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Efficacy of different variants of corn flour (*Zea mays*) and peanut flour (*Arachis hypogea*) on quality characteristics of designer low sodium fish (*Pangasius pangasius*) balls

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Abstract: The present study was undertaken to analyze the effect of incorporation of different combinations of corn flour (*Zea mays*) and peanut flour (*Arachis hypogea*) on quality characteristics of low sodium fish (*Pangasius pangasius*) balls. Replacement of 50% sodium chloride by 40% KCl, 30% citric acid and 30% sucrose was optimum for preparation of low sodium fish balls. Three different levels of corn flour with peanut flour viz. 25:75, 50:50 and 75:25 were incorporated for preparation of low sodium fish balls and were compared with fish balls prepared by incorporating 10% refined wheat flour (control). On the basis of physico-chemical and sensory evaluation, emulsion stability, cooking yield and overall acceptability were significantly ($p<0.05$) found to be higher with incorporation @ 75% corn flour and 25% peanut flour for preparation of low sodium fish balls. The FFA, TBA and all microbiological profile were found to be significantly ($p<0.05$) increasing with increase in days of storage. The pH, cooking yield, FFA, TBA, total plate count, pychrotropic count, yeast and mould count, overall acceptability were found to be 5.74 ±0.14, 87.06 ± 0.43, 0.36 ± 0.01, 0.72 ± 0.19, 4.43 ± 0.12,3.74 ± 0.2 ,2.60 ± 0.2 ,7.09±0.09 respectively on 21st day of refrigeration storage. The prepared designer low sodium fish balls were found to be fit for human consumption till 21st day of refrigeration storage (4±1°C).

Keywords: *Arachis hypogea,* Low sodium fish balls, Quality attributes, Storage quality, *Zea mays*

INTRODUCTION

Today, consumers concern about dietary sodium intake and its direct correlation with increased risks of hypertension and cardiovascular diseases have motivated meat food researchers to develop newer processing technique to reduce sodium content in meat products (Ruusunen and Puolanne, 2005; Doyle and Glass, 2010). Common salt is the major contributor of sodium in diet and also a vital ingredient in the development of meat products especially restructured and reformed meat products like steaks, roll, meatballs, nuggets, patties etc (Colmenero *et al*., 2005; Desmond, 2006). Salt brings about conformational changes in myofibrillar proteins, which ultimately enhance the ability of protein to bind water (Guardia *et al*., 2012). Reduction in salt content in such products will therefore, inevitably result in not just reduction of saltiness perception but also loss of desirable properties like water binding, tenderness and juiciness(Colmenero *et al*., 2005, Ganie *et al*., 2016). Strategies to reduce common salt in meat products include the use of salt replacers like potassium chloride, magnesium chloride, calcium chloride etc. potassium chloride (KCl) is the most common salt substitute used in low salt meat products, but at higher concentrations can confer a bitter taste to the product (Doyle and Glass, 2010; Horita *et al*., 2011; Canto *et al*., 2014). Addition of carbohydrates like starch flours and non meat proteins can compensate for losses owing to reduction of salt in meat products (Verma and Banerjee, 2012). Several studies have reported that whey protein can improve water binding, cooking yield and texture of meat products (Ulu, 2004; Pietrasik *et al*., 2007; Szerman *et al*., 2012) and thus, can be utilized in developing low salt meat products. Legume flours like black gram flour reportedly increase moisture retention and yield in meat products (Modi *et al*., 2003; Serdaroglu *et al*., 2005). Although a number of studies have focused on the development of low salt meat products using various salt replacers and extenders, but not examined the simultaneous effect salt replacement and incorporation of non meat proteins and binders (Guardia *et al*., 2008; Askin and Kilic, 2009; Lopez-Lopez *et al*., 2010; Horita *et al*., 2011; Szerman *et al*., 2012). Researches undertaking the development of low salt restructured meat products are scarce (Cofrades *et al*., 2011; Canto *et al*., 2014) and studies employing statistical analysis to investigate the interaction of salt and salt replacer in meat products are even lesser (Pietrasik *et al*., 2007; Saricoban *et al*., 2009). Fish, in comparison to other meat types viz. mutton,

