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Research Paper

Effect of Modified Soy Protein on Quality Attributes of Extended Chicken Nuggets

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ABSTRACT

A study was conducted to assess the effect of incorporation of modified soy protein (MSP) on the functional and quality attributes of extended chicken nuggets. Negative control group nuggets were prepared by utilizing 67% meat, 10% ice flakes and other non-meat ingredients (binders, spice, condiments, etc.), whereas the positive control and MSP nuggets were prepared with 62% meat, 15% ice flakes and other non-meat ingredients. In case of MSP nuggets, 0.5% of binder was replaced with modified soy protein. No significant ($p>0.05$) difference in pH, emulsion stability, cooking yield, expressible water and proximate composition was observed among the negative control, positive control and modified soy protein incorporated chicken nuggets. The negative control group exhibited significantly higher ($p<0.01$) redness value (a*), cohesiveness, hardness, and fracturability score and lower lightness (L*) value than the positive control and MSP groups. The negative control group also showed significantly ($p<0.01$) higher texture and overall acceptability scores. However, the incorporation of MSP and additional water did not make any significant ($p>0.05$) change in the storage stability of nuggets in terms of oxidative rancidity and microbial counts compared with negative control group. The results revealed that incorporation of modified soy protein enable to reduce the proportion of meat in the chicken nuggets without adversely affecting the quality and sensory attributes.

Keywords : *Chicken products, Soy proteins, Products quality, Storage stability*

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INTRODUCTION

Producing consistent quality products and improving the profitability are the important goals for meat processors. Processors want to reduce the quantity of meat used in the product preparation and optimize costs while improving the texture and taste. Incorporation of non-meat ingredients viz., binders, extenders and additives is an important strategy to produce better quality products and cut down the cost of production. The addition of certain non-meat proteins improves water binding, stabilize fat and control cost (Biswas *et al*. 2016).

Soy proteins are one of the important non-meat additives used by the meat industry. Soy proteins are typical of vegetable proteins with health-enhancing activity. They are thought to be effective in preventing cardiovascular disease, cancer and osteoporosis. Soy-based ingredients also contain another group of bioactive components such as isoflavones, saponins, etc. (Arihara, 2006). Soy proteins (flours, concentrates and isolates) are more commonly used in processed meat products for their functional properties and relatively low cost as compared to lean meat. Being an economical source of food protein, soy proteins have been used extensively in meat products as a binder for improving yields, gelling agent to enhance emulsion stability, meat replacement to reduce costs and fat replacer. Despite the many advantages of soybean, its use as a food material has been limited because of oû-ûavour such as ''beany ûavour'' generated during processing. The lipoxygenases in soybean catalyze the hydroperoxidation of polyunsaturated fatty acids by molecular oxygen which results in the generation of oû-ûavours associated with compounds such as n-hexanal and n-hexanol (Matoba *et al*. 1985).

Many attempts have been made to control such oû-ûavour generation during soybean processing. The grinding of soybean at high temperature is eûective to control lipoxygenase activities and results in reduction of oû-ûavour generation and maximizes protein extraction. Appropriate heat processing also eliminates or inactivates anti-nutritional factors. Recently microbial fermentation technology is also employed to produce beany flavor free soy proteins. However, these processing have significant influence on the functional properties of soy proteins, which primarily depends upon degree of protein denaturation. Protein solubility could affect emulsification, whipping and gelling capacity. In this study,

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the modified soy protein (MSP) manufactured by microbial fermentation process was utilized to develop extended chicken nuggets and evaluate its effect on the quality attributes of chicken nuggets.

MATERIALS AND METHODS

Preparation of chicken emulsion: Fresh chicken procured from local market of Hyderabad was utilized. After removing fat and connective tissues, lean meat was stored at 4 ^{0}C for 24 h before use. The experimental trials were carried out in seven replications. For each of the seven trails, chicken was obtained freshly and utilized for nuggets preparation to nullify the variation in the quality of meat between batches. Chilled chicken samples were cut into small cubes and minced twice (13 mm plate followed by 6 mm plate) using a meat mincer (Scharfen, Germany). The chicken emulsion was prepared by chopping minced chicken and non-meat ingredients in a bowl chopper (Dadaux, France) till the desired consistency. The proportion of chicken, refined wheat flour (maida), modified soy protein (MSP) (Praras Biosciences Pvt. Ltd., Bangalore) and water in the control and treatment groups are as follows:

