OPTIMIZATION AND QUALITY EVALUATION OF BUFFALO CALF MEAT SAUSAGES INCORPORATED WITH SKIM MILK POWDER AS A COMPLETE FAT REPLACER

Gauri Jairath¹, Diwakar Prakash Sharma², Randhir Singh Dabur², Pradeep Kumar Singh^{3,*} and Ashok Kumar Pathera⁴

ABSTRACT

The study was conducted to maintain the leanness of meat while processing by substituting added fat with skim milk powder (SMP) without breaching quality attributes. Four treatments viz. control: 20% fat, T1: sausages with 3% SMP, T2: sausages with 6% SMP, T3: sausages with 9% SMP, were tried to develop low-fat sausages. Water holding capacity and emulsion stability of T3 were higher (P<0.05) amongst all, but could not pass sensory acceptability, however all attributes of T1 and T2 were higher than control, but comparable to each other. Thus T1 was selected for further quality evaluation and compared with high fat products. The cooking yield, pH, protein content, oxidative stability along with sensory attributes of selected products (low-fat) were significantly (P<0.05) higher, however, vice-versa was true for shear press value and fat and did not affect textural properties. The results concluded that SMP (3%) incorporation could be a way to substitute fat while processing of meat giving 42% lower calorie content.

Keywords: *Bubalus bubalis*, buffaloes, skim milk powder, buffalo male calf meat, lean meat, fat replacer, calorie content

INTRODUCTION

Buffalo calf meat has gained more preference because of its lean character. Consumers of 21st century are becoming more health conscious, paying attention to their platter. Further, domestic and export potential of buffalo meat has raised their importance. But, in India, carabeef is mainly obtained from spent buffaloes. Such meat usually comes out to be dark, coarse and tough in texture with poor processing and eating quality attributes due to more collagen content. Therefore, meat from young male buffalo (<18 months) animals with more collagen solubility (Kandeepan et al., 2009) can definitely provide prime choice to the consumers. Though, the meat of buffalo calf tends to be leaner and well capable to satisfy consumers' demand, but sometimes it needs 15 to 20% fat (vegetable/ animal) incorporation to improve quality of product

¹Division of Livestock Products Technology, Indian Veterinary Research Institute, Regional Station Palampur, Himachal Pradesh, India

²Department of Livestock Products Technology, LalaLajpat Rai University of Veterinary and Animal Sciences, Haryana, India

³Department of Livestock Products Technology, College of Veterinary Science and Animal Husbandry, Nanaji Deshmukh Veterinary Science University, Madhya Pradesh, India, *E-mail: drpradeeplpt@yahoo.co.in

⁴School of Bioengineeirng and Food Technology, Shoolini University, Himachal Pradesh, India

during processing, however the practice raises the suspicion among health conscious consumers as fat consumption comes with caution. Development of obesity, cardiovascular diseases, stroke, and cancer are directly or indirectly linked with high fat intake (Azad *et al.*, 2016). In addition, high fat diet is considered as one of top contributors to obesity development owing to its ability to provide 2.25 times more energy in comparison to carbohydrates and proteins (Mallika *et al.*, 2009).

Hence, the consumers' desire has inclined more towards low-fat meat products which have further stimulated the researchers to focus on the strategies for fat replacement. However, merely reduction of fat doesn't solves the need of leaner character maintenance as it results in dried, hard and less juicy products of low cooking yield with rubbery texture and high shear force value (Kumar and Sharma, 2004). Thus, to comply today's demand, the identification of appropriate fat replacer is much needed.

Skim milk powder (SMP) is widely used as neutral filler with good water binding and structure enhancing property in comminuted meat products (Serdaroglu and Deniz, 2004) and may also be used as fat replacers to develop low-fat products. Along with that, replacement of fat with milk proteins significantly improves the cooking characteristics in low fat meat products (Andic and Boran, 2015). Therefore, utilization of SMP as an alternate to maintain the lean character of buffalo calf meat while processing may be explored. The objectives of present study were the optimization of SMP level in model meat system and study the quality attributes of the developed product.

MATERIALS AND METHODS

Raw meat and materials

Three healthy male buffalo calves (Murrah breed) of around 130 kg each and 10 to 12 months of age were humanely slaughtered in experimental slaughter house of Department of Livestock Products Technology, LUVAS, Hisar, India, as per the standard procedures and animal welfare guidelines. The carcasses were pre-chilled for one day followed by cutting, deboning and trimming. The meat was kept in deep freezer (-18±1°C) after packing in low density polythene bags for further study. All the chemicals, reagents and media were purchased from some Indian firms like CDH Chemicals. New Delhi; Hi-Media, Mumbai and Merck's, New Delhi and skimmed milk powder (Sterling Agro Industries Ltd., New Delhi) was purchased from super market.

