DEVELOPMENT AND QUALITY ATTRIBUTES OF CHICKEN MEAT POWDER AND FISH PROTEIN ISOLATE INCORPORATED FUNCTIONAL BREAD

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ABSTRACT

To increase the functionality and protein content, standardization of incorporation levels of chicken meat powder (CMP) and fish protein isolate (FPI) in whole wheat bread was done at varying levels. Three different levels each of CMP (20%, 25% and 30%) and FPI (1%, 3% and 5%) were tried, replacing whole wheat flour in standardized formulation, yielding nine treatments. It was observed that the incorporation of CMP and FPI had a significant impact ($p \le 0.05$) on proximate composition, textural profile and instrumental colour values. The moisture (%) in final product displayed a significant decrease ($p \le 0.05$) with the addition of CMP and FPI. Conversely, protein (%), fat (%) and ash (%) values exhibited a significant increase ($p \le 0.05$) with the substitution of CMP and FPI in whole wheat flour-based bread. A significant increase ($p \le 0.05$) in most of the textural profile parameters was observed with the inclusion of both CMP and FPI. Furthermore, redness and yellowness values of treated products significantly increased ($p \le 0.05$) as compared to control. On sensory profile analysis, the colour of the crust, crumb and flavor exhibited a significant increase ($p \le 0.05$) with the increasing level of incorporation till T-6, while porosity and texture scores decreased significantly ($p \le 0.05$). Based on proximate, colour, textural and sensory attributes, incorporation levels of 25 % CMP and 5% FPI was found to be optimum replacing wheat flour in formulation for the development of functional meat bread, which could provide health benefits beyond basic nutrition.

Keywords: Chicken meat powder, Fish protein isolate, Functional, Meat bread, Proximate, Textural

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The changing lifestyles and globalization has affected dietary patterns with more insistence on convenient and ready-to-eat foods. Many of these products are not nutritionally balanced and pose greater health risks to consumers, owing to sedentary lifestyles (Mehta et al., 2015a; Mehta et al., 2013). Regular food items like bread are excellent convenient foods that act as base materials for various recipes but lack essential elements like quality proteins (Umaraw et al., 2021). They are an important source of carbohydrates and dietary fiber and the primary component is starch followed by gluten-forming protein. This gluten matrix is essential for forming cohesive dough necessary for generating a porous structure in bread. Consumers are becoming aware of their health and are interested in consuming balanced and nutritious products. Incorporation of alternate protein sources in bread can be instrumental in enhancing functionality and improving amino acid balance (Prieto-Vazquez del Mercado et al., 2022). Thus, the enrichment of bread with animal protein is a novel area of research that could provide a product which is self-sufficient and could meet the key nutritional needs. Chicken meat holds significant importance as a widely consumed and highly versatile protein source in the global food industry. It is valued for its nutritional profile and affordability, making it the most sought-after meat

(Mehta et al., 2015b). High protein along with a relatively lean structure is the characteristic feature that makes it an ideal choice for value-added product development. In addition, utilization of fish proteins may have additional benefits in terms of protein enrichment alongside improvements in textural and processing attributes. Proteins from the aquatic sources improves cardiovascular health and other associated health conditions. The inclusion of fish protein can be a good alternative to improve the overall nutritional quality and also increase fish consumption amongst masses (Kaur et al., 2024). Thus, the present study envisioned the development of functional meat bread incorporated with chicken and fish proteins and its quality evaluation to formulate a functional product that could provide key elements for maintaining human health.

MATERIALS AND METHODS

Raw Materials

Spent hens were procured from the University Poultry Farm of Guru Angad Dev Veterinary and Animal Sciences University, Ludhiana. The birds were slaughtered in the university's experimental abattoir and the carcasses were chilled at $4\pm1^{\circ}$ C for 12-18 hours. After deboning and removal of skin, fat and connective tissues, the deboned meat was packaged in LDPE bags and stored at $-18\pm1^{\circ}$ C.

