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Optimization of Amaranth Incorporation in Bread Prepared with Combination of Chicken Meat Powder and Whole Wheat Flour

Pramila Umaraw, Geeta Chauhan, S.K. Mendiratta, Akhilesh K. Verma 10.18805/ajdfr.DR-1941

ABSTRACT

Background: Bread is baked product generally prepared with use of refined wheat flour which is deficient in protein and fiber. Addition of Amaranthus flour and chicken meat powder in bread enhanced the protein and fiber content which are essential component of human diet.

Methods: Amaranthus flour was utilized for fibre enrichment of chicken-meat protein fortified whole wheat bread (meat bread) by substituting whole wheat flour at 5, 10, 15% level. Physico-chemical characteristics, proximate analysis, instrumental colour and texture profile and sensory evaluation were conducted.

Result: Moisture content, moisture retention, dietary fibres, insoluble dietary fibres and redness of bread increased significantly (P<0.05) at each level of amaranth fortification while fat, moisture-protein ratio, total phenolic content and texture parameters like hardness, springiness, stringiness and chewiness values increased significantly (P<0.05) at higher levels *i.e.* 10% and 15%. Colour and appearance of crust and crumb, flavour and after-taste of treatment increased significantly (P<0.05) while porosity, texture and meat flavour intensity decreased non-significantly (P>0.05). The overall acceptability scores of 10% substitution were significantly (P<0.05) higher than control. The study revealed that 10% amaranth incorporation can be successfully done for the development of fibre enriched chicken meat-protein fortified whole wheat bread (meat bread) without adversely affecting the bread characteristics.

Key words: Amaranth, Dietary fibres, Meat bread, Texture profile.

INTRODUCTION

The fast moving and changing lifestyle have also changed eating habits based largely on ready-to-eat and convenient foods which in turn has boosted the food processing industry. The shift from home cooking to convenience has drastically changed our diet composition. Such foods are generally low in fibre, proteins and essential minerals and vitamins (Verma *et al.,* 2014). Nowadays prevalent lifestyle diseases such as obesity, type 2 diabetes and cardiovascular diseases have been associated with imbalanced and inadequate diet which is nutritionally poor. It is considered that adequate levels of dietary fiber and antioxidants induce immunological processes and increase the defensive abilities of cells in an appropriate manner (Koç *et al*., 2020). The awareness of the consumer towards the benefits of high fibre diets has also taken a leap. Bakery products however provide ideal matrix for increasing functionality and are considered promising for functional supplementation.

Amaranth is a nutritionally rich pseudocereal with high protein, essential amino acid especially lysine; mineral and dietary fibre content (Tamsen *et al*., 2018). Thus, amaranth incorporation in bread formulations may potentially increase its nutritional and functional value. The objective of present study was to develop fibre-rich novel meat bread (chicken meat protein fortified whole wheat bread).

MATERIALS AND METHODS

Ingredients

Whole wheat flour and amaranth flour were procured from reputed firms while spent hen chicken meat powder was prepared in laboratory under standard conditions. Yeast, flavourings, sugar and salt were procured from local market, Bareilly. All the chemicals used were of analytical and food grade and were obtained from standard firms (Hi-Media, CDH *etc*). Breads were prepared according to the formulation given in Table 1 using method described by Umaraw *et al*., (2020). The bread baking was repeated for triplication of the experiment. The research work was carried out at the Division of Livestock Products Technology, ICAR-Indian Veterinary Research Institute, Izatnagar, Bareilly, Uttar Pradesh, India during 2013-2016 as PhD research work of the first author.

Division of Livestock Products Technology, ICAR-Indian Indian Veterinary Research Izatnagar, Bareilly-243 122, Uttar Pradesh, India.

Corresponding Author: Pramila Umaraw, Department of Livestock Products Technology, College of Veterinary and Animal Sciences, Sardar Vallabhbhai Patel University of Agriculture and Technology, Meerut-250 110, Uttar Pradesh, India. Email: pramila1303@gmail.com

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Physico-chemical analysis

The pH of dough and bread was determined by combination electrode digital pH meter. Water activity meter was utilized for assessing water activity of samples. Baking yield was determined by measuring the difference in the sample weight before and after cooking.

$$\text{cooking yield (\%)} = \frac{\text{Weight of cooked bread loaf}}{\text{Weight of uncooked dough}} \times 100$$

