




# Evaluation of nutrients and organoleptic value of novel value added multibran cookies using multivariate approach

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**Abstract** Cereal and legume flours are intensively being used by food experts to formulate cookies. But their byproducts are discarded in spite of being nutrient rich. The study was conducted to determine nutrients, organoleptic properties and shelf-life of highly nutritive multibran cookies formulated with partial replacement of wheat flour along with the milling byproducts i.e., chickpea husk, moong bean husk, rice bran, broken rice, and wheat bran. The percentages of the byproduct flour, taken for the formulation of the product, was determined using central composite design of response surface methodology. According to the obtained data, Multibran cookies (MBC) possessed rich nutrient composition in comparison with the control sample i.e., the wheat flour cookies (WFC). MBC showed 18% crude protein, 5% crude fiber, higher than the crude protein (7.78%) and crude fiber (2%) of WFC. However, total sugar concentrations of MBC (3.08 g/100 g) was lower than WFC (4.89 g/100 g). Calcium and phosphorus present in MBC were 115.06 mg/100 g and 195.88 mg/100 g respectively, significantly higher ( $p < 0.05$ ) than WFC. The overall acceptability of MBC as indicated by

9-point hedonic scale (8.13) was satisfactory. On the basis of the obtained data it can be said that the selected milling byproducts can be used as potential plant-based sources to develop significant functional products like cookies without affecting its sensory quality and to improve nutritional status of consumer.

**Keywords** Cookies · Microwave heating · Chickpea byproduct · Moong bean husk · Response surface methodology

## Abbreviations

RSM Response surface methodology  
CCD Central composite design  
DPPH 2,2-Diphenyl-1-picrylhydrazyl  
TIA Trypsin inhibitor activity

## Introduction

Rise in global population without any intervention strategy might lead to hunger issues in the near future. According to the survey data and the statistical analysis of population growth, food consumption, along with the food wastage, without any strategic intervention by 2050 there will be an emergence of global food shortage (McKenzie and Williams 2015). Hence, conservation of various food sources is crucial along with lowering food wastage to maintain food and nutrition security. Due to the presence of essential health beneficial factors in plants, researchers and food experts are more interested in the conservation of plant sources viz., grains, vegetables, traditional herbs etc. for novel value-added food production (Niño-Medina et al. 2019). However, during the processing of these plant sources in industries, large amount of byproducts is generated, labelled

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as agro-industrial byproducts. Proper utilization of these byproducts gets hindered due to poor control of processing techniques, limited access to available suppliers, poor marketing channels and difficulty in transferring existing technologies (Oluwajuyitan et al. 2020). Thus a small portion of these agro-industrial byproducts viz., milling byproducts (bran, husks, hulls) are used for animal feed and chemical industries. Rest unused portion of these byproducts are burnt to get rid of them resulting in the production of silica and Green House Gas (Giroto et al. 2015) causing environmental pollution. Therefore, it becomes necessary to find out probable way to exploit these byproducts and their untapped properties.

Studies revealed that the milling byproducts, obtained after the processing of the cereals and legumes, have higher content of nutrients rather than the processed grains. Valorization of milling byproducts can improve the bioavailability of these nutrients viz., protein, lipids, starch, dietary fibers along with micronutrients, and other bioactive molecules (Giroto et al. 2015). Among cereal byproducts rice bran (Sohail et al. 2017), wheat bran (Hassanzadeh-Rostami et al. 2020), broken rice (Tiwari et al. 2011) are already well-known nutrient sources among consumers and regarded as most utilized byproducts for food purposes. Being the most-cultivated cereals, rice and wheat cultivation generates large quantity of byproducts. Legume production also generates a large amount of byproducts. These byproducts, obtained from dhal milling industries are mainly used for animal feed. Chickpea (*Cicer arietinum* L.), also known as garbanzo bean, is the third most cultivated (after dry beans and field peas) and consumed bean throughout the world (Chibbar et al. 2010). Despite being highly nutritious (Niño-Medina et al. 2019), chickpea husk did not get worth acceptance yet in the food industry, rather it is more used in the chemical industry, dye industry, as absorbent. Moong bean is another most cultivated and consumed legumes worldwide, especially in Asian countries (Tiwari et al. 2011). Due to the presence of high fiber and essential micronutrients, the moong bean as a whole and even its outer skin (husk) are being used as a food source. Formulated products utilizing moong husk have shown potential nutrient content and bioactivity (Tiwari et al. 2011).