Preparation of the sausages

On the basis of preliminary work, formulation (Table 1) and processing protocol to develop sausages were optimized. The deboned meat was minced in an electrical meat mincer (3 mm plate) (Mado Primus Meat Mincer, MEW-613; Dr. Froeb India Pvt. Ltd.) followed by manual mixing of all ingredients and then vacuum tumbled for 2 hours. Emulsion was stuffed in sausage filling machine manually to fill cellulose casings and then were steam cooked for 35 minutes. The cooked sausages were taken out, cooled in chilled water for 5 minutes and casings were peeled off. The lowfat sausages were prepared in the similar manner as mentioned for control sausages, except that SMP was added instead of vegetable fat. Total four treatments were tried viz. control = with 20% fat, T1 = sausages with 3% SMP, T2 = sausages with 6% SMP, T3 = sausages with 9% SMP. The SMP level was selected on the basis of water holding capacity (WHC), emulsion stability (ES) and sensory evaluation. The developed low-fat sausages were compared to high fat control sausages for the various physico-chemical properties [cooking yield, shear press value, pH and thiobarbituric acid reactive substances (TBARS)], texture profile, proximate composition and sensory evaluation.

Analysis

Physico-chemical parameters

WHC: The water holding capacity was estimated as per procedures described byWardlaw *et al.* (1973).

ES: The emulsion stability was determined using the method of Baliga and Madaiah (1970).

Cooking yield: The yield of the products in percentage was calculated as below:

Cooking yield = <u>Weight of cooked product</u> x 100 Weight of raw product

Shear press value: It was analysed using Warner-Bratzler shear probe in Texture Analyser in terms of force (Newton; N), required to shear a sample of 20 mm diameter transversely.

pH: Double electrode pH meter (Cyber Scan pH 510, Eutech Instruments; Thermo Fisher Scientific, Navi Mumbai) was used to determine pH of samples (Trout *et al.*, 1992).

Thio-barbituric acid reacting substances (TBARS): TBARS, the measure of oxidative stability was calculated as mg malonaldehyde/kg of sample (Witte *et al.*, 1970) by multiplying O.D. with K factor (5.2).

Texture profile analysis

Texture exponent program of texture analyser (TA. HD plus, Stable Micro Systems Ltd., Surrey, England) and a probe with compression platform (70 mm diameter) were used to study the textural attributes like hardness, springiness, cohesiveness, gumminess and chewiness of the sausages (Bourne, 1978).

Proximate composition

Proximate composition i.e. moisture, protein, fat, and ash content was estimated using automatic moisture analyser, Kelplus, Socs Plus, and Muffle furnace, respectively, according to the procedures mentioned in AOAC (2005). In addition, Atwater values (fat: 9 kcal/g, protein: 4.02 kcal/g, carbohydrate: 4 kcal/g) were used to account total calories in the product on the basis of 100 portions.

Sensory evaluation

Eight-point descriptive scale described by Keeton (1983) was used to sensory evaluate the samples. Six panellists including faculty members and postgraduate students of the department evaluated the samples after two training sessions and same panel evaluated the samples every time. The warmed samples were offered randomly in coded form after microwaving for 20 seconds along with water for in between mouth rinsing.

Statistical analysis

'SPSS-16.0' (SPSS Inc., Chicago, II USA) software package was used to analyze the data (Snedecor and Cochran, 1994). All the trials were conducted three times and data was subjected to one-way analysis of variance (ANOVA) at 5% level (P<0.05) of significance. Duplicate samples were drawn for each parameter in each experimental trial except for texture and shear press value (n=5) and sensory evaluation (n=6) to have total No. of observations N=6 for all parameters, whereas N=15 for texture profile as well as for shear press value and N=18 for sensory evaluation.

RESULTS AND DISCUSSION

Assessing incorporation level of skim milk powder in model meat system

The selection of SMP level was done on the basis of WHC, ES and sensory attributes (Table 2 and 3).