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Frozen meat was thawed overnight at $4\pm1^{\circ}$ C before use. The meat was minced using meat mincer (MADO Eskimo Mew 714, Germany), with each batch consisting of 4 kg of meat. The minced meat was then dried in an industrial tray drier (Macro Scientific Works MAC-MSW 215) at 68-70° C for 48 hours, resulting in dry flakes. These flakes were ground into a fine powder using a grinder. The chicken meat powder (CMP) was stored in airtight, dark-coloured jars under refrigeration ($4\pm1^{\circ}$ C) to preserve its freshness and quality till further use. Fish protein isolate (FPI) was procured from the Department of Fish Processing and Technology, GADVASU, Ludhiana. Wheat flour, edible oil, multigrain flour, sugar, salt and other ingredients were purchased from the local market.

Preparation of functional meat-based bread

Three different levels of CMP (20%, 25% and 30%) and FPI (1%, 3% and 5%) were incorporated in the prestandardized formulation of bread (Table 1), resulting in nine treatments. The multigrain flour (MGF) used in formulation consisted of sorghum, ragi, maize, oats and soya in equal proportions. The flow diagram for the preparation of bread is mentioned below:

Mix wheat flour, MGF, CMP, FPI and Salt Addsugar, Yeast, and 20-30 ml Lukewarm Water for Yeast activation Ţ Incubate at 37°C for 15-30 Minutes ↓ Slowly add oil to the batter Mix slowly for 2 Minutes and add 5 ml Water Kneading and Sheeting to prepare Dough (2-3 Min) Cover dough with muslin cloth (37°C,45 Min): 1st Proofing Ţ Softly sheeting and rolling Incubate (37°C, 45 Min): 2nd Proofing Bake in the oven for 30 minutes Cooling, cutting, packaging and storage at 4±1°C Evaluation and sample analysis

Proximate Composition

Moisture, protein, fat, and ash content of the product was estimated following the standard procedures of AOAC (2000).

Instrumental texture and colour profile analysis

Instrumental texture profile analysis was carried out using a texture analyzer (TMS-PRO, Food Technology Corporation, USA). Asample of $1 \text{ cm} \times 1 \text{ cm} \times 1 \text{ cm}$ was used with a pre-test speed of 30 mm/sec, a post-test speed of 100 mm/sec, and a test speed of 100 mm/sec. A load cell with a 2500 N capacity was used to apply the double compression cycle. For colour profile analysis, CR-400 Konica Chromameter (Konica Minolta, Japan) was used and the results were recorded as L*, a* and b* values.

Sensory evaluation

A panel of seven experienced individuals (scientific staff and students) from the department evaluated the samples for colour and appearance of crust and crumb, porosity, flavour, texture, meat flavor intensity and overall acceptability using an 8-point descriptive scale by Keeton (1983), where 8= extremely desirable and 1= extremely undesirable.

Statistical Analysis

Using IBM SPSS Statistics-20.0 software, the data was statistically analyzed following one way ANOVA at 5% level ($p \le 0.05$) and evaluated with Duncan's Multiple Range Test (DMRT) (Snedecor and Cochran, 1994). For each parameter, duplicate samples were drawn and the experiments were carried out in triplicate, yielding six observations (n=6). For sensory evaluation, a panel of seven judges conducted analysis and the experiment was replicated thrice, thus yielding 21 observations (n=21). The analyzed results were presented as Mean \pm S.E.

RESULTS AND DISCUSSION

Proximate composition

The proximate composition of the functional meat bread incorporated with different levels of CMP and FPI is presented under Figure 1. Perusal to this figure, it was observed that the moisture content of the control and treatment breads ranged from 2.5% to 4.79%. The highest moisture content was observed in T-9, whereas control had the lowest value of moisture (%). Combination of CMP and FPI at a higher level might have contributed to a looser bread structure, allowing for higher moisture retention (Umaraw et al., 2021). The moisture content is also dependent on many other factors viz. baking time, temperature and storage conditions. Also, these factors can affect the evaporation of moisture during baking and postbaking storage, ultimately impacting the moisture content of the bread samples. Similar observations have also been made by Giannou et al. (2003). The protein content of the