Rising awareness of consumers about their health is resulting in increased demand of natural, healthy plant-based products. However, westernization in modern times, compromised the vision regarding lifestyle and consumption of food. Hence, in many countries, researchers are using the most popular ready-to-eat and easily accessible snack product such as, cookies as functional food for nutrient supplement (Mohd Basri et al. 2020). Optimization process is a vital step for the formulation of novel products. Response surface methodology (RSM) using central composite design (CCD) is considered as potential tool to

process optimization. This statistical model has been testified as best model to study interaction and optimization. RSM has been used widely in many studies for the formulation of food products. Mohd Basri et al. (2020) used RSM to formulate oat-based cookies based upon central composite design. Value addition of cookies using wheat and oat bran (Milićević et al. 2020) along with rice bran (Sohail et al. 2017) have been carried out in some studies. However, as per best knowledge, no study has been found on multibrand cookies formulation using byproducts of chickpea, moong bean, rice, and wheat in combination using multivariate approach. Hence the present study took initiative to determine the nutrient composition of value added multibrand cookies as potential functional food using multivariate approach for optimization of composite flour from selected cereal legume byproducts and analysis of antioxidant property, in vitro digestibility, sensory attributes, and shelf life evaluation of prepared cookies.

## Experimental design

### Methodology

The milling byproducts of chickpea, moong bean, rice, and wheat were procured from the local dhal mill of Mahendergarh city, Haryana (India) along with other necessary ingredients (spices, salt, Yoghurt/curd) from the local market of the same place. All reagents and chemicals utilized for the analysis of the nutrient composition of the formulated products were of analytical grade. The experiment was carried out in the Department of Nutrition Biology, Central University of Haryana, India.

### Processing of selected milling byproducts

Washed and soaked milling byproducts of chickpea, moong bean, rice, and wheat were blanched separately and kept in room temperature (at  $40 \pm 2$  °C) for natural air drying for 5 h. Due to presence of relatively higher amount of antinutrient in chickpea husk (Niño-Medina et al. 2019), they were processed through soaking, blanching and microwave heating. These pretreatment techniques are considered to be the potent way out to diminish antinutrient by inactivating their inhibitory activity towards nutrient bioavailability (Suhag et al. 2021). The byproducts were subjected to microwave heating at 800 W for 1.5 min. followed by grinding in a laboratory grinder [Philips HL7756/00 Mixer Grinder, 750 W (20,000 rpm)] for 5–7 min to get the fine powder (flour) and sieved through a 200 µm sieve. The processed byproduct-flours were then subjected to formulation of composite flour according to the experimental design using Response Surface Methodology (RSM).

## Multivariate approach for multibran cookie formulation

### *Selection of independent variables and responses*

Selected variables and responses were designed according to the central composite rotatable design (CCRD) of RSM. Literature have been reviewed for optimization of suitable concentration of rice bran flour, wheat bran flour, broken rice flour to develop a desirable product (Sohail et al. 2017; Hassanzadeh-Rostami et al. 2020; Tiwari et al. 2011). For the preparation of composite flour, chickpea husk flour and moong bean husk flour were considered as independent variables and the concentration of rice bran flour, wheat bran flour, broken rice flour were maintained at  $(20 \pm 2) \%$ ,  $(40 \pm 1) \%$ ,  $(10 \pm 2) \%$  respectively. Upper and lower range for chickpea husk flour and moong bean husk flour were nominated between 5 and 15% and 10–25%, respectively. Sensory parameters viz., texture, taste, and overall acceptability were selected as responses in the design. The CCRD was designed (Fig. 1) into thirteen experiments with four factorial points, five replicates i.e. center points, and four axial points.

Responses were determined utilizing regression analysis by fitting second-order polynomial equation:

$$Y = \beta_0 + \sum_{i=1}^n \beta_i X_i + \sum_{i=1}^n \beta_{ii} X_i^2 + \sum_{i \neq j=1}^n \beta_{ij} X_i X_j$$

where  $\beta_0$  = value of response at the center points (0,0),  $X_i$ ,  $X_{ij}$  = variables of design,  $\beta_i$ ,  $\beta_{ii}$ ,  $\beta_{ij}$  = regression coefficients,  $n$  = number of variables.

