Each attribute was discussed and tests were initiated after panelists were familiarized with scales. Samples were prepared by steeping the sausages in boiling water for 5 min. Tap water and bread were served between the samples to cleanse the palate. Each sample was coded with randomly selected 3-digit numbers.

Statistical analysis

The trial was performed twice and the data was evaluated by two-way analysis of variance (ANOVA). Significance of differences was defined as (p < 0.05).

RESULTS AND DISCUSSION

The pH ranged from 5.86 to 6.04 for batter and from 6.04 to 6.1 for sausage. The pH of cooked sausages increased upon heat processing. Neither fat level nor fat replacers had a significant effect on pH values of batters or finished product (p > 0.05). Proximate analyses indicated that the fat levels in the raw sausage batter were close to targeted fat values but did not exactly match the formulated fat levels and ranged from 5.8% to 6.3%, 10.5% to 11.8% and 21.5% to 22.1%. Incorporation of soy protein (SP), whey powder (WP) or wheat gluten (WG) into formulations did not affect the fat content in the finished product (p > 0.05). Fat levels both in batter and sausages affected moisture content significantly (p < 0.05). Uncooked 5% fat sausages had moisture content. Any increase in fat content may be related to moisture loss on processing. Percentage moisture decreased with increased level of fat (p < 0.05). If the fat content is low, the moisture content is tending to be high. Since with a low fat content, more protein per g of fat is available for moisture retention. Similar results reported by other researchers [10, 19, 41].

Moisture content of cooked sausages was lower than batter, for cooked sausages moisture ranged 62.0% to 73.1% (<u>Table 1</u>). Adding fat replacers did not affect moisture levels (p > 0.05). Slight differences in protein content between treatments were observed, protein levels varied between treatments and ranged from 14.0% to 16.0% for sausages (<u>Table 1</u>). Due to moisture losses during heat processing protein levels were higher in sausages than in uncooked batter. Mittal and Barbut [34], reported that protein levels increased on cooking which could be attributable to changes in total product mass during cooking.

Treatmen t	pH ^a raw	pH cooke d	Fat raw [%]	Fat cooked [%]	Moisture raw [%]	Moisture cooked [%]	Protein raw [%]	Protein cooked [%]
5/0 5/1 5/2 5/3 10/0 10/1 10/2 10/3 20/0 20/1 20/2	5.87 5.93 5.95 5.99 5.88 5.92 5.96 5.86 5.91 6.04 5.95	6.04 6.08 6.10 6.07 6.07 6.07 6.08 6.09 6.05 6.08 6.08 6.08	$\begin{array}{c} 6.2^{a} \\ 5.8^{a} \\ 6.3^{a} \\ 5.9^{a} \\ 11.3^{c} \\ 12.1^{c} \\ 10.9^{c} \\ 11.0^{c} \\ 21.5^{d} \\ 21.9^{d} \\ 22.1^{d} \end{array}$	8.1 ^b 7.4 ^b 7.1 ^{ab} 6.6 ^a 11.8 ^c 10.5 ^c 10.8 ^c 10.9 21.8 ^d 21.8 ^d 22.6 ^d	75.1 ^c 74.4 ^c 74.2 ^c 71.8 ^b 71.5 ^b 70.8 ^{ba} 70.3 ^a 70.0 ^a 69.1 ^a 68.7 ^a	70.8 ^d 71.6 ^d 71.3 ^d 66.6 ^c 68.1 ^c 66.1 ^b 65.9 ^b 63.9 ^a 65.2 ^b 62.4 ^{a b}	13.3 ^a 12.9 ^a 13.4 ^a 13.5 ^a 12.9 ^a 14.3 ^b 13.9 ^a 14.9 ^b 13.7 ^a 12.7 ^a 12.2 ^a	16.0 ^b 15.5 ^a 15.7 ^a 16.0 ^b 15.7 ^a 16.0 ^b 15.0 ^a 14.0 ^a 14.8 ^a 14.1 ^a
20/2	5.92	6.07	22.1 21.6 ^d	22.0 ^d	68.5 ^ª	62.0 ^a	13.1 ^a	14.7 ^a

a Values are mean of 4 replicates. The first letter refers to the fat:

0 no added fat replacer; 1 soy protein; 2 whey powder; 3 wheat gluten. Different letters in the same column (with in each main effect) indicate significant differences (p > 0.05).

Effects of fat level and fat replacers on WHC, emulsion stability and cooking loss were shown in Table 2. Water holding capacity (WHC), varied from 43.0% to 77.9% (<u>Table 2</u>), reducing the fat level from 20% to 5% significantly decreased WHC (p < 0.05). This has been reported for various types of meat products such as frankfurters and beefburgers and turkey frankfurters [2, 37, 21, 22, 34, 35, 47]. Fat replacers increased WHC and emulsion stability (p < 0.05). Higher binding ability of wheat gluten, due to interaction with myosin was reported by Siegel et al. [43] previously. It was reported in many works that adding soy proteins and whey proteins to frankfurter types meat products resulted high WHC [9, 24, 29, 39].