Negative Control: 67% meat, 10% water, 3 % maida, and no

Positive Control: 62% meat, 15% water, 3 % maida, and no MSP

62% meat, 15% water, 2.5 % maida, and 0.5 % MSP

The proportion of all other ingredients (vegetable oil (10%), salt (1.5%), sodium tripolyphosphate (0.3%), spice mix (1.5%), condiments (onion: garlic; 3:1, 3.5%), sugar (0.2%) and sodium nitrite (0.01%) and processing conditions remained same for both the controls and treatment. The protein, moisture, fat and ash content of modified soy protein were 40.62, 6.34, 0.33 and 21.37 %, respectively. The modified soy protein showed rehydration ability of 259%.

Preparation of chicken nuggets: Chicken nuggets were prepared by manually filling the emulsion into rectangular stainless steel box smeared with vegetable oil. The filled boxes were cooked in a double jacketed stainless steel electric water heater. The chicken emulsion was steam cooked without any pressure until the core temperature of the cooked emulsion block reaches 80^{0}C. After cooking, the boxes were cooled at room temperature for 1 h. After cooling, the cooked blocks were taken out by loosening the sides with sharp knife and sliced in the form of nuggets (3.5 cm L X 1.5 cm WX 1.5 cm H) using meat slicer (Sirman, Italy). Both the control and treatment nuggets were aerobically packaged in a low density polyethylene pouches and stored at 4°C. The nuggets were analyzed for pH, water expression, emulsion stability, cooking yield, instrumental color, texture profile, sensory attributes and proximate composition on the day of preparation. The thiobarbituric acid reactive substances (TBARS) and microbial count (total plate counts and psychrotrophic counts) were estimated for four trials on 0, 5, 10, 15 and 20 days of refrigerated storage (4±1^{0}C). The freeze thaw loss was calculated for three cycles.

Analysis of samples

Proximate composition: Moisture, fat, protein and ash content of MSP and chicken nuggets were determined according to AOAC (2005).

pH: The pH of the chicken nuggets was determined by blending 10 g sample with 50 ml distilled water for 60 s in a homogenizer (ART Moderne Labortechnik, Germany). The pH values were measured using a standardized electrode attached to a digital pH meter (Thermo Orion, USA).

Emulsion stability, cooking yield and expressible water: The emulsion stability was measured by taking 20 g emulsion moulded into a shape of ball and placed in a polyethylene pouch. This was heated in a temperature controlled water bath at 80^{0}C for 20 min. After cooling at room temperature, fluid released was drained and cooked emulsion was wiped with tissue paper to make it dry and weighed (Sen *et al*. 2008). The emulsion stability was calculated as

ES (%) = (Weight of the emulsion after heating / Weight of raw emulsion) X 100

Cooking yield was determined by dividing cooked product weight by the raw uncooked weight and multiplying by 100.

The expressible water (EW) was determined by wrapping 5 g of chicken nuggets in a Whatman No. 1 filter paper, placing in a centrifuge tube and centrifugation at 1500 rpm (Sorvall Biofuge Stratos, Germany) for 5 min. Immediately after centrifugation, the nugget samples were reweighed. The amount of expressible water = (Initial weight of the sample - Final weight of the sample)/Initial weight of the sample X 100 (Ramirez *et al*. 2002).

Instrumental color: Colour measurements were conducted on the surface of samples with a Miniscan XE plus (Hunter Associated Labs, Inc, USA) that had been calibrated against black and white reference tiles (X=78.6, Y=83.4, Z=89.0). CIE L* (lightness), a* (redness) and b* (yellowness) values were obtained using a setting of D65 (daylight, 65-degree light angle). An average value from 4 random locations in each sample surface was used for statistical analysis.

Texture profile analysis: Texture profile analysis (TPA) of cooked chicken nuggets was recorded as per the procedure described by Bourne (1978) using Texturometer (Tinius Olsen, England). Four samples were analyzed under each formulation and the readings were averaged. From the resulting force/deformation curves, the textural parameters of cohesiveness, fracturability, hardness and springiness were calculated.