### *Data analysis*

For the optimization of composite flour formulation, State-Ease Design expert software (Ver. 13.0) was used. Two function variables were considered to plot the response surfaces and 3D- contour graphs. Regarding the accuracy of the designed model coefficient of determination was estimated (significant level of  $p < 0.05$ ).

### **Formulation of value added Multi-bran cookies**

The basic ingredients were the byproduct (chickpea husk, moong bean husk, rice bran, wheat bran, broken rice) flours, wheat flour, jaggery (traditional non-centrifugal cane sugar) (100 g), vegetable oil (115 g), salt (pinch of salt, as per taste), and baking powder (2.5 g) (Fig. 1). 200 g of combined flour composition (byproduct flour and wheat flour) was taken for the formulation where 60% was byproduct flour and rest was wheat flour (40%). The dry ingredients were kneaded with creamy mixture of sugar

and vegetable oil (Okaka 1997) for 6–7 min. using kneader [TECHMATE (India), Planetary mixer, Model 10]. The formed dough was cut out by a round cookies cutter (3.6 X 3.6") to get preferred shapes. After the baking at 200 °C for 20–25 min, the cookies were cooled and stored in polyethylene bags at room temperature for further uses. After baking the dimension of the cookies was increased by 1 inches. Same procedure was followed to formulate 'Control' wheat flour cookies to get a comparative study with novel multibran cookies.

### **Sensory evaluation**

The acceptance and successful formulation of Multibran cookies was determined by sensory evaluation using a 9-point hedonic rating scale (Peryam and Pilgrim 1957) by a panel of 10 judges. They were informed before the process that they would be evaluating cereal bran and legume husk incorporated cookies. They were also instructed to have some water to wash out their mouth by gargling in between the sample products to lessen the residual effect of the prior sample.

### **Characterization of the formulated product**

#### *Proximate analysis*

The proximate analysis [Moisture (925.09), ash (923.03), crude protein (979.09), crude fat (920.39), crude fiber (962.09)] was carried out in triplicate (AOAC 2000). For the analysis of the crude protein and crude fat content Kjeldahl method and soxhlet extraction method were used respectively. However, the carbohydrate content was estimated by using the formula:

$$100 - (\text{Moisture} + \text{Ash} + \text{protein} + \text{fiber} + \text{fat})$$

Soluble and insoluble dietary fiber of defatted products were estimated by the enzymatic method.

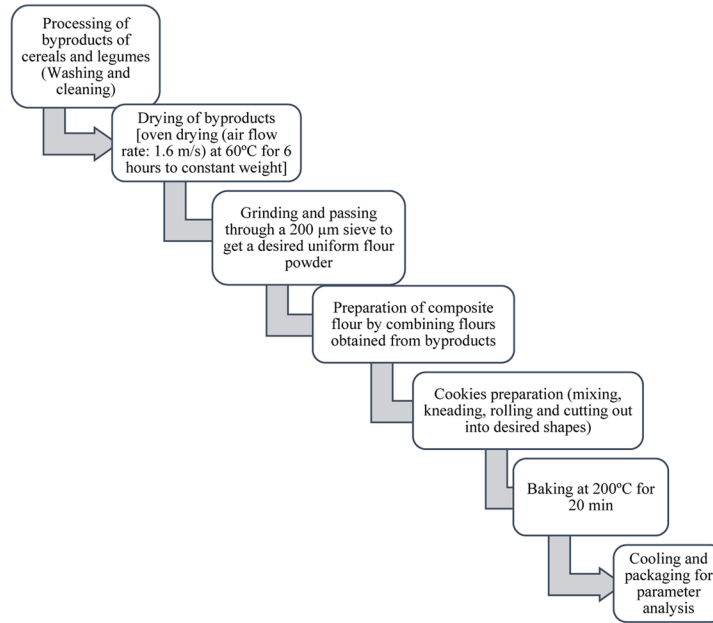
#### *Sugar and starch*

Total soluble sugar was determined from samples diluted with 25 ml ethanol (80%) (Yemm and Willis 1954). The absorbance at 625 nm using UV- VIS spectrophotometer was obtained after one ml of the diluted solution was reacted with freshly prepared Anthrone reagent. Reducing sugar estimation (Somogyi 1945) was carried out by heating up one ml of sugar extract along with Copper reagent A and Copper reagent B for 20 min in boiling water bath. Data was obtained at 520 nm using UV- VIS spectrophotometer. Extracted sugar residue was treated with Perchloric acid for the starch content. For total soluble sugar, the