The WHC of T3 was significantly higher (P<0.05) than T2 followed by T1 and control. The WHC of raw sausages emulsion ranged from 45.06 to 57.44%. The WHC was found significantly higher in low-fat raw emulsions in comparison to control. The WHC of treated samples increased with increase in level of SMP; however, it was at par in T2 and T3. The increase in WHC with SMP can be attributed to the milk proteins resulting in more firm and stable protein complex. This stable meat protein matrix might have reduced the chances of water and fat release, thus, further enhancing the water binding ability of samples (Pietrasik and Shand, 2003). Increase in WHC has been reported by Serdaroglu and Deniz (2004) in turkey rolls incorporated with 3% milk powder.

The stability of emulsion was variably affected with addition of both fat and fat replacers. With increase in level of incorporation, the emulsion became significantly (P<0.05) more stable owing to emulsification of free fat firstly by milk proteins and leaving salt soluble proteins free for water binding (Martien, 1987). Free fat prefers milk proteins more during emulsion formation over both salt and water soluble proteins in fat-water interface. Milk proteins also negatively influence the size of fat globules resulting in smaller sized fat globules which further, aid in water binding as tendency of water expulsion while heating is lower in small fat globules. These results endorse the findings obtained by Eswarapragada *et al.* (2010) in low fat pork sausages.

The colour and appearance score did not differ significantly; however, were higher than control. The flavour scores improved significantly $(P \le 0.05)$ at 3 and 6% level of SMP; however, scores of T3 were lower than of that of T1 and T2, but comparable to control. The lower flavour scores in T3 were probably due to lack of meaty flavour (Serdaroglu and Deniz, 2004) at higher level of SMP incorporation. The texture and tenderness scores did not differ significantly with the increase in incorporation level however, were higher than control. Similarly, no effects on colour and appearance, texture and tenderness were found in pork patties when milk co-precipitates were incorporated by Kumar and Sharma (2003). Juiciness scores increased with increase in SMP percentage. The credit of higher juiciness scores went to milk proteins, improving water binding ability of emulsions, which result in higher moisture retention. Overall acceptability was comparable at 3 and 6% level of incorporation, however lower in sausages with 9% level of incorporation. On the same lines, improved sensory attributes were noticed in chicken steaks when milk co-precipitate were incorporated (Bhoyar et al., 1998).

Though, rise in SMP level of incorporation, increased both WHC and ES, but the sensory scores did not show same trend at 9% level of incorporation. Since, overall acceptability of T3 was significantly lower than other treated products, thus, T1 and T2 clear the gross screening process for final selection. Since WHC and ES did not differ significantly between T1 and T2, lower incorporation level (3%) was chosen while considering economics as well as meaty flavour for the development of finished product for further study.

Quality evaluation of finally developed product (3% SMP)

Three percent skim milk powder was used to develop low-fat buffalo calf meat sausages which were compared with full fat sausages (20% fat) with respect to various quality attributes.

Physico-chemical evaluation of developed product

The products were evaluated for physicochemical properties like cooking yield, shear force, pH and TBA (Table 4) to assess the effect of SMP on cooking losses, texture and oxidative stability of developed sausages.

Cooking yield of developed low-fat product (3% SMP) was found significantly (P<0.05) higher in comparison to control high fat product (20% fat) owing to its higher protein and lower fat contents. Protein itself not only has the properties to trap moisture, but also lead to formation of coherent protein matrix which further lessen the release of fat and water and improves the water binding (Abiola and Adegbaju, 2001; Pietrasik and Shand, 2003). In addition, total amount of fat as well as fat type are the factors which depict the cooking loss in meat products (Hong et al., 2004; Choi et al., 2009). Thus, high content of proteins and lower content of fat, collectively contributed towards improvement in cooking yield of low-fat developed product. The cooking yield was also found higher with the incorporation of milk co-precipitates in pork patties (Kumar and Sharma, 2003). The control samples had the lower cooking yield which may be attributed to the excessive fat separation and water release during cooking (Mansour and Khalil, 1999).

The final developed product required significantly (P<0.05) less force to shear the products than high fat product as replacement of fat with water conferred much slushy texture to sausages (Keeton, 1994; Khalil, 2000). Konjac blend and sodium alginate also imparted the soft texture to low-fat products like bologna and pork patties, respectively (Chin *et al.*, 1998; Kumar *et al.*, 2007) depicted by their lower shear force values.

The recording of significantly (P<0.05) high pH in treatment directly corroborates with the higher water binding properties of the fat replacers. On similar lines, when milk coprecipitate, alginate and carrageenan were used as a fat replacer at 2, 0.15 and 0.5% level, respectively in emulsion products, also resulted in significant rise in pH (Eswarapragada *et al.*, 2010; Lin and Keeton, 1998).