Bake loss was calculated:

$$\frac{\text{Weight of the loaf before baking-}\ \text{Weight of the loaf after baking and cooling}}{\text{Weight of the loaf before baking}} \times 100$$

Proximate composition

The proximate composition of meat bread was estimated by methods described by AOAC (1995). Atwater values (fat (9 kcal/g), protein (4.02 kcal/g) and carbohydrate (4 kcal/g) were used to calculate total calories content of cooked products on the basis of 100g portion. An analysis of the percentage of carbohydrate in the samples was determined by numerically formulae (carbohydrate = 100-moisture + protien + fat+ ash). Moisture to protein ratio was derived by dividing the moisture content of chicken meat bread to protein content of the same bread. While, moisture retention of meat breads was calculated by given formula (Umaraw *et al*., 2015).

Moisture retention % =

$$\frac{\text{(\% cooking yield)} \times \text{(\% moisture in bread loaf)}}{100}$$

Total dietary fibre (TDF), soluble dietary fibre (SDF) and insoluble dietary fibre (IDF) were determined by slight modification of an enzymatic method.

Estimation of total phenolics

The total phenolic content in bread was quantified using the Folin-Ciocalteau (F-C) colorimetric method.

Texture and colour profile analysis

Texture profile analysis (TPA) of bread was performed using a Texture Analyser (TMS-PRO, Food Technology Corporation, Maries Road, Suite 120 Sterling, VA, USA). The colour evaluation was done by Lovibond Tintometer (Model F, Greenwich,U.K.). Formula, $(\tan^{-1})^{b/a}$ and $(a^2 + b^2)$ ½, where, a = Red unit. b = Yellow unit.

were used respectively for deriving the hue and chroma values.

Sensory evaluation

Eight-point descriptive scale (Keeton, 1983) with 8 as excellent and 1 as extremely poor, rest being in between was used. Before beginning of the trials, the panelists were briefed about the experiment and product development and all evaluation parameters.

Statistical analysis

The IBM SPSS Statistics software (Version 20.0 for Windows; IBM SPSS Inc, Chicago, 111, USA) was used for analysing all data recorded during the experiment by One-way ANOVA and Mean±S.E. Duncan's multiple range test was used to compare the means. The smallest difference ($D_{5\%}$) for two means was reported as significantly different ($P<0.05$).

RESULTS AND DISCUSSION

Physico-chemical characteristics

The fibre enrichment in the form of amaranth flour produced significant differences in the pH of dough and breads (Table 2). The pH of dough increased with amaranth fortification but statistically the mean values differed significantly ($P<0.05$) only at 15% level. The pH of breads increased with baking. The amaranth flour incorporated breads had significantly ($P<0.05$) higher pH than control which might be due to higher pH of amaranthus flour than that of wheat flour which was replaced (Tamsen *et al*., 2018).

Incorporation of amaranth flour produced an increase in water activity values which might be due to the higher water absorption capacity of fibres as well as due to the smaller particle size of amaranth flour and as the size decreases the surface area increases which tends to absorb more water. The amount of fiber can play an important role in binding water and increasing the moisture content of final products. Higher water binding capacity is associated with the fibre and protein content as water percolates inside fibre pores and binds through hydrogen bonds (Liu *et al*., 2019). The bread with highest level of amaranth (T3) showed the highest water activity that differed significantly ($P<0.05$) from rest.

The baking yield was positively affected by amaranth incorporation as the yield increased with the level of incorporation (Fig 1). The increased yield might be attributed to the higher fibre content of amaranth flour especially IDF and various studies indicate that IDF such as lignin, cellulose and hemicelluloses usually have high water-holding capacity.

The baking loss of the treatments decreased significantly ($P<0.05$) with the increase in level of replacement Fig 1. The lower baking losses might be due to higher moisture content as well as because of lower fermentation due to higher replacement of whole wheat flour

Table 1: Formulation for preparation of amaranth flour incorporated meat bread.

Ingredients (% w/w)	Control	Treatments		
	C	T_1	T_2	T_3
Whole wheat flour (WWF)	70.00	65.00	60.00	55.00
Chicken meat powder (CMP)	30.00	30.00	30.00	30.00
Amaranth flour (AF)	0.00	5.00	10.00	15.00
Yeast	3.00	3.00	3.00	3.00
Sugar	5.00	5.00	5.00	5.00
Salt	1.20	1.20	1.20	1.20
Fat	5.00	5.00	5.00	5.00
Water	75.00	75.00	75.00	75.00

C = Meat bread without amaranth flour (AF), T1= Meat bread with 5% AF, T2 = Meat bread with 10% AF, T3= Meat bread with 15% AF.

which is the major source of starch. Similarly, lower moisture loss was observed in amaranth breads in comparison to whole wheat breads (Liu *et al*., 2019).