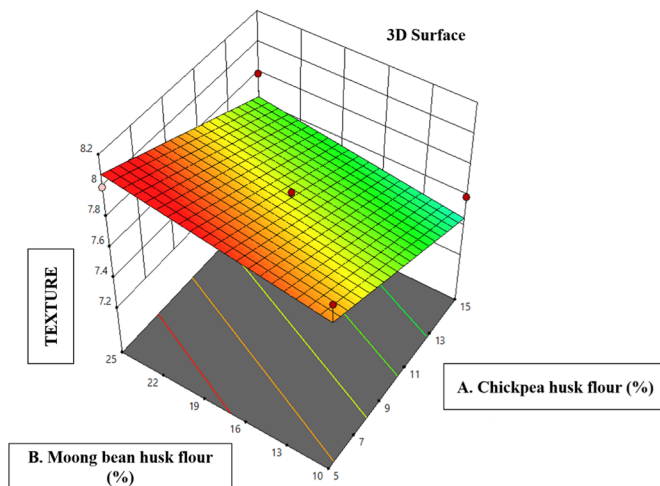
**Fig. 1** Formulation of value added nutrient rich Multibran cookies (a–e)

Std	Run	Space Type	Factor 1 A:Chickpea husk... %	Factor 2 B:Moong bean ... %	Response 1 Texture	Response 2 Taste	Response 3 Overall acceptability
1	3	Factorial	5	10	8	8.2	8.3
2	2	Factorial	15	10	7.6	7.5	7.7
3	4	Factorial	5	25	8	8.5	8.6
4	10	Factorial	15	25	7.8	7.5	7.7
5	11	Axial	2.92893	17.5	8	8	8
6	8	Axial	17.0711	17.5	7.2	7.5	7.6
7	7	Axial	10	6.8934	7.5	7.5	7.6
8	5	Axial	10	28.1066	7.9	8	8
9	12	Center	10	17.5	7.8	7.9	8
10	1	Center	10	17.5	7.8	7.9	8
11	13	Center	10	17.5	7.8	7.9	8
12	9	Center	10	17.5	7.8	7.9	8
13	6	Center	10	17.5	7.8	7.9	8

(a) Central composite design arrangement to optimize the composite flour preparation