Ultra structural studies: Emulsion samples were fixed in 2.5% gluteraldehyde in 0.1 M phosphate buffer (pH 7.2) for 24 hrs at 40^{0}C and post fixed in 2 % aqueous osmium tetroxide for 4 hrs. Dehydrated in series of graded alcohols and dried to critical point drying with CPD unit. The samples were scanned under scanning electron microscope (JOEL JSM 5600, USA) at required magnification as per the standard procedure (Bozzola and Russel 1998).

Sensory evaluation: An experienced panel evaluated chicken nuggets on the day of preparation. The panelists rated each sample on an 8-point descriptive scale (8, extremely desirable; 1, extremely undesirable) for colour and appearance, flavor, texture and overall acceptability (Keeton 1983).

Thiobarbituric acid reactive substances (TBARS) value: The thiobarbituric acid reactive substances (TBARS) value (mg malonaldehyde/kg) of chicken nuggets was determined using the extraction method described by Witte *et al*. (1970) with slight modifications as the slurry was centrifuged at 3000 g for 10 min instead of filtration through Whatman No. 42 filter paper.

Microbiological quality: Microbial quality in terms of aerobic plate count (APC) and psychrotrophic count (PTC) was determined as per the procedure of ICMSF (1983).

Freeze thaw loss: The aerobically packed chicken nuggets were frozen and stored in a still freezer at-20 ^{0}C for 3 days before being thawed in a refrigerator (5 ^{0}C) until the temperature at the center of nuggets reached 0 ^{0}C. The thawed nuggets were wiped with tissue papers and weighed. Immediately after weighing, nuggets were refrozen and stored for 3 days and again thawed in a refrigerator (cycle 1). The freeze–thaw cycle was repeated for three cycles. The freeze- thawing loss was determined from the known weights of frozen nuggets and after thawing and expressed as % Freeze thaw loss = (weight of frozen nuggets - weight of thawed nuggets)/weight of frozen nuggets X 100

Statistical analysis: Mean values for various parameters were calculated and compared by analysis of variance using SPSS (SPSS version 13.0 for windows; SPSS, Chicago, IL, USA). The pH, emulsion stability, expressible water, freeze thaw loss, proximate composition, texture profile, instrumental colour, cooking yield and sensory attributes were analyzed using one-way ANOVA. A 3 x 5 factorial design with six replicates was employed for storage data (TBARS value, APC and PTC) with treatments and storage time as main effects using two-way ANOVA. Statistical significance was determined at 95% confidence level ($P < 0.05$).

RESULTS AND DISCUSSION

Proximate composition: The proximate composition of control and treated chicken nuggets are given in Table 1. There was no significant difference ($p>0.05$) in protein, fat, moisture and ash content among the control and treated groups due to incorporation of MSP. Low level of incorporation of soy product/carrageenan and not much difference in the cooking yield could be the reason. Das *et al*. (2008) also reported no change in proximate composition, except protein content due to addition of 15% soy paste.

pH: There was no difference ($p>0.05$) in pH among the control and treated groups due to incorporation of MSP (Table 1). Das *et al*. (2008) also reported that addition of soy protein did not increase the pH signiûcantly compared to control samples.

Emulsion stability, cooking yield and expressible water: The ability of the protein matrix to bind water upon heating and fat stability are crucial in the manufacture of comminuted meat products. These properties determine the emulsion stability and final cooking yield (Nakaiand Li- Chan 1988). The positive control and MSP group showed non-significantly lower emulsion stability and cooking yield and higher expressible water compared to negative controls (Table 1). This might be due to higher moisture content in the formulation. Hughes *et al*. (1998) found that emulsion stability and cooking loss was affected by changing the fat and water content in the formulation. Crehan *et al*. (2000) also indicated that cooking loss increased as added water/ice increased. However, among the higher moisture content formulations, MSP had numerically higher values for emulsion stability and cooking yield and lower expressible water than positive control. This indicates participation of soy proteins in fat holding and water binding in meat systems (Youssef and Barbut 2011). Reichet (1991) reported that the soy proteins act as an emulsifier and fat encapsulating agent by supplementing myosin and actomyosin and thereby prevent fat and water

Table 1: Proximate composition, functional attributes, instrumental colour values, textural profile and sensory attributes of chicken nuggets incorporated with modified soy protein