Ingredients (%w/w)	Control (C)	T-1	T-2	T-3	T-4	T-5	T-6	T-7	T-8	T-9
Wheat Flour	92	58	56	54	53	51	49	39	37	35
Yeast	3	3	3	3	3	3	3	3	3	3
Baking Powder	-	3	3	3	3	3	3	3	3	3
Sugar	3	3	3	3	3	3	3	3	3	3
Salt	1	1	1	1	1	1	1	1	1	1
Water (ml)	100	100	100	100	100	100	100	100	100	100
Fat/Oil (ml)	1	1	1	1	1	1	1	1	1	1
CMP	-	20	20	20	25	25	25	30	30	30
FPI	-	1	3	5	1	3	5	1	3	5
MGF	-	10	10	10	10	10	10	10	10	10

CMP: Chicken Meat Powder; **FPI:** Fish Protein isolate; **MGF:** Multigrain Flour, T-1 (20% CMP+1% FPI), T-2 (20% CMP+3% FPI), T-3 (20% CMP+5% FPI), T-4 (25% CMP+1% FPI), T-5 (25% CMP+3% FPI), T-6 (25% CMP+5% FPI), T-7 (30% CMP+1% FPI), T-8 (30% CMP+3% FPI) and T-9 (30% CMP+5% FPI)

bread ranged from 21.51% to 28.78%. Out of all the treatments, the highest value for protein (%) was observed in T-9, which consisted of highest quantity of both CMP and FPI. Irrespective of the levels of incorporation, addition of chicken meat powder and fish protein isolate significantly increased ($p \le 0.05$) the protein content of the all bread samples as both of them are rich sources of protein (Umaraw et al., 2021). Further, the processing conditions, such as mixing, fermentation, and baking, might have also affected protein content. Fat content of control and all the treatments (T-1 to T-9) ranged from 1.51% to 13.09%, with control having lowest values. The highest ($p \le 0.05$) fat content was observed in bread containing 30% CMP and 5% FPI, which could be attributed to fat already present in these sources. The specific ratios of CMP and FPI in the bread formulations can impact the overall fat content. Higher levels of CMP and FPI in the bread formulation may have contributed to increased fat content and both CMP and FPI may have different abilities to absorb and retain fat during flour mixing and baking, leading to variations in the final fat content of the bread samples (Nawaz et al., 2019). The highest ash content was observed in bread containing 25% CMP and 5% FPI (T-6), while the lowest ash content values were observed in bread containing 20% CMP and 1% FPI (T-1). Higher ash content in bakery products is indicative of higher fiber content, which is generally suggested due to positive health impact (Cakmak et al., 2016)

Colour and textural attributes

The colour attributes of control as well as treated products are depicted under Figure 2. Control product had a significantly lighter ($p \le 0.05$) colour than all other treatments and with an increase in level of incorporation of CMP and FPI, a significant decrease ($p \le 0.05$) was observed in the lightness value of bread samples. Consequently, an increase in the redness value of treated products was observed with increasing levels of CMP and FPI. This could be due to increased protein and amino acid content which could have led to enhanced interaction between protein and carbohydrate moiety leading to more browning (Roncolini *et al.*, 2019; Madenci and Bilgicli, 2014). Similar findings have been reported by Umaraw *et al.* (2021). The value for yellowness i.e. b* value significantly increased (p \leq 0.05) with increasing levels of CMP and FPI, which could be attributed to change in O-R potential and non-enzymatic browning reaction during baking (Chinma *et al.*, 2015). Similar findings have been reported by Farouk *et al.* (2018) in red meat incorporated breads.

Texture is an essential attribute in bakery products, particularly breads and out of all the components, hardness of the product holds critical importance. The findings of texture profile analysis for the bread samples are presented in figure 3. It was observed that hardness value was significantly affected (p < 0.05) with the incorporation of CMP and FPI. As compared to control, an increase in hardness in treated samples was observed. It could be due to lower starch gelatinization owing to higher absorption and retention of water by the added proteins (Karimi et al., 2021). Similar findings have been reported by Zhou et al. (2018) on the addition of whey and soy protein in breads made up of wheat flour. Bread cohesiveness indicates the holding of the dough while chewing and with incorporation of CMP and FPI, an increase $(p \le 0.05)$ in cohesiveness value was observed. With the addition of protein sources in bread, the springiness value decreased ($p \le 0.05$) indicating a reduction in the ability to recover after deformation due to compression forces. Millar et al. (2019) reported similar findings in white bread on addition of raw, germinated and toasted pea flour. An increase ($p \le 0.05$) in chewiness value with the addition of CMP and FPI in functional bread was observed. It could be attributed to a lesser amount of gluten in treated samples due to replacement with protein sources, as compared to control. These proteins have a stronger