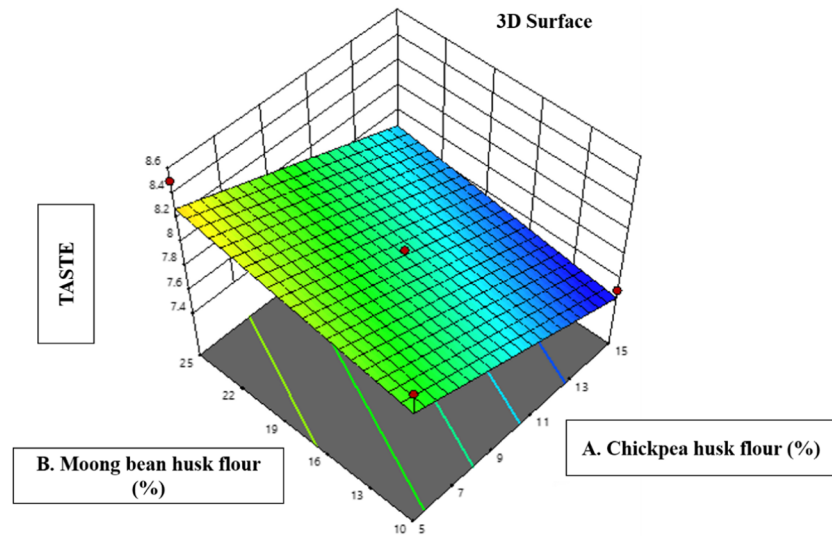


(b) Preparation of cookies substituted with cereal-legume husk flour

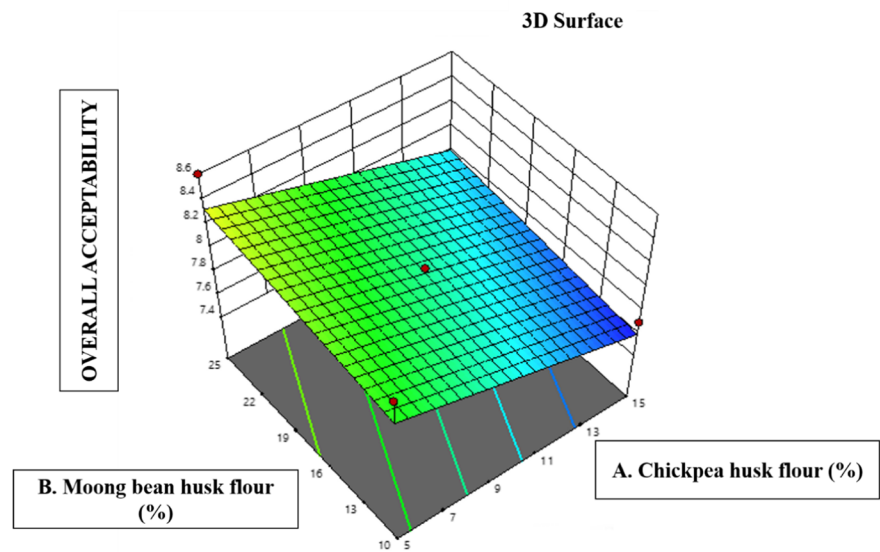


(c) Observation obtained from the optimized design: effect of variables on Texture

Fig. 1 (continued)



(d) Observation obtained from the optimized design: effect of variables on Taste



(e) Observation obtained from the optimized design: effect of variables on Overall acceptability

extracted residue was subjected to glucose estimation. Starch was calculated by multiplying glucose value with 0.9.

#### Minerals

Available iron and phosphorus were estimated followed by UV-spectrophotometer method. However, calcium was evaluated by the titration of acid digested samples (AOAC 2000).

#### Determination of antioxidant activity

Extracted Sample was kept in 4 °C in the dark to determine antioxidant activity. 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical scavenging activity (Hudec et al. 2007) was recorded at 517 nm wavelength using UV–VIS spectrophotometer.

### *Total phenolic activity estimation*

Total phenolic activity (Singleton et al. 1965) was evaluated by using UV–VIS spectrophotometer at 750 nm. Standards were calibrated using Galic acid.

### *In vitro digestibility estimation*

Kjeldahl was used to record the available protein after in vitro digestion of protein available in samples (Mertz et al. 1983). In vitro starch digestibility were obtained by the adapted method of Singh et al. (1982) using defatted sample, treated with pancreatic amylase, dinitrosalicylic acid reagent.

### *Antinutrient evaluation*

After the digestion of samples with nitric acid (0.5 M), it was used to estimate phytic acid level (AOAC 2000) at 465 nm using UV-spectrophotometer. Trypsin inhibitor activity (TIA) was determined from sample treated with trichloroacetic acid (TCA). TCA soluble proteins available in supernatant were estimated (AOAC 2000).

### *Shelf life evaluation of the formulated products*

Shelf life of products were evaluated at the interval of 0, 15, 30, 45, 60, 75, and 90 days.

*Sensory evaluation of stored food products* Sensory profiling was evaluated using 9-point hedonic rating scale by the same panel of 10 judges. Sensory parameters viz., colour, appearance, flavor, taste, texture and overall acceptability of the formulated products were studied during the storage of the products.

*Determination of free fatty acid generated during storage* Free fatty acid level (AOAC 2000) was estimated by dissolving the samples in 50 ml of neutralized Isopropyl alcohol and titrated against 0.25 N NaOH for the quantification.

*Peroxide value analysis* Peroxide value was obtained from the iodine, released from the sample dissolved in 30 ml of acetic acid and chloroform mixture (AOAC 2000).

### *Microbial load determination*

Growth of yeast, mold, and other viable microorganism count (Rico et al. 2019) was recorded to evaluate microbial load and find out the microbial safety of the product. Total Plate Count under aerobic conditions were counted at incubation temperature 30 °C for 72 h. respectively. For Yeast

and Mold Count, plates with sample dilution were incubated at 25 °C for 3–5 days.