Amaranth flour incorporation increased the moisture protein ratio of bread Fig 1. The 15% amaranth containing bread showed the highest ratio while control the lowest. A low moisture protein ratio is desirable for stability of a product. Moisture retention values influences the product eating quality thus higher moisture retention is related to a better product. The moisture retention increased significantly (P<0.05) with each level of substitution Fig 1. This increase in moisture retention might be due to higher amaranth flour level which provided higher insoluble fibres than wheat flour (San-Penella *et al*., 2013).

Proximate and physicochemical attributes

The moisture values increased with the level of incorporation and 15% (T3) amaranth containing bread evinced the highest moisture content (Table 2). The moisture increase might be due to the inclusion of a greater amount of insoluble dietary fibre with the amaranth flour. The higher fibre content of amaranth might be implicated for the increase in moisture content of treatments as the amount of fibre can play important role in water binding and thus increasing the moisture content of breads (Pourafshar *et al*., 2015).

The protein content of samples did not show significant (P>0.05) differences. Results were similar to Pourafshar *et al*. (2015) reporting that amaranth incorporation at 20% in traditional Iranian breads did not much affect their protein content. But amaranth incorporation at higher levels *i.e.* more than 20 per cent have been reported to increase the protein content in breads (Bodroza-Solarov *et al*., 2008; Tosi *et al*., 2002). The fat content increased with replacement but the differences were more prominent (P<0.05) in 10% and 15% level. The results were in agreement with the findings of Nasir *et al*. (2020). The ash content of breads was not much affected by incorporation until the level of incorporation was high enough *i.e.* at 15% level. Dyner *et al*. (2007) observed significant increase in mineral and dietary fibre contents in bread and pasta by amaranth flour substitution at levels up

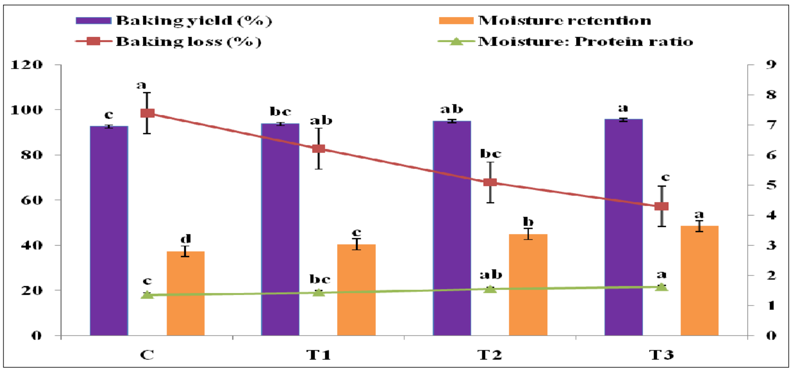

Fig 1: Baking attributes of amaranth flour incorporated meat bread.

Mean values bearing different superscripts in a row differ significantly (P<0.05) n =6

C = meat bread without amaranth flour (AF), T1= meat bread with 5% AF, T2 = meat bread with 10% AF, T3= meat bread with 15% AF

Table 2: Physico-chemical characteristics of amaranth flour incorporated meat bread.