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chevon and poultry, has several important nutritional characteristics in its favour especially PUFA content, as well as certain health benefits-principally; that it is low in cholesterol, high in good-quality protein, and it contains average of 16g unsaturated fatty acids, including the omega-3 type per 100 gm, which are reported to help prevent several human illnesses (Heinz and Hautzinger, 2007; Luckose *et al*., 2015).Therefore, the present study was aimed at standardization and optimization of processing conditions for the preparation of corn flour (*Zea mays*) and peanut flour (*Arachis hypogea*) fortified designer low sodium fish (*Pangasius pangasius*) balls and its quality characteristics as well as storage quality analysis at refrigeration temperature.

MATERIALS AND METHODS

Sources of materials: Spineless meat of *Pangasius pangasius* fish belonging to Pangasidae family was used for preparation of low sodium fish balls. All the chemicals used were of analytical grade and were obtained from standard firms (Qualigens, CDH, Hi-Media etc.). Low-density polyethylene film was used packaging during storage study at refrigeration temperature.

Analytical techniques: The pH of cooked nuggets sample was measured by the method of Keller *et al.* (1974). The pH was recorded by digital pH meter (Systronics Digital pH meter 802, Serial No. 603), The proximate composition moisture, crude protein, crude fats, emulsion stability and cooking yield in both control and treated samples were done as per standard procedures. (AOAC, 2000), emulsion stability was calculated as per Townsend *et al.* (1968), Thio Barbituric Acid (TBA) was determined using method of Witte *et al.* (1970), Free Fatty Acid (FFA) was determined by method described by Koniecko (1979), Total plate count, Psychrotrophic count, Coliform count and Yeast and Mould count in the sample were determined by method described by APHA (1984). Readymade media (Hi-Media) were used for the analysis. Sensory Evaluation was carried for various attributes viz. color and appearance, flavor, juiciness, texture and overall acceptability by a panel of trained members composed of scientists based on a eight-point Hedonic scale, wherein 8 denoted "extremely desirable" and 1 denoted "extremely undesirable" (Seman *et al*., 1987). Seven members of the panel replicated the experiment thrice (n = 21). Panellists were comfortably seated in a noise and odour free room which was suitably illuminated. Coded samples for sensory evaluation were prepared. Results were analyzed statistically for analysis of variance in one way as well as two way and least significant ($p<0.05$) difference tests as per (Snedecor and Cochran, 1980).

Methodology of preparation of fish balls: Methodology for the preparation of fish balls is described in following steps i.e. preparation of emulsion, filling in manual extruder, cooking of fish balls.

Preparation of emulsion: Several preliminary trials were conducted to standardize the formulation for preparation of emulsion for fish balls. Fish meat was cut into smaller chunks and minced in a Sirman mincer (MOD-TC 32 R10 U.P. INOX, Marsango, Italy) with 6mm plate. The common salt, refined wheat flour (maida), sodium glutamate, sodium tripolyphosphate, spice mixture and condiment mixture were added to weighed meat according to formulation. Meat emulsion for fish was prepared in Sirman Bowl Chopper [MOD C 15 2.8G 4.0 HP, Marsango, Italy]. Minced meat was chopped with all the curing ingredients for 1.5 minute. Water in the form of crushed ice was added and blending continued for 1 minute. This was followed by addition of spice mixture, condiments and other ingredients and again mixed for 1.5 to 2 minutes to get the desired emulsion. Adequate care was taken to keep the end point temperature below 18^0C by preparing the emulsion in cool hours of morning, by addition of meat and other ingredients in chilled/partially thawed form and by addition of crushed ice or ice water. Low sodium fish balls can be efficiently prepared by substituting 50% NaCl by 20% KCl, 15% Sucrose and 15% citric acid. Formulation (%) of fish balls were fish meat 59.7, Added water (ice flakes) 10, Condiment mixture 5, vegetable oil 9, refined wheat flour 10, Spice mixture 2.5, Salt + KCl+ citric acid+sucrose1 +0.5+0.25+0.25 (low sodium standardized salt combi-

Table 1. Effect of replacement of NaCl by other salts on pH, emulsion stability and proximate composition of low sodium fish balls.