### *Statistical analysis*

The data obtained were subjected to analysis of variance for a completely random design using MS Office Excel (2016). All experiments were evaluated in triplicates. The data were presented as means  $\pm$  standard error. Assessment of the statistical significance was carried out by using one-way analysis of variance (ANOVA) followed by Tukey HSD Test ( $p < 0.05$ ).

## **Results and discussion**

### **Analysis of design**

Evaluation of the regression equations of response was done to check the model fitting. The designed model was modified to optimum level to study the quadratic and linear effect of variables on selective responses. Regression equation was also studied to understand the interactive effect of the variables. The adequacy of linearity and quadratic combination of variables was described by examining the significance of data with reference to regression, lack of fit, and residual values (Table 1) by using ANOVA.

Based upon the three dimensional graphs of the responses it is observed that those responses were affected by different level of predictive data. Texture quality was higher when level of variation moves towards the axial position of high moong bean husk proportion. Comparatively lesser substitution of chickpea husk was seemed to be associated with higher texture quality (Fig. 1). During previous study evaluation by Niño-Medino et al. (2019), change in sensory parameters of composite flour (utilized for the formulation of the cookies) was noticed due to variation in different legume substitution. Same pattern was also observed in case of another response, taste parameter (Fig. 1). In case of overall acceptability, the three dimensional graph showed higher score toward the axial portion of moong bean husk and chickpea husk substitution (Fig. 1). Saddle points were pointed out to get the optimized value of the response variables within the given range of formulated value-added products.

### **Optimization and justification of response variables**

Based on the maximization of response values to definite level (obtained using State-Ease Design expert software Ver. 13.0), model design conveyed the data at different level of substitutions of variables. These obtained data were further used for actual experimental responses.

**Table 1** Analysis of variance (ANOVA) result for the optimization of variables and responses

Source	Sum of Squares	Df	Mean Square	F-value	<i>p</i> -value
<i>Texture</i>					
Model	0.4480	2	0.2240	14.03	0.0013 (significant)
A-Chickpea husk flour	0.3747	1	0.3747	23.46	0.0007
B-Moong bean husk flour	0.0733	1	0.0733	4.59	0.0578
Source	Sum of Squares	Df	Mean Square		
Residual	0.1597	10	0.0160		
Lack of fit	0.1597	6	0.0266		
Pure error	0.0000	4	0.0000		
Cor total	0.6077	12	–		
The Model F-value of 14.03 implies the model is significant. There is only a 0.13% chance that an F-value this large could occur due to noise					
<i>Response</i>					
Source	Sum of squares	Df	Mean square	F-value	<i>p</i> -value
Model	0.8511	2	0.4255	17.75	0.0005 (Significant)
A-Chickpea husk flour	0.7243	1	0.7243	30.21	0.0003
B-Moong bean husk flour	0.1268	1	0.1268	5.29	0.0443
Source	Sum of Squares	Df	Mean Square		
Residual	0.2397	10	0.0240		
Lack of fit	0.2397	6	0.0400		
Pure error	0.0000	4	0.0000		
Cor total	1.09	12	–		
The Model F-value of 17.75 implies the model is significant. There is only a 0.05% chance that an F-value this large could occur due to noise					
<i>Overall acceptability</i>					
Source	Sum of squares	Df	Mean square	F-value	<i>p</i> -value
Model	0.6271	2	0.3135	10.32	0.0037 (Significant)
A-Chickpea husk flour	0.5334	1	0.5334	17.56	0.0019
B-Moong bean husk flour	0.0937	1	0.0937	3.08	0.1096
Source	Sum of Squares	Df	Mean square		
Residual	0.3037	10	0.0304		
Lack of fit	0.3037	6	0.0506		
Pure error	0.0000	4	0.0000		
Cor total	0.9308	12	–		
The Model F-value of 10.32 implies the model is significant. There is only a 0.37% chance that an F-value this large could occur due to noise					

According to the set of experiments with minimum deviation of actual to predictive data responses were validated. Same pattern was also observed in a study conducted by Mohd Basri et al. (2020). According to the data provided by CCRD design, the optimized value of independent variables for composite flour obtained to formulate cookies were 10% of chickpea husk flour and 17.5% moong bean husk flour.