Parameters	C	T1	T2	T3	P-value
pH of dough	5.79^{b}±0.02	5.83^{ab}±0.03	5.85^{ab}±0.03	5.88^{a}±0.02	0.142
pH of baked bread	5.91^{c}±0.02	6.06^{b}±0.02	6.12^{a}±0.02	6.14^{a}±0.01	0.000
Water activity (a_W)	0.921^{b}±0.003	0.923^{ab}±0.002	0.928^{ab}±0.002	0.931^{a}±0.003	0.066
Moisture (%)	40.35^{d}±0.76	43.17^{c}±0.85	47.42^{b}±1.11	50.70^{a}±0.78	0.000
Protein (%)	29.70^{a}±0.47	30.34^{a}±0.35	30.82^{a}±0.82	31.39^{a}±0.51	0.217
Fat (%)	3.97^{c}±0.06	4.13^{bc}±0.07	4.21^{ab}±0.04	4.34^{a}±0.04	0.002
Ash (%)	2.25^{b}±0.06	2.30^{ab}±0.02	2.33^{ab}±0.03	2.39^{a}±0.04	0.146
Carbohydrate (%)	23.73^{a}±1.03	20.06^{b}±0.62	15.22^{c}±0.43	11.18^{d}±0.83	0.000
Energy (Kcal)	250.04^{a}±3.41	239.38^{b}±3.48	222.65^{c}±4.22	209.95^{d}±3.60	0.000
Dietary fiber (%)	2.77^{d}±0.08	3.10^{c}±0.05	3.54^{b}±0.05	4.10^{a}±0.03	0.000
Insoluble	1.40^{d}±0.04	1.63^{c}±0.03	1.98^{b}±0.03	2.43^{a}±0.02	0.000
Soluble	1.37^{c}±0.04	1.47^{b}±0.02	1.55^{b}±0.02	1.67^{a}±0.01	0.000
Total phenolic (mg TAE/100 g dwb)	21.98^{a}±0.61	20.59^{ab}±0.59	19.88^{bc}±0.60	18.29^{c}±0.55	0.002

Mean values bearing different superscripts in a row differ significantly (P<0.05) n =6

C = Meat bread without amaranth flour (AF), T1= Meat bread with 5% AF, T2 = meat bread with 10% AF, T3= Meat bread with 15% AF.

to 20 g/100 g. Liu *et al*. (2019) studied the feasibility of amaranth as an alternative gluten-free ingredient to improve the nutritional quality of gluten-free breads and reported that acceptable breads were produced which were also characterized by having significantly higher levels of minerals.

The carbohydrate values decreased significantly (P<0.05) with each level of replacement. This decrease in carbohydrate values might be attributed to the proportionate decrease of whole wheat flour from the formulation resulting into lower starch level. The carbohydrate content of amaranth is lower than that of whole wheat (Liu *et al*., 2019). The energy values showed significant (P<0.05) decrease with the increase in amaranth flour level. There was significant (P<0.05) decrease at each level of replacement of whole wheat flour with amaranth flour.

The dietary fibre contents increased significantly (P<0.05) with amaranth flour incorporation in breads. Similar results were also reported by Sanz-Penella *et al*. (2013); Tosi *et al*. (2002) and Bodroza-Solaroza *et al*. (2008) in amaranth incorporated breads. Gambus, *et al*. (2002) also observed higher fibre content in amaranth incorporated breads as compared to their gluten free counterparts. Among fibre components the insoluble fraction evinced similar significant (P<0.05) increase in values with amaranth inclusion. The soluble fraction showed significant (P<0.05) increase in values from control but among treatments the 5% and 10% amaranth containing breads differed non-significantly (P>0.05) in soluble fibre content; while 15% amaranth containing bread contained significantly (P<0.05) highest soluble fibre fraction.

Total phenolics

The total phenolic content decreased with whole wheat flour replacement by amaranth flour (Table 2). The decrease was statistically significant (P<0.05) at higher levels of amaranth incorporation. The lower total phenolic content in treatments might be due to lower phenolic content in amaranth flour than whole wheat flour. Thus, higher level of replacement significantly affected the total phenolic content. Gelinas and McKinnon (2006) found that breads baked from wholegrain flours contain much higher amount of phenolic compounds compared with those baked from refined flours or white breads. Ragaee *et al*. (2011) reported that inclusion of 30 g/ 100 g of wholegrain flours into the bread formula resulted in an increase in the content of both phenolic fractions, free and bound, as compared to the control bread.

Instrumental texture profile analysis

The hardness values of amaranth incorporated meat breads increased with the level of incorporation (Table 3). The T3 and T2 samples required significantly (P<0.05) higher force of compression than control. The effect might be due to the dilution of gluten content.

Springiness values indicate the rapidity and degree of recovering after a deforming force. Pourafshar *et al*. (2015) reported that firmness of barbari bread increased by replacing 20% of wheat flour with amaranth flour. Whereas contrary to our results Sanz-Panella *et al*. (2013) observed amaranth substitution did not produce significant changes in crumb hardness until 40 g/100 g substitution. The springiness values increased with amaranth incorporation which differed significantly (P<0.05) from control. This increased springiness of treatment might be attributed to the six times higher lipid content of amaranth flour than wheat flour, which act as a surface-active agent (Sanz-Panella *et al*., 2013). The higher lipid content of amaranth improves elasticity in breads by stabilising entrapped gases (Alvarez-Jubete *et al*., 2010). Similar studies indicating higher elasticity in amaranth incorporated bread with increasing level of incorporation have been reported earlier (Nasir *et al*., 2020).