	WHC	Emulsion s	stability [%]	Cooking loss
	[%]	TEF [%]	Fat [%]	[%]
A: Fat level				
5	50.4 ^a	29.4 ^a	8.3 ^a	15.4 ^a
10	63.5 ^b	18.5 ^b	6.8 ^b	13.4 ^b
20	73.0 ^c	9.8 ^c	2.7 ^c	9.26 ^c
SL	0.0	0.0	0.0	0.0
B: Fat replecer				
0	57.5 ^a	23.1 ^a	6.4 ^a	14.4 ^a
1	68.4 ^b	19.8 ^b	6.2 ^a	11.6 ^b
2	62.7 ^b	17.4 ^c	5.5 ^b	12.6 ^b
3	60.5 ^b	16.7 ^c	5.6 ^b	12.2 ^b
SL	0.0	0.0	0.01	0.01
INT AxB	ns	ns	0.01	ns
5/0	43.0	34.2	8.7	17.5
5/1	57.7	30.3	8.3	14.8
5/2	50.9	27.0	8.1	15.3
5/3	49.8	26.3	8.1	14.2
10/0	59.4	22.8	7.6	15.4
10/1	69.7	19.0	7.2	12.4
10/2	64.2	16.7	6.1	12.7
10/3	60.5	15.6	6.5	13.0
20/0	70.3	12.3	3.1	10.1
20/1	77.9	10.1	3.1	7.6
20/2	72.8	8.6	2.3	9.9
20/3	71.1	8.2	2.3	9.2

Table 2. Effect of fat level and fat replacers on WHC, emulsion stability and cooking loss

The first letter refers to the fat: 0 no added fat replacer, 1 soy protein, 2 whey powder, 3 wheat gluten, ns non significant, SL significance level, TEF total expressible fluid. Different letters in the same column (with in each main effect) indicate significant differences (p > 0.05).

Total expressible fluid and fat release of batters was significantly affected by fat level (p < 0.05). Reducing the fat level significantly decreased emulsion stability (p < 0.05). The higher fat level in the formulation resulted in lower expressible fluid. Cavestany et al. [4] found that fat level significantly influenced the total expressible fluid and released fat in bologna and increased emulsion stability. Within each fat level incorporation of soy protein, whey powder and wheat gluten as fat replacer caused a decrease in total expressible fluid (p < 0.05). The effect of interaction between fat level and fat replacers was significant for percentage fat release (p < 0.05). WP and WG had significant effect on fat release at10% and 20% fat levels. Gnanasambandam and Zayas [18] also reported the significant effect of wheat gluten on WHC and emulsion stability in frankfurter type meat products. ND Soy protein had no effect on fat release within each fat level (Table 2). Hughes et al. [22] also reported increased emulsion stability and decreased fat release in sausages formulated with WP. In the present research whey protein bonded fat in the high fat formulation. El-Magoli et al. [13] concluded that addition of whey protein concentrate would increase fat binding in the meat system even at lower fat levels (10%). Whey proteins having excellent surface-active properties that allow them to re-orient and reduce interfacial tension with increased opportunity for fat- protein interacting [31]. Soy proteins can improve water and fat binding and hence aid in emulsion stabilization in meat products, such as bologna and frankfurters [17]. Soy proteins are hydrophilic (absorb and retain water), can form a gel that act as a matrix for holding moisture and fat and possess film forming as well as adhesive properties [36].

In the present research, cooking losses varied from 7.6% to 17.5%, reducing the fat content, caused significant increased in cooking loss, cooking loss was 17.5% in 5% fat controls and 10.1% in 20% fat controls (<u>Table 2</u>). The smallest cooking loss recorded in the present study was for sausages formulated with soy protein and 20% fat. Our findings agree with observations of some authors [7, 22, 34, 45]. However cook yield has been reported to not differ much in the 5-20% range of fat [3]. Controlling the cooking loss is very important to maintain the proper juiciness in the product [47]. Sausages formulated with fat replacers had significant lower cooking losses than ingredient free controls. This is agreement with other studies on a variety of meat products.

In the present study reducing fat level decreased inside L* values (p < 0.05) but had no effect on a* and b* values of sausages (p > 0.05) (<u>Table 3</u>). These results are in agreement with those of Decker et al. [8] who found that lowering fat content from 30 to 15/100g significantly increased the cured meat colour of sausages. The lower the fat level the higher the cured color intensity in sausages due to increase in meat and thus in muscle pigment. Fat is known to influence the internal colour of meat products as reported by Hughes et al. [45]. Mittal and Barbut [47] also reported no difference of a* and b* values with fat in frankfurters. When the myoglobin content is kept constant the colour of frankfurter type products is mostly influenced by fat content and added water. Fat level and fat replacers had no affect on external colour parameters (p > 0.05). The relationship between fat level and sausage colour was observed in other studies [22, 34, 35, 41, 45, 46] and WG increased internal yellowness regardless of the fat level (p < 0.05) (<u>Table 3</u>). This could be explained by the dilution of myoglobin of meat with fat replacers and some extent the colour of the additives. Dzudie et al. [11] who found that sausages with common bean flour had higher yellowness values. Yellowness values of beef burgers were affected by addition of wheat fibers [33].