### Detailed study of nutritional compositions of MBC Cookies

#### *Proximate compositions*

Addition of protein and fiber rich legume husk in the formulation, showed enhanced protein level almost by 50% accompanied by a significant increase ( $p < 0.05$ ) in the level of fiber as compared to the control i.e. wheat flour cookies

**Table 2** Analyzed nutrient composition of formulated value added product- Cookies per 100 g- Multi-bran cookies (MBC) and wheat flour cookies (WFC)

	Multi- bran cookies (MBC)	Wheat flour cookies (WFC)
<i>Proximate composition</i>		
Moisture* (%)	5.10 ± 0.07 <sup>a</sup>	4.05 ± 0.02 <sup>b</sup>
Crude protein (%)	18.01 ± 0.01 <sup>a</sup>	7.78 ± 0.06 <sup>b</sup>
Crude fat (%)	4.54 ± 0.04 <sup>b</sup>	17.98 ± 0.19 <sup>a</sup>
Crude fiber (%)	5.01 ± 0.06 <sup>a</sup>	2.01 ± 0.02 <sup>b</sup>
Ash (%)	6.56 ± 0.10 <sup>a</sup>	1.10 ± 0.02 <sup>b</sup>
Total carbohydrate (%)	59.82 ± 2.01 <sup>b</sup>	70.47 ± 0.01 <sup>a</sup>
<i>Sugars and starch</i>		
Non-Reducing sugar (g/100 g)	0.91 ± 0.01 <sup>b</sup>	1.94 ± 0.01 <sup>a</sup>
Reducing sugar (g/100 g)	2.17 ± 0.18 <sup>b</sup>	2.95 ± 0.02 <sup>a</sup>
Total soluble sugar (g/100 g)	3.08 ± 0.05 <sup>b</sup>	4.89 ± 0.01 <sup>a</sup>
Starch (g/100 g)	26.08 ± 0.04 <sup>b</sup>	27.01 ± 0.01
<i>Minerals (mg/100 g)</i>		
Iron	16.89 ± 0.08 <sup>a</sup>	1.05 ± 0.06 <sup>b</sup>
Calcium	115.06 ± 4.03 <sup>a</sup>	6.07 ± 0.01 <sup>b</sup>
Phosphorus	195.88 ± 0.20 <sup>a</sup>	21.22 ± 0.01 <sup>b</sup>
<i>Antinutrient</i>		
Trypsin inhibitor activity (TIU/mg)	1.71 ± 2.01 <sup>a</sup>	1.02 ± 1.06 <sup>b</sup>
Phytic acid (mg/100 g)	293.01 ± 1.32 <sup>a</sup>	140.02 ± 1.12 <sup>b</sup>
<i>Antioxidants</i>		
Total phenolic content (mg GAE/ g)	3.11 ± 0.05 <sup>a</sup>	1.59 ± 0.22 <sup>b</sup>
DPPH Radical Scavenging Activity (%)	45.99 ± 0.05 <sup>a</sup>	12.03 ± 0.25 <sup>b</sup>
<i>In vitro digestibility of Starch and Protein</i>		
In vitro digestibility of Starch (mg maltose released/ g of product starch)	30.51 ± 0.07 <sup>b</sup>	32.99 ± 0.05 <sup>a</sup>
In vitro digestibility of Protein (%)	69.89 ± 0.15 <sup>a</sup>	52.01 ± 0.01 <sup>b</sup>
<i>Dietary fiber</i>		
Soluble dietary fiber(g/100 g)	6.85 ± 0.12 <sup>a</sup>	0.98 ± 0.01 <sup>b</sup>
Insoluble dietary fiber (g/100 g)	3.53 ± 0.11 <sup>a</sup>	1.04 ± 0.02 <sup>b</sup>
Total dietary fiber (g/100 g)	10.38 ± 0.06 <sup>a</sup>	2.02 ± 0.05 <sup>b</sup>

Data presented are significantly different,  $p < 0.05$  (Statistical analysis has been done row wise)