Parameter	Level of NaCl and other salts			
	100:0	50:50	25:75	75:25
		KCl 40% +Citric acid 30% + Sucrose 30%	KCl 50% + Citric acid 25% + Sucrose 25%	KCl 60% + Citric acid 20% + Sucrose 20%
Moisture (%)	57.96±0.41[a]	59.52±0.69[a]	59.70±1.13[a]	58.55±0.97[a]
Protein (%)	20.05±0.87[a]	20.26±0.67[a]	20.94±0.55[a]	19.60±0.59[a]
Fat (%)	11.10±0.20[a]	11.11±0.34[a]	11.38±0.49[a]	10.92±0.36[a]
Ash (%)	3.94±0.22[a]	4.32±0.42[a]	4.30±0.48[a]	4.33±0.39[a]
Cooking yield (%)	86.59±0.48[c]	86.46±0.51[c]	80.62±0.40[a]	83.62±0.55[b]
pH	6.29±0.17[a]	6.30±0.20[a]	6.22±0.21[a]	6.47±0.20[a]
Emulsion stability (%)	85.97±0.45[c]	86.15±0.29[c]	81.06±0.37[a]	83.57±0.41[b]

[*]Mean ±SE with different superscripts in a row differs significantly ($P<0.05$). n = 6 for each treatment

Table 2. Effect of different combinations of corn flour and peanut flour on pH, emulsion stability, cooking yield and proximate composition of low sodium fish balls.

Parameters	Levels of corn flour and peanut flour (%)			
	0	25:75	50:50	75:25
pH	6.33 ± 0.17[a]	6.15 ± 0.28[a]	6.39 ± 0.20[a]	6.21 ± 0.27[a]
Emulsion stability (%)	82.61±0.60[a]	84.78±0.44[b]	84.93±0.33[bc]	86.16±0.21[c]
Moisture (%)	57.01 ± 0.75[a]	58.09 ± 0.92[a]	57.97± 1.05[a]	57.87± 1.01[a]
Protein (%)	20.47± 0.62[a]	22.32± 0.75[ab]	20.43± 0.76[ab]	19.36 ± 0.77[b]
Fat (%)	10.87 ± 0.32[a]	10.52 ± 0.35[a]	10.79 ± 0.31[a]	10.81 ± 0.29[a]
Ash (%)	3.93 ± 0.22[a]	4.64 ±0.32[b]	4.47 ±0.40[b]	4.36 ±0.32[b]
Cooking yield(%)	82.71 ± 0.54[a]	84.58 ± 0.32[b]	84.94 ± 0.62[b]	87.06 ± 0.43[c]

[*]Mean ±SE with different superscripts in a row differs significantly ($P<0.05$). n = 6 for each treatment

Table 3. Effect of different combinations of corn flour and peanut flour on sensory attributes of low sodium fish balls.

Sensory attributes	Levels of corn flour and peanut flour (%)			
	0	25:75	50:50	75:25
Appearance	6.25±0.309[ab]	6.09±0.166[a]	6.11±0.265[a]	6.92±0.160[c]
Flavour	6.12±0.267[a]	5.83±0.234[a]	5.89±0.293[a]	6.86±0.183[b]
Juiciness	6.31±0.292[ab]	6.01±0.244[a]	5.99±0.240[a]	6.91±0.154[b]
Texture	6.39±0.315[ab]	5.98±0.330[a]	5.88±0.155[a]	6.97±0.126[b]
Overall acceptability	6.11±0.204[a]	5.78±0.189[a]	5.87±0.328[a]	7.09±0.085[b]

[*]Mean ±SE with different superscripts in a row differs significantly ($P<0.05$). Mean values are scores on 8 point descriptive scale where 1- extremely poor and 8- extremely desirable. n = 21 for each treatment.

nation),sodium tripolyphosphate 0.3 monosodium glutamate 0.5 and egg white 1.