Low-fat sausages were found to be more oxidative stable as expected due to fat replacement. Highest TBARS value of control was because ofits significantly higher fat content (11%). The report of Muhlisin *et al.* (2012) supports the data of oxidative stability in buffalo calf sausages. Similarly, lower TBA values were recorded on day 0 in milk co precipitate and sodium alginate incorporated pork patties (Kumar and Sharma, 2003; Kumar *et al.*, 2007).

Textural attributes

Developed sausages were softer than control one, though the hardness values in Newton were at par statistically (Table 5). More water binding and less release because of milk proteins were the probable reasons for the reduced hardness. Softer texture have also been reported by many authors on addition of fat replacers (Khalil, 2000; Kotwaliwale et al., 2007; Verma et al., 2015). Further, reduced hardness stamped the results of shear force value (Table 4). The springiness and cohesiveness of all the sausages were comparable. Similar results were observed by Khalil (2000) in beef patties. Reduction of fat content in muscle foods, generally, leads to increase in cohesiveness and decrease in springiness in comparison to control (Confrades et al., 1997). But here in sausages, as fat replacer was added, springiness and cohesiveness values came closer to the control owing to properties of SMP to retain moisture content. In addition, protein-protein and proteinwater interactions were more due to milk proteins contributingimproved texture in developed products (Rao et al., 1997). Gumminess, chewiness and resilience values of the developed low fat products were comparable to high fat control which showed that fat replacer was able to impart the texture similar to that of control sausages.

Proximate composition

Perusal of Table 6 indicates the significant difference in proximate composition between control and developed sausages. The moisture content in SMP treatedproduct was significantly (P<0.05) higher attributing to improved WHC, formation of stable protein matrixon addition of milk proteins (Berry and Wergin, 1993). Higher moisture content was also observed in beef patties incorporated with fat replacers (Khalil, 2000; Muhlisin et al., 2012). The fat content in developed productmade it a low-fat product as it was within the limits, set for meat products declared to be lowfat i.e. 10% (Keeton, 1994). The fat and moisture contentshowed inverse relationship as a result of substitution of fat withwater (Kumar et al., 2007). Significantly (P<0.05) improved protein and ash

content in treated productwas attributable to higher respective content in SMP. On the same lines, finding were observed in dairy ingredient enriched turkey meat and oat flour enriched beef patties (Serdaroglu and Deniz, 2004; Serdaroglu, 2006). The moisture to protein ration was higher in the developed products and vice-versa for the control products as predicted by results presenting higher moisture content in treated products. The calorie content of developed low-fat sausages was 40% lower in comparison to high fat sausages owing to fat replacement with SMP. Thus, fat replacement with SMP offered a low-fat, protein rich, low calorie finished product.

Sensory evaluation

SMP showed positive effects on the sensory scores right from colour to overall acceptability (Table 6) and a notable effect on the colour of sausages was perceived by the sensory panellist. The fat replacers incorporated products had significantly higher colour scores in comparison to control sausages due to increase in redness as a result of fat replacement (Hughes et al., 1998). Similarly, mil co-precipitates enhanced the colour and appearance scores when added in pork patties (Kumar and Sharma, 2003). Flavour and textural/ tenderness scores were also significantly ($P \le 0.05$) higher due to SMP incorporation. The lactose content of SMP might have imparted the better flavour scores to the developed sausages (Kumar and Sharma, 2003). The juiciness scores were attributable to their respective, water holding capacity. The overall acceptability of developed and control products were at par. Similar results were reported by Bhoyar et al. (1998) in chicken steaks incorporated with milk co-precipitate.

Nama of ingradiants	Percentage (w/w)			
Name of ingredients	Control	T1	T2	Т3
Lean meat	100.00	100.00	100.00	100.00
Salt	02.00	02.00	02.00	02.00
TSPP	0.40	0.40	0.40	0.40
Sodium nitrite	0.02	0.02	0.02	0.02
Spice mix	02.00	02.00	02.00	02.00
Condiment mix (onion: garlic) 2:1	03.00	03.00	03.00	03.00
Albumen	05.00	05.00	05.00	05.00
Refined wheat flour	02.00	02.00	02.00	02.00
Ice flakes	08.00	08.00	08.00	08.00
Vegetable oil	20.00	-	-	-
Skim milk powder	-	03.00	06.00	09.00

Table 1. Formulation of buffalo calf meat sausages.

Control: sausages with 20% fat, T-1 = sausages with 3% SMP;

T-2 = sausages with 6% SMP, T-3 = 9% SMP.