Quality attributes	Parameter	Negative Control	Positive Control	MSP
Proximate composition	Protein (%)	17.05±0.11	16.42±0.18	16.54±0.20
	Fat (%)	9.38±0.21	9.25±0.09	9.21±0.15
	Moisture (%)	68.40± 0.20	69.09± 0.43	69.77± 0.24
	Ash (%)	2.96±0.28	2.85±0.45	3.05±0.20
Functional attributes	pH	6.29 ± 0.02	6.26 ± 0.02	6.32± 0.02
	Emulsion stability (%)	96.06 ± 0.54	94.47 ± 0.88	95.91± 0.30
	Cooking yield (%)	96.10 ± 0.27	95.35 ± 0.09	96.05± 0.19
	Expressible water (%)	13.89 ± 0.56	14.71 ± 0.77	14.11± 0.45
Instrumental colour values	L*	55.81 ± 1.34^{a}	59.11 ± 1.14^{b}	58.96 ± 0.65^{b}
	a*	9.83 ± 0.02^{b}	8.60 ± 0.14^{a}	8.68 ± 0.34^{a}
	b*	42.50±0.70	39.78±1.24	40.94±1.85
Textural profile	Springiness (cm)	0.841 ± 0.06^{a}	1.085 ± 0.07^{b}	0.882 ± 0.07^{a}
	Cohesiveness (ratio)	1.027 ± 0.12^{c}	0.890 ± 0.13^{a}	0.980 ± 0.13^{b}
	Hardness (N/cm^2)	48.35 ± 2.35^{c}	40.24 ± 2.30^{a}	46.94 ± 1.63^{b}
	Fracturability (N/cm^2)	48.35 ± 2.35^{b}	40.68 ± 1.25^{a}	47.23 ± 1.16^{b}
Sensory attributes	Appearance	7.13±0.08	6.91±0.09	6.97±0.15
	Flavour	6.97 ±0.11	6.78±0.15	6.76±0.13
	Juiciness	6.87 ±0.05	6.82±0.11	6.81±0.17
	Texture	6.94 ± 0.05^{c}	6.08 ± 0.08^{a}	6.58 ± 0.13^{b}
	Overall acceptability	6.93 ± 0.11^{c}	6.00 ± 0.00^{a}	6.53 ± 0.08^{b}

Means ±SE with different superscripts in a row differ significantly ($p<0.05$) ($n=7$)

separation while cooking. Further, studies of cross-linking on gelation of myofibrillar and soy protein mixtures have indicated that a substantial amount of cross-linking of soy and myofibrillar protein occurred at temperatures over 50^{0}C (Ramírez-Suarez and Xiong 2003). This agrees with Pietrasik and Duda (2000) who reported improvement in cooking yield with increasing soy protein levels in the formulation. Reduced cooking loss in soy proteins added meat products was also reported by Matulis *et al*. (1995).

Instrumental color: Colour is an important sensory attribute, which determines the products acceptability rate. Redness values of chicken nuggets were affected by addition of excess water. The negative control group exhibited significantly ($p<0.01$) higher redness value (a*) and lower lightness (L*) value than the positive control and treatment group (Table 1). The red colour of the meat is mainly due to the myoglobin content. According to Candogan and Kolsarici (2003), meat products with higher lean meat contents are expected to be redder in color. Increasing the proportion of non-meat ingredient (water) resulted in lighter and less red products compared to the corresponding higher meat negative control group.

Texture profile analysis: The ability of the protein matrix to bind water upon heating is crucial in determining the texture and other attributes defining final product quality (Nakaiand Li- Chan 1988). The cohesiveness, hardness and fracturability score were significantly ($p<0.01$) higher in the negative control group than the positive control and treatment groups (Table 1). It has been reported that when soy protein replaced lean meat, the resulting product was softer (Keeton 1994). However, when soy protein was added while keeping the level of lean constant (constant muscle protein level), the hardness of the product increased (Matulis *et al*. 1995). Das *et al*. (2008) opined that the higher moisture content might be a contributing factor to the lower hardness in soy paste extended nuggets. Further, Chin *et al*. (2004) reported that the products hardness decrease with increase in moisture content. Similarly, a decline in shear force value with lower lean meat content was reported by Das *et al.* (2008). On the other hand, Chin *et al.* (1999) indicated that replacing 2.2% meat proteins with soy protein isolate did not affect hardness of bologna. The results suggest a high correlation between some of the textural parameters and moisture content of the meat products.