Experimental design: The standardization and optimization for preparation of low sodium fish ball using corn flour (*Zea mays*) and peanut flour (*Arachis hypogea*) in the ratio of 25:75, 50:50 and 75:25 along with 100 ratio refined wheat flour as control was eperimented. The designer products were analyzed for its proximate, physio-chemical characteristics and sensory characteristics. The best variant of designer product selected was analyzed for its storage quality parameters at refrigeration temperature of 4±1°C.

RESULTS AND DISCUSSION

pH, physiochemical and proximate composition: The pH value of fish balls prepared by incorporating 25:75, 50:50 and 75:25 corn flour with peanut flour were comparable with pH value of fish balls prepared with 10% incorporation of refined wheat flour (control). The moisture value of fish balls prepared by incorporating 10% refined wheat flour (control) was lowest but was comparable to moisture value of fish balls prepared by incorporating 25:75, 50:50 and 75:25 corn flour with peanut flour. This might be possibly due to decreasing water absorbing property of all flours used. The results were in contrast to the findings of (Luckose *et al*., 2015) who reported that during heat processing of legume flours gelatinization of the carbohydrates and swelling of the crude fiber occur which leads to increased water absorption. The protein content was highest for fish balls prepared by incorporating 25:75 corn flour with peanut flour and varied significantly from protein value of fish balls prepared by incorporating 50:50 and 75:25 corn flour with peanut flour as well as fish balls prepared by incorporating 10% refined wheat flour (control). The fat content was highest for fish balls prepared by incorporating 75:25 corn flour with peanut flour although it was comparable to fat content of fish balls prepared by incorporating 25:75 and 50:50 corn flour and peanut flour as well as fish balls prepared by incorporating 10% refined wheat flour (control). Decrease in the fat content could be attributed to defatted nature of corn flour used in the formulation of fish balls. The ash content was highest for fish balls prepared by incorporating 25:75 corn flour and peanut flour and it varied significantly from ash content of fish balls prepared by incorporating 10% refined wheat flour (control) though it was comparable to ash content of fish balls prepared by incorporating 50:50 and 75:25 corn flour and peanut flour. The lower ash content in corn flour and peanut flour incorporated fish balls might be due to lower ash content of flours. This trend of reduction fat and ash contents in meat products using corn flour and peanut flour was also found by Yang *et al.* (2007), who used different flours in the formulation of duck sausages. The emulsion stability of fish balls prepared by incorporating 25:75, 50:50 and 75:25 corn flour with peanut flour varied significantly ($p<0.05$) from emulsion stability of fish balls prepared by incorporating 10% refined wheat flour. The emulsion stability of fish balls prepared by incorporating 75:25 corn flour with peanut flour was highest and varied significantly from emulsion stability of fish balls prepared by incorporating 25:75 and 50:50 corn flour with peanut flour as well as fish balls prepared by incorporating 10% refined wheat flour (control). The increase in emulsion stability with increase of extension level could be attributed to ge-

Table 4. Effect of refrigerated temperature storage on physicochemical characteristics of aerobically packaged low sodium fish balls incorporated with different flour combinations.

Treatment	Storage period in days			
	0	7	14	21
pH				
RWF	6.07 ± 0.18^{Aa}	6.06 ± 0.23^{Aa}	5.78 ± 0.17^{Aa}	5.99 ± 0.16^{Aa}
CP	6.04 ±0.18^{Aa}	5.88 ±0.14^{Aa}	5.62 ± 0.19^{Aa}	5.74 ±0.14^{Aa}
TBARS(mg malonaldehyde/kg)				
RWF	0.40 ± 0.21^{Aa}	0.45 ± 0.24^{Aa}	0.64 ± 0.20^{Ab}	0.71 ± 0.17^{Ac}
CP	0.40 ± 0.16^{Aa}	0.52 ± 0.20^{Bb}	0.62 ± 0.23^{Ac}	0.72 ± 0.19^{Ad}
FFA(% oleic acid)				
RWF	0.090± 0.007^{Aa}	0.131 ± 0.003^{Ab}	0.233 ± 0.005^{Ac}	0.388 ± 0.009^{Ad}
CP	0.101 ± 0.007^{ABa}	0.125 ± 0.008^{Aa}	0.231 ± 0.007^{Ab}	0.358 ± 0.010^{Ac}
Total plate count(log cfu/g)				
RWF	2.37 ± 0.05^{Aa}	2.62± 0.06^{Aa}	3.31 ± 0.11^{Ab}	4.17 ±0.21^{Ac}
CP	2.33 ± 0.05^{Aa}	2.70 ± 0.08^{ABb}	3.32 ± 0.06^{Ac}	4.43 ± 0.12^{Ad}
Psychrotrophic count (log cfu/g)				
RWF	ND	ND	2.24 ± 0.04^{Ab}	3.35 ± 0.26^{Ac}
CP	ND	ND	2.73 ± 0.29^{Ab}	3.74 ± 0.26^{Ac}
Coliform count (log cfu/g)				
RWF	ND	ND	ND	2.63 ± 0.13^{Ab}
CP	ND	ND	ND	3.33 ± 0.23^{Bb}
Yeast and Mould count (log cfu/g)				
RWF	ND	ND	ND	2.40 ± 0.22^{Ab}
CP	ND	ND	ND	2.60 ± 0.23^{Ab}