	External colour			Internal colour			Shear force (kg)
A: Fat level	L*	a*	b*	L*	a*	b*	
5	50.6	11.6	11.1	51.7 ^a	10.2	9.8	6.3
10 20	50.7 51.4	10.9 10.6	10.4 10.7	57.1 ^b 58.3 ^c	10.3 10.5	10.1 10.5	6.8 6.2
SL	ns	ns	ns	0.01	ns	ns	ns
B: Fat replacer							
0	50.6	10.6	9.8	53.7	10.8	9.2 ^a	7.4ª
1	49.9	10.1	9.4	56.8	10.0	10.3 ^b 10.7 ^b	7.3 ^a 4.6 ^b
2 3	51.8 51.4	11.5 11.9	10.7 10.0	55.3 57.0	10.5 10.2	10.7 11.0 ^b	4.0 6.4 ^b
SL	ns	ns	ns	ns	ns		

The first letter refers to the fat level: 0 no added fat replacer, 1 soy protein, 2 whey powder, 3: wheat gluten, ns: non significant, SL significance level. Different letters in the same column (with in each main effect) indicate significant differences (p > 0.05).

Fat level had no effect on shear force values (p > 0.05), similar results were recorded by other researchers [4, 19, 38]. However there is some disagreement about fat level and hardness of sausages, a number of researchers reported on the textural characteristics of meat emulsions with varying fat levels and generally it was concluded that products with increased fat content present less hardness [5, 25]. This contradictory results may explained by different protein content. According to Panareas et al. [38], protein has a greater influence on texture than fat. Within each fat level, shear force values for WP or WG added sausages significantly less than sausages formulated with 20% fat and no added fat replacer. No significant influence adding SP on shear force values of sausages was observed (p > 0.05). Holding protein content steady in low fat products resulted in softer products. WP and WG bonded and retained water and resulted tenderer product. Decreases in shear values of wheat gluten added sausages were also reported [18]. However, in contrast to our findings, Ensor et al. [14] reported addition of WP (0-3%) in full fat knockwurst increased final product hardness. Results from sensory evaluation indicated that (Table 4) fat level significantly affected texture scores of sausages (p < 0.05). Increasing fat level 5% to 20% increased texture scores. 20% fat sausages had higher texture scores. This is in agreement with previous studies on affect of fat level on texture [41,45]. However in contrast to our findings, Cofrades et al. [6], found that high fat frankfurters were harder than low-fat frankfurters. Some authors have reported that overall palatability of sausage is not always dependent on fat content [1, 30]. Adding SP, WP or WG had no effect on texture scores (p > 0.05).

	Apperance	Textur	Flavour			
A: Fat level						
5	3.7	1.9 ^a	3.5			
10	3.6	2.2 ^a 2.6 ^b	3.4			
20	3.6	2.6 ^b	3.4			
SL	ns	0.01	ns			
B: Fat replacer						
0	3.7	2.1	3.7			
1	3.6	2.0	3.3			
2	3.6	2.2	3.5			
3	3.7	2.1	3.4			
SL	ns	ns	ns			

The first letter refers to the fat level: 0 no added fat replacer, 1 soy protein, 2 whey powder, 3 wheat gluten, ns non significant, SL significance level. Different letters in the same column (with in each main effect) indicate significant differences (p > 0.05).

Neither fat level nor fat replacers had an effect on appearance and flavour of sausages (p > 0.05). Smoking and spices may enhanced sausage flavour so the panel could not detected the flavour differences between low and high fat sausages. This is in agreement with other authors who have reported that the flavour of the sausage is not always dependent on fat [6, 13, 30, 34]. Porcella et al. [40] concluded that 5% soy protein level was not detected in chorizo, a kind of raw sausage. Lyonns et al. [32] demonstrated that increasing concentrations of whey protein (8%) decreased flavour scores by masking of spice flavor. In the present study added whey protein level (4%) was lower than this level.

CONCLUSIONS

Decreasing fat content significantly alters WHC, emulsion stability and cooking loss. Decreasing fat level had no effect on flavour scores and shear force values. Fat replacers (soy protein, whey powder and wheat gluten) were successfully used to improve binding properties and had no detrimental effects on sensory characteristics. Neither fat level nor fat replacers had an effect on external colour parameters. WP and WG resulted in soft texture regardless of the fat level. SP, WP and WG could be used successfully in reduced fat all beef sausages.

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