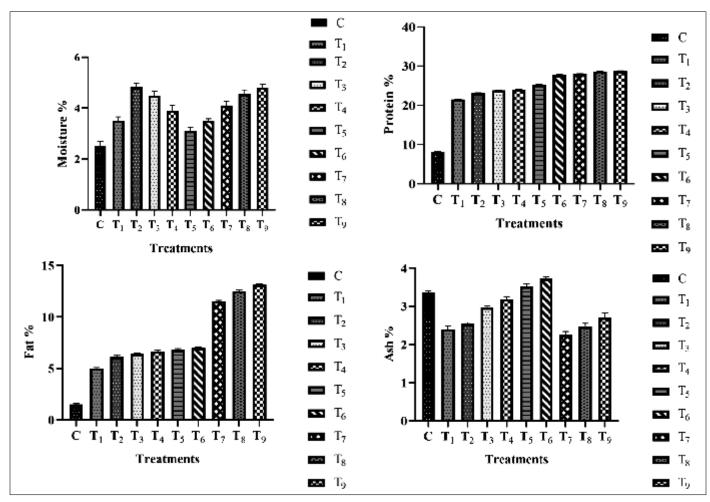


Fig. 1. Proximate Composition (mean ± standard error) of functional meat bread incorporated with different levels of CMP and FPI; n=6. CMP: Chicken Meat Powder; FPI: Fish Protein isolate; T-1 (20% CMP+1% FPI), T-2 (20% CMP+3% FPI), T-3 (20% CMP+5% FPI), T-4 (25% CMP+1% FPI), T-5 (25% CMP+3% FPI), T-6 (25% CMP+5% FPI), T-7 (30% CMP+1% FPI), T-8 (30% CMP+3% FPI) and T-9 (30% CMP+5% FPI)

water binding capacity which might have led to denser structure in bread mass. Similar findings have been reported by Mao *et al.* (2022) who reported higher values of hardness and chewiness in dairy-based high protein steamed breads.

Sensory Analysis

The scores for colour of crumb and crust corresponded to the values of colour profile analysis, wherein the darker colour of the product was accepted by panelists to a certain level i.e. T-6 but beyond 25% of CMP inclusion, the panelists scored the products on the lower side (Fig. 4). A decrease ($p \le 0.05$) in porosity attribute as compared to control was observed in bread incorporated with CMP and FPI. This could be due to the replacement of gluten-rich wheat flour with meat which is devoid of gluten. Further, the absence of fiber due to addition of meat also contributed to decreased porosity. Similar findings have been reported by Cercel *et al.* (2016) and Umaraw *et al.* (2021) in fish protein concentrate incorporated wheat breads and chicken meat incorporated whole wheat bread, respectively. An

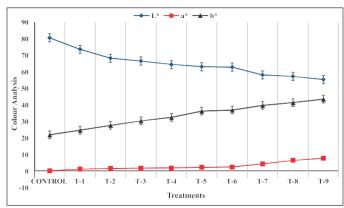
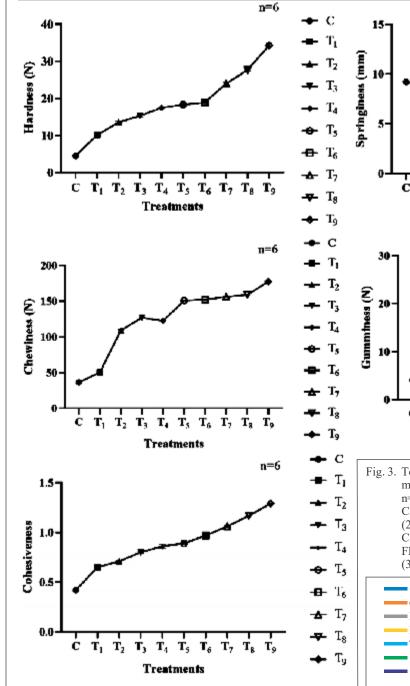