Mean ± SE with different superscripts in a row wise (lower case alphabet) and column wise (upper case alphabet) differ significantly ($p<0.05$).RWF=Refined wheat flour, CP=Corn and peanut flour.(n=6).

latinizing property of increasing starch component on heating, which stabilized the emulsion (Canto, 2014). Gelatinization of all types of this starch improves the binding properties of meat proteins (Purnomo and Rahardiyan, 2008). The cooking yield was highest for fish balls prepared by incorporating 75:25 corn flour and peanut flour and it varied significantly from cooking yield of fish balls prepared by incorporating 25:75 and 50:50 corn flour and peanut flour as well as fish balls prepared by incorporating 10% refined wheat flour (control). Horita (2011) reported that cooking yield increased in low sodium reduced-fat mortadella. Similar results were obtained by (Pietrasik *et al*., 2007) who observed increase in cooking yield in while studying effect of non meat protein on hydration and textural properties of pork meat.

Sensory attributes: The mean values of sensory scores of fish balls prepared by incorporating different combinations of corn flour with peanut flour are presented in table 3.Appearance, flavour, juiciness, texture and overall acceptability was highest for fish balls prepared by incorporating 75:25 corn flour and peanut flour and varied significantly from appearance, flavour, juiciness, texture and overall acceptability of fish balls prepared by incorporating 25:75 and 50:50 corn flour and peanut flour as well as fish balls prepared by incorporating 100 refined wheat flour (control). Similar results were obtained by Huda *et al.* (2012) for colour who found that lighter colour of the fish snacks was preferred by the sensory panellists. Increased puffiness due to increase in corn flour level could be the cause of decrease in the texture scores of fish balls. The present findings were in accordance with the findings obtained by Boonyasirikool and Charunuch (2000), who found increased corn flour levels, increased the expansion of corn extruded snacks. Similar findings were reported by Ross and Smith (2006) while working on lipid oxidation in muscle foods and Gelabert *et al.* (2007) in low sodium fermented sausages. The overall acceptability of fish balls was significantly highest at 75:25 level of inclusion of corn flour with peanut in the formulation as compared to control and other treatments. Hence, 75:25 level of inclusion of corn flour with peanut was found optimum for the preparation of fish snacks.

Based on the results of cooking yield, emulsion stability and sensory evaluation, the fish balls prepared by incorporating 75% corn flour with 25% peanut flour were selected for storage studies and were compared with fish balls prepared by incorporating 10% refined wheat flour. They were aerobically packaged in low density polyethylene pouches and were analyzed at regular intervals of 0, 7, 14 and 21 days kept at refrigerated storage. The physico-chemical characteristics, microbiological characteristics are presented in table 4.