Sensory evaluation: The nuggets prepared without additional water (negative control group) had significant higher texture and overall acceptability scores than others. Among the higher moisture content formulations, MSP had significantly better texture and overall acceptability scores than positive control (Table 1). Sensory scores indicated that nuggets with additional water were very tender and also reflected by lower hardness score compared to that of others. Though the aroma and ûavour are probably the most important attributes that adversely inûuence the sensory properties of comminuted meat products extended with non-meat protein additives, incorporation of MSP in this experiment does not impart any off flavour. However, Singh *et al.* (2002) reported that addition of texturized soy protein significantly reduced the acceptability of goat meat patties in a dose dependent manner.

Ultra structural studies: The scanning electron microscopy revealed denser matrix (more protein aggregation) with less number of large fat globule in the negative control group and denser matrix with more number of smaller fat globules in batters where meat protein substituted with modified soy proteins (Figure 1). Similar observations were also reported by Van den Hoven (1987) and Youssef and Barbut (2011).

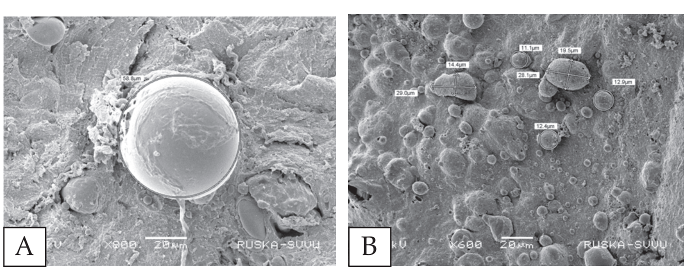

Figure 1: Scanning electron microscope of chicken emulsion of negative control (A) and MSP group (B)

Thiobarbituric acid reactive substances (TBARS) value: The effect of storage on the Thiobarbituric acid reactive substances value of control and treatment groups is presented in Figure 2. The TBA values routinely used as index of lipid oxidation in stored meat products and onset of rancid flavor is usually detected in meat products between TBA values of 0.5 and 2.0 (Raharjo and Safoes 1993). However, TBARS values below 1 mg malonaldehyde/kg product are considered acceptable (Candogan and Kolsarici 2003). The incorporation of MSP and additional water in the formulation does not have significant ($p>0.05$) effect on the TBARS value. However, Das *et al.* (2008) reported that the goat nuggets made with 15% soy granules had signiûcantly lower TBARS value. There was a significant ($P< 0.05$) increase in the TBARS value during the storage period in both control as well as treated samples. A general trend of increase in TBARS during refrigerated and frozen storage of meat and meat products has been reported by many workers (Muthukumar *et al.* 2012). Fat oxidation is responsible for increase in TBARS during refrigerated storage. But the values of TBARS in both control and treated samples remained lower than the acceptable level for rancidity (1.0 mg/kg) and oû-ûavour was not detected by the taste panelists during 20 days of storage.

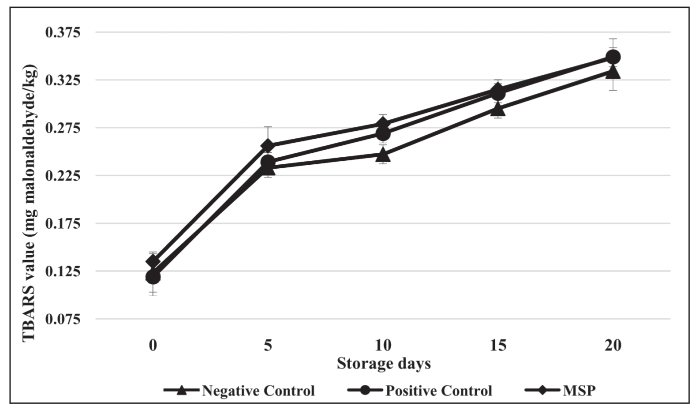

Figure 2: Thiobarbituric acid reactive substances (TBARS) value of chicken nuggets incorporated with modified soy protein during refrigerated storage