Table 2. Effects of different levels of skim milk powder on WHC and ES of raw male buffalo calf meat emulsion (Mean ± S.D., n=6).

Treatments/Parameters	WHC	ES
Control	45.06±1.47ª	62.88±1.83ª
T-1	53.15±1.64 ^b	84.60±2.90 ^b
T-2	55.76±2.6 ^{bc}	86.53±2.72 ^b
T-3	57.44±3.33°	90.92±2.68°

Control = sausage with 20% fat; T-1 = sausages with 3% SMP;

T-2 = sausages with 6% SMP; T-3 = sausages with 9% SMP

Means with different superscripts in a row differ significantly ($P \le 0.05$)

Treatments/	Colour	Flavour	Toxturo	Inicipase	Overall
Sensory attributes	Colour	Flavour	lexture	Juiciness	acceptability
Control	7.17±0.24ª	7.03±0.32ª	$7.06{\pm}0.16^{a}$	7.11±0.21ª	7.14±0.23ª
T-1	7.89±0.2 ^b	7.86±0.23 ^b	7.69±0.35 ^b	7.47±0.21 ^b	7.83±0.24°
T-2	7.97±0.12 ^b	7.64±0.23 ^b	7.72±0.35 ^b	7.75±0.26°	7.86±0.23°
T-3	7.97±0.12 ^b	7.25±0.3ª	7.72±0.39 ^b	7.78±0.26°	7.56±0.24 ^b

Table 3. Effects of different levels of skim milk powder on sensory attributes of buffalo calf meat sausages (Mean \pm S.D., n=18).

Control = sausage with 20% fat; T-1 = sausages with 3% SMP; T-2 = sausages with 6% SMP; T-3 = sausages with 9% SMP. Means with different superscripts in a row differ significantly ($P \le 0.05$).

Table 4. Physico-chemical properties of 3% SMP incorporated low-fat buffalo calf meat sausages (Mean \pm S.D., n=6).

Parameters/Treatments	Control	Low-fat sausages
Cooking yield	$75.38{\pm}1.70^{a}$	90.13±1.24 ^b
Shear press value (N)	6.20 ± 0.52^{b}	5.74±0.39ª
pH	5.96±0.01ª	6.05 ± 0.03^{b}
TBA (mg malonaldehyde/Kg	0.592±0.01 ^b	0.558±0.01ª

Means with different superscripts in a row differ significantly (P≤0.05).

Table 5. Textural scores of developed low-fat sausages containing 3% SMP (Mean±S.D., n=15).

Parameters/Treatments	Control	Low-fat sausages
Hardness (N)	23.13±2.26ª	22.31±2.33ª
Springiness	0.81±0.02ª	0.81±0.13ª
Cohesiveness	0.45±0.04ª	$0.45{\pm}0.06^{a}$
Gumminess	10.34±1.86ª	$10.01{\pm}1.70^{a}$
Chewiness	8.33±1.44ª	7.99±1.24ª
Resilience	0.19±0.02ª	0.19±0.03 ^{ab}

Means with different superscripts in a row differ significantly ($P \le 0.05$)

Composition/Treatments	Control	Low-fat sausages		
Moisture	64.71±0.92ª	72.77±0.32°		
Fat	11.57 ± 0.40^{d}	2.33±0.37 ^b		
Protein	17.82±0.60ª	18.57±0.12°		
Ash	1.99±0.05 ^{ab}	2.40±0.06°		
Moisture: protein	3.63	3.91		
Energy content (Kcal/100g)	191.40	111.34		
Sensory attributes				
Colour	7.17±0.24 °	7.89±0.21 ^b		
Flavour	7.03±0.32 ª	7.86±0.23 b		
Texture/Tenderness	7.06±0.16 ª	7.69±0.35 b		
Juiciness	7.11±0.21 ª	7.47±0.21 ^b		
Overall acceptability	7.14±0.23 °	7.83±0.24 ^b		

Table 6. Effects of skim milk powder on the proximate composition and sensory evaluation of developed low-fat buffalo calf meat sausages (Mean±S.D., n=6).

Means with different superscripts in a row differ significantly ($P \le 0.05$).

CONCLUSION

Above results concludes the suitability of skim milk powder as a substitute of fat in buffalo calf meat to maintain the lean character while processing. SMP may be incorporated at 3% level without effecting the meat flavour and other quality characteristics including texture. Further the developed low-fat meat product would be able to satisfy demands of health conscious consumers.

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