Storage quality (Physico-chemical characters)

pH: The different flour combinations selected for storage study had a significant effect on pH value. The effect of storage on pH followed a decreasing trend at progressive storage intervals. There was a significant decrease in pH of most of the variants of fish balls; however the mean pH value was comparable among low sodium fish balls prepared by incorporating different flour combinations. The decrease in pH might be attributed to the availability of more readily utilizable carbohydrate molecules by the microbes and thereby formation of lactic acid. It is an established fact that a decrease in pH is usually attributed to the metabolic activity of bacteria (Luckose *et al*., 2015. Garcia *et al.* (2002) also observed a similar decrease in pH of low fat fermented dry sausages prepared with cereals and fruit fibres. Devatkal *et al*. (2008) reported that pH decreased significantly with the increase of storage periods but non-significant changes were observed in pH among different loaves. However the results were in contrast to those obtained by Bingol *et al.* (2010) on low fat Turkish type meat balls.

Free fatty acid (FFA): The FFA followed a significant increasing trend from day 0 to 21 in low sodium fish balls prepared by incorporating different flour combinations. FFA content increased non-significantly up to day 7 and significantly on subsequent intervals i.e, day 14 and 21. The significant increase in FFA content of meat products during storage might be due to growth of lipolytic microorganisms as lipids being most vulnerable for easy deterioration (Das *et al*., 2008).

Thiobarbituric acid (TBA) value: The TBA followed a non significant increasing trend from day 0 to day 7 and significant increasing trend on subsequent intervals i.e, day 14 and day 21st.The increase in TBARS values on storage might be attributed to oxygen permeability of packaging material that led to lipid oxidation. A general trend of increased TBARS value during refrigerated and frozen storage of meat and meat products had been reported by Ratanatriwong *et al.* (2011) and Singh *et al.* (2011) in beef and chicken meat respectively stored at room temperature.

Microbiological profile: The mean microbiological characteristics of fish balls prepared by incorporating 10% refined wheat flour (control), 50:50 rice flour with kidney bean flour, 75:25 corn flour with peanut flour and 25:75 barley flour with pea flour. The Total plate count increased significantly from day 0 to day 21 in fish balls prepared by incorporating 10% refined wheat flour as well as fish balls prepared by incorporating 75:25 corn flour with peanut flour. Similar results were obtained by Siddique *et al.* (2013) on quality and shelf life of fish sausage and fish balls prepared from Bombay duck. Chand (2011) also reported an increase in TPC at each storage interval in meat snacks. The psychrotrophs were not detected on day 0 and day 7of storage in any of the fish balls prepared by incorporating different flour combinations but was observed on day 14 and day 21 in all products. Absence of psychrotrophs upto day 7 might be probably due to high temperature used for frying fish balls which crossed the thermal death point of bacteria and sudden appearance on 14th day may be as a result of contamination. A detectable count on day 14 onwards while nil on preceding observations might be attributed to the fact that bacteria generally need some lag phase before active multiplication is initiated. The coliforms were not detected in any of the product prepared by incorporating different flour combinations upto day 14 but were observed in all the products on day 21st of storage. Appearance of coliforms later onwards could be because of contamination. Serdaroglu *et al.* (2004) had reported similar results while working on quality of low-fat meatballs containing Legume flours as extenders. The yeast and moulds were not detected in any of the product prepared by incorporating different flour combinations upto day 14 but were observed in all the products on day 21st of storage. The detection of yeast and molds on day 21st possibly could be due to contamination which might have occurred during post processing. Askin and Kilic (2009) observed that yeast and mold appeared during the last day of storage while study in effect of microbial transglutaminase, sodium caseinate and non-fat dry milk on quality of salt-free, low fat turkey kebab.

Conclusion

Fish balls prepared by incorporating 75:25 corn flour with peanut flour was optimum for preparation of fish balls. Replacement of 50% sodium chloride by 40% KCl, 30% citric acid and 30% sucrose was optimum for preparation of low sodium fish balls. Fish balls prepared by incorporating best combinations of flours were having comparable emulsion stability (86.15±0.2), cooking yield (87.06 ± 0.4). The pH, cooking yield, FFA, TBA, total plate count, pychrotropic count, yeast and mould count, overall acceptability were found to be 5.74 ±0.14, 87.06 ± 0.43, 0.36 ± 0.01, 0.72 ± 0.19, 4.43 ± 0.12,3.74 ± 0.2 ,2.60 ± 0.2 ,7.09±0.09 respectively on 21st day of refrigeration storage. The novelty of developed product was low in sodium having improved sensory scores and enhanced shelf-life.

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