The cohesiveness and gumminess values decreased non-significantly (P>0.05) with the amaranth replacement. The decrease in values might be attributed to the poorer matrices of bread due to higher gluten reduction. The chewiness value increased with the increase in amaranth flour level in breads. Resilience is breads is a combined function of proteins, fibres and carbohydrates (Liu *et al*., 2019). Amaranth replacement did not significantly affect the resilience of breads which might be due the fact that the increase in fibre content was counteracted by the decrease in carbohydarate while, change in protein content was insignificant (P>0.05).

Instrumental colour analysis

Colour is a perceived quality that influences consumer purchasing behavior (Umaraw and Verma, 2016). The result revealed that redness values increased from 2.68 in control to 3.94 in T3 (Fig 2). A proportionate significant (P<0.05) increase was observed with level of incorporation which

Table 3: Instrumental texture profile of the amaranth flour incorporated meat bread.

Parameters	C	T1	T2	T3	P-value
Hardness (N)	$8.96^{c}\pm0.22$	$9.49^{bc}\pm0.23$	$10.34^{b}\pm0.40$	$12.02^{a}\pm0.50$	0.000
Springiness (ratio)	$0.43^{c}\pm0.02$	$0.51^{b}\pm0.03$	$0.56^{b}\pm0.02$	$0.66^{a}\pm0.02$	0.000
Stringiness (mm)	$22.56^{c}\pm0.90$	$23.05^{bc}\pm0.80$	$24.85^{ab}\pm0.62$	$26.62^{a}\pm0.51$	0.001
Cohesiveness (ratio)	$0.81^{a}\pm0.03$	$0.78^{a}\pm0.04$	$0.76^{a}\pm0.03$	$0.72^{a}\pm0.05$	0.288
Gumminess (N)	$0.98^{a}\pm0.04$	$0.94^{a}\pm0.05$	$0.91^{a}\pm0.04$	$0.85^{a}\pm0.03$	0.195
Chewiness (J)	$0.91^{c}\pm0.05$	$1.02^{bc}\pm0.06$	$1.09^{ab}\pm0.04$	$1.20^{a}\pm0.02$	0.001

Mean values bearing different superscripts in a row differ significantly (P<0.05) n = 9.

C = meat bread without amaranth flour (AF), T1= meat bread with 5% AF, T2 = meat bread with 10% AF, T3= meat bread with 15% AF.

Table 4: Sensory attributes of amaranth flour incorporated meat bread.

Parameters	C	T1	T2	T3	P-value
Colour and appearance of crust	6.88^{b}±0.12	7.00^{ab}±0.11	7.17^{ab}±0.12	7.24^{a}±0.10	0.114
Colour and appearance of crumb	6.78^{b}±0.11	6.90^{ab}±0.10	7.10^{ab}±0.13	7.14^{a}±0.12	0.087
Porosity	7.14^{a}±0.12	7.10^{a}±0.13	7.07^{a}±0.12	7.00^{a}±0.11	0.861
Flavor	6.81^{b}±0.10	7.00^{b}±0.11	7.21^{ab}±0.13	7.26^{a}±0.11	0.021
Texture	7.14^{a}±0.13	7.10^{a}±0.11	7.07^{a}±0.13	6.81^{a}±0.15	0.277
Meat flavor intensity	7.19^{a}±0.12	7.17^{a}±0.10	7.14^{a}±0.15	7.05^{a}±0.10	0.855
After taste	6.83^{b}±0.10	6.98^{ab}±0.09	7.12^{ab}±0.10	7.21^{a}±0.11	0.045
Overall acceptability	6.81^{b}±0.11	6.98^{ab}±0.10	7.17^{a}±0.12	7.07^{ab}±0.12	0.148

Mean values bearing different superscripts in a row differ significantly (P<0.05) n =21.

C = meat bread without amaranth flour (AF), T1= meat bread with 5% AF, T2 = meat bread with 10% AF, T3= meat bread with 15% AF.