Microbiological quality: There was no difference ($p>0.05$) in total plate count and pschyrotrophic count among the control and treated group due to incorporation of MSP and additional water (Table 2). The microbiological stability of low-fat meat products during storage has been studied because the shelf life of these products can be lowered due to their increased water content, which favors microbial growth (Colmenero 2000). During the storage period, the TPC and PTC increased significantly ($p<0.05$) in the control as well as treated samples. However, TPC of nuggets in both control as well as treated samples had not exceeded the permissible level of microbial standards (log 106cfu/g of sample) in cooked meat products as reported by Jay (1996) upto 20 days of storage. Similarly, the psychrotrophic counts were increased significantly ($p<0.05$) between storage periods, however, the counts did not exceed the limit of 4.6 log cfu/g (Cremer and Chipley 1977) in both the control as well as treated samples, suggesting that the nuggets were kept well without microbial spoilage up to 20 days of refrigerated storage.

Freeze-thaw loss: The percentage of freeze thaw loss for negative control, positive control and MSP were 1.76, 2.8 and 1.83,

Table 2: Total viable counts and pschyrotrophic counts (log cfu/g) of chicken nuggets incorporated with modified soy protein during refrigerated storage

Parameter	Treatment groups	Storage period (Days)Overall mean (Storage period)					
		0	5	10	15	20	Overall mean (Treatment)
Total viable counts (log cfu/g)	Negative Control	2.86±0.13[a]	2.87±0.14[a]	3.32±0.15[b]	3.51±0.15[b]	3.58±0.15[b]	3.23±0.08
	Positive Control	2.86±0.13[a]	2.91±0.16[a]	3.36±0.16[b]	3.43±0.16[b]	3.48±0.15[b]	3.21±0.08
	MSP	3.04±0.16[a]	3.09±0.16[a]	3.19±0.15[a]	3.50±0.19[b]	3.59±0.13[b]	3.28±0.08
Overall mean (Treatment)		2.92±0.08[A]	2.96±0.09[A]	3.29±0.09[B]	3.48±0.09[BC]	3.55±0.08[C]	
Pschyrotrophiccounts (log cfu/g)	Negative Control	2.81±0.16[a]	2.79±0.16[a]	3.04±0.26[a]	3.40±0.15[ab]	3.60±0.12[b]	3.13±0.09
	Positive Control	3.00±0.18[ab]	2.94 ±0.14[a]	3.14±0.13[ab]	3.31±0.25[ab]	3.50±0.14[b]	3.18±0.08
	MSP	3.18±0.16[ab]	2.90±0.22[a]	3.32±0.15[ab]	3.30±0.30[ab]	3.74±0.15[b]	3.29±0.10
Overall mean (Treatment)		2.99± 0.10[A]	2.88±0.10[A]	3.17±0.11[AB]	3.34±0.14[BC]	3.61±0.08[C]	

Means ±SE with different superscripts in a row differ significantly ($p<0.05$) ($n=4$).

respectively. The positive control showed significantly ($p<0.05$) higher freeze thaw loss among the products. This is in accordance with Colmenero *et al*. (1996) that freezing-thawing of emulsion meat products has been found to cause loss of quality especially binding properties, the extent of which is dependent on water content of the product. The lower loss noticed in the MSP might be due to soy proteins, which act as an emulsifier and fat encapsulating agent by supplementing myosin and actomyosin and thereby prevent fat and water separation while subjecting to external pressure (Reichet 1991).

CONCLUSION

The functional and sensory attributes of chicken nuggets incorporated with modified soy protein (MSP) as a non-meat ingredient to replace high priced meat is comparable with the nuggets made with higher proportion of meat. Further, the nuggets incorporated with MSP had no beany flavor. There was no significant difference ($p>0.05$) in pH, expressible water, freeze thaw loss, proximate composition and cooking yield between nuggets made with MSP and higher water content and the negative control group made with higher proportion of meat and lesser water. However, replacement of meat with water resulted in lighter coloured tender nuggets. Further, the incorporation of MSP and additional water did not make any significant change in the storage stability of nuggets compared with negative control group. Hence, the MSP has great potential to be used as a meat replacer for the emulsion based meat products with good acceptability and cost benefit.

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