Fig. 2. Colour analysis (mean ± standard error) of functional meat bread incorporated with different levels of CMP and FPI; n=6 CMP: Chicken Meat Powder; FPI: Fish Protein isolate; T-1 (20% CMP+1% FPI), T-2 (20% CMP+3% FPI), T-3 (20% CMP+5% FPI), T-4 (25% CMP+1% FPI), T-5 (25% CMP+3% FPI), T-6 (25% CMP+5% FPI), T-7 (30% CMP+1% FPI), T-8 (30% CMP+3% FPI) and T-9 (30% CMP+5% FPI)

increase in flavor attribute with increasing concentration of CMP was observed till 25% of CMP level, however, at 30% level, a decrease (p ≤ 0.05) in flavor attribute was reported



by sensory panelists. Higher CMP levels may have led to the production of some of the compounds by microbial fermentation during incubation, which might have produced poor flavor. Meinert *et al.* (2016) also reported off odour and bitter taste compound during hydrolysis of bovine heart. With the higher level of inclusion of CMP, the texture scores were rated lower by the sensory panelists. The meat flavor intensity was significantly higher (p≤0.05) in treatments with higher concentrations of CMP due to possible Maillard reaction products. An increase (p≤0.05) in overall acceptability scores of bread with the inclusion of CMP and FPI has been observed till T-6,

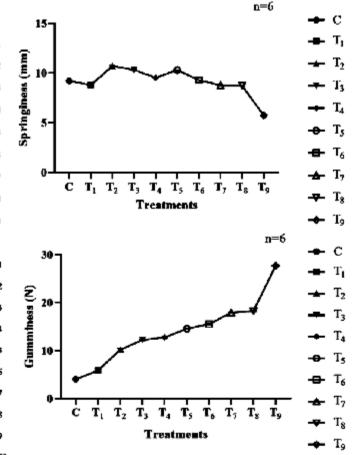


Fig. 3. Texture Profile Analysis (mean ± standard error) of functional meat bread incorporated with different levels of CMP and FPI; n=6

CMP: Chicken Meat Powder; FPI: Fish Protein isolate; T-1 (20% CMP+1% FPI), T-2 (20% CMP+3% FPI), T-3 (20% CMP+5% FPI), T-4 (25% CMP+1% FPI), T-5 (25% CMP+3% FPI), T-6 (25% CMP+5% FPI), T-7 (30% CMP+1% FPI), T-8 (30% CMP+3% FPI) and T-9 (30% CMP+5% FPI)

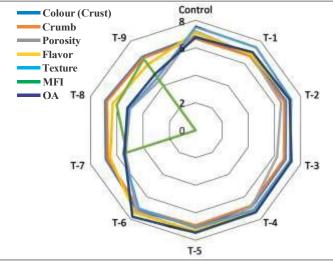


Fig. 4. Effect of different levels of CMP and FPI on sensory profile of functional meat bread; n=21; CMP: Chicken Meat Powder; FPI: Fish Protein isolate; T-1 (20% CMP+1% FPI), T-2 (20% CMP+3% FPI), T-3 (20% CMP+5% FPI), T-4 (25% CMP+1% FPI), T-5 (25% CMP+3% FPI), T-6 (25% CMP+5% FPI), T-7 (30% CMP+1% FPI), T-8 (30% CMP+3% FPI) and T-9 (30% CMP+5% FPI)

thereafter a decrease in scores was observed. It could be due to decrease in other sensory attributes, which might have affected scores for overall acceptability. Khare *et al.* (2015) also found that all the independent sensory variables along with their interaction significantly affected the overall acceptability scores of chicken noodles.

CONCLUSION

Based on the findings of the present study, it can be concluded that the incorporation of chicken meat powder at 25% level and fish protein isolate at 5% level was found to be optimum for development of functional meat bread. The bread had higher protein content, better textural, colour and sensory properties as compared to control, which will be instrumental in providing quality protein to the consumers without affecting any sensory attribute. The study concluded that T-6, consisting of CMP (25%) and FPI (5%) are best among all tried formulations and can be used for the development of functional high-protein breads. These products have a potential to supplement proteins from meat as well as fish which will make them an ideal food capable of maintaining and enhancing human health. Future research can focus on enhancement of keeping quality of protein enriched breads to make them shelf stable for longer period of time.

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