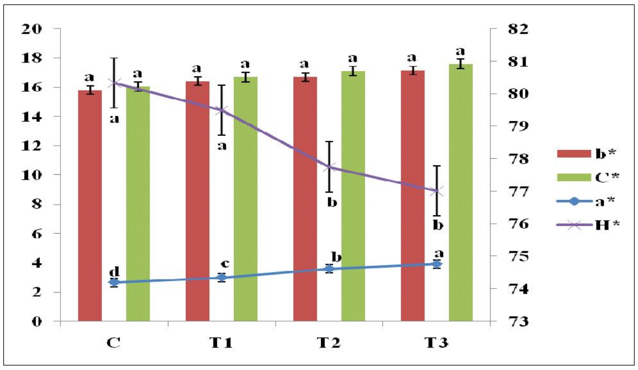

Fig 2: Colour indices of amaranth flour incorporated meat bread. Mean values bearing different superscripts in a row differ significantly (P<0.05) n =9

C = meat bread without amaranth flour (AF), T1= meat bread with 5% AF, T2 = meat bread with 10% AF, T3= meat bread with 15% AF.

might be attributed to the inherent dark colour of amaranthus. The yellowness values increased non-significantly (P>0.05) with the level of amaranth addition in breads. The mean values for chroma also increased non-significantly (P>0.05) from 16.05 in control to 17.60 in T3. Amaranth addition to developed breads brought about significant effects at higher level of incorporation. The mean values differed significantly (P<0.05) at 10 % and 15% replacement from control and T1 (5%).

Sensory evaluation

Colour and appearance of crust and crumb of developed amaranth incorporated breads was rated higher than that of control by the sensory panelists (Table 4). Amaranth incorporation produced darker crusts which was preferred over lighter colour of control. Increased darkness might be attributed to the inherent colour of the amaranth flour. Amaranth flour contains betacyanin pigment which imparts darker colour to the final product in comparison to wheat which lacks this pigment (Tamsen *et al*., 2018). Similarly, Rosell *et al*. (2009) observed that replacement of wheat flour by up to 50% of amaranth led to darker breads with good sensory perception.

The mean porosity values did not differ significantly (P>0.05) among all the samples. This could be due to higher stabilization of gases by high polar lipids present in amaranth during bread making improving its elasticity (Avarez-jubete *et al*., 2010) that counteracted the gluten dilution effect of amaranth. The flavour of bread crumbs is largely affected by enzymatic fermentation while that of crust is affected by heating during baking. Sensory panelists rated T3 significantly (P<0.05) higher than the control for flavour. Rosell *et al*. (2009) observed that sensory analysts preferred nutty flavour of amaranth breads over white breads. The texture of breads evinced non-significant (P>0.05) differences in scores. The lower scores of treatments could be due to the more closed structure. Similar to texture meat flavour intensity did not differ significantly (P>0.05) in all samples but panelists scores decreased with the increase in amaranth flour content. The lower scores could be attributed to the stronger masking effect of amaranth over meat.

The aftertaste of bread samples as perceived by panelists increased with the level of amaranth that was highest at 15% replacement. This might be due to the higher fat content of the sample as amaranth flour contains approximately six times higher fat content than wheat flour (Alvarez-jubete *et al*., 2010). The overall acceptability scores of sensory panelists rated T2 highest which differed non-significantly (P>0.05) with T1 and T2 but significantly (P<0.05) from control. This might be attributed to the texture, porosity and meat flavour intensity which was perceived strongly in T2 without affecting the bread qualities. The flavour attribute, the freshness, colour, texture and biting properties dramatically influence the overall perception of bread. Similarly, Tosi *et al*. (2002) reported that upto 8% substitution of wheat flour with hyperproteic whole amaranth flour produced breads with greater quality and acceptability.

CONCLUSION

Development of novel products is a challenging task that requires controlled monitoring and effect of its ingredients on its nutritional characteristics. As the study reveals incorporation of amaranth flour in developed meat bread (chicken meat protein fortified whole wheat bread) as fibre source is a promising approach where yield, dietary fibres and total phenolic content increased significantly. Textural characteristics like hardness and springiness and colour characteristics like redness were improved. Consumer acceptability and flavour was much appreciated for 10% incorporation. Thus, it can be concluded that incorporation of 10% amaranth flour in chicken meat protein fortified whole wheat bread produces a desirable product with positive consumer response.

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