(WFC) (Table 2). Crude protein content (%) in MBC and WFC was estimated (18.01 ± 0.01) % and (7.78 ± 0.06) % respectively. The moisture content of the MBC was around 5%. The moisture content of the product is directly linked with its shelf life as it measures the probability and vulnerability of microbial contamination. The estimated moisture content was found to be acceptable enough to store for a longer duration at appropriate conditions as low moisture content between 1 and 5% indicates less perishable. However, moisture in the control sample i.e., WFC was lower [(4.05 ± 0.02) %] than the MBC [(5.10 ± 0.07) %]. WFC was

found to be crispier than MBC. Higher moisture content in comparison with control might attributed to the fact of degradation of carbohydrates. Besides, lower carbohydrate level also contributes to the increase absorption of oil as a binding agent to form an appropriate cookies dough (Singh Sibian and Singh Riar 2020). Crude fat (%) was found lesser in MBC [(4.54 ± 0.04) %] than WFC [(17.98 ± 0.19) %]. The Proximate composition of novel MBC cookies was found to be enhanced in comparison to wheat flour cookies (WFC). The higher level of fiber content present in MBC makes the novel product appropriate for improved digestion. Due to lesser carbohydrate content in MBC, it might be an eligible food source for the diet dealing with weight management, blood sugar, blood pressure. Low-carb food products in the diet can lower the triglyceride level (Kämmerer et al. 2021). Consumption of MBC might attribute to improved health status rather than the wheat flour cookies available in the market.

*Minerals*

Higher level of ash content (%) indicates enhanced mineral proportion. Ash content (%) was significantly higher ( $p < 0.05$ ) in MBC (6.56 ± 0.10) % than WFC (1.10 ± 0.02) %. Iron (16.89 ± 0.08 mg/100 g), calcium (115.06 ± 4.03 mg/100 g), and phosphorus (195.88 ± 0.20 mg/100 g) levels were found considerably higher than control (WFC) (Table 2). Enhanced protein, fiber, and ash content due to legume fortification was acknowledged by Niño-Medino et al. (2019). Presence of jaggery, moong bean husk, and rice bran in the formulation can contribute in improved iron level. According to a study by Sohail et al. (2017), rice bran contains significant amount of iron. Micronutrients are essential for the ideal functioning of living. In WFC, Iron content was 1.05 ± 0.06 mg/100 g, calcium level was 6.07 ± 0.01 mg/100 g, and phosphorus found was 21.22 ± 0.01 mg/100 g, which are lesser than minerals present in MBC. Hence, micronutrient-rich MBC can be an addition to a nutritious snack diet to enhance health status. Although there are micronutrient rich products available in market, they also provide high calorie which increases risk of overweight or metabolic disorders upon consumption. In contrast, being low-calorie bakery product, MBC could be beneficial to prevent the micronutrient deficiencies among women as well as the children worldwide.

*Antinutrient*

Trypsin inhibitor activity was found 1.71 ± 2.01 TIU/ mg in MBC along with 293.01 ± 1.32 mg/100 g of phytic acid content (Table 2). Despite presence of high level of antinutrient in chickpea husk (Niño-Medina et al. 2019), MBC showed negligible antinutrient content in comparison to source. This might occur due to various processing



techniques used viz., washing, soaking, blanching during preparation of the product (Shi et al. 2018). As most of the antinutrient contents are water soluble, soaking easily dissolves those compounds. Moreover, some of these contents protein in nature. Hence blanching along with soaking has been found to be effective to lower their activity (Avilés-Gaxiola et al. 2018; Dagostin 2017). Although heating effects very moderately upon phytic acid content, trypsin inhibitor activity decreased significantly (Shi et al. 2018). However, WFC contained lower antinutrient contents due to absence of the byproducts. Trypsin inhibitor activity in WFC was found to be  $1.02 \pm 1.06$  TIU/mg and phytic acid was  $140.02 \pm 1.12$  mg/100 g.

#### *Phenolic compounds and antioxidant properties*

Total Phenolic content and DPPH Radical Scavenging Activity was enhanced with the substitution of developed composite flour. Total Phenolic content and DPPH RSA for the MBC were  $3.11 \pm 0.05$  mg GAE/g and  $(45.99 \pm 0.05)$  % respectively. Whereas, the control type (WFC) showed  $1.59 \pm 0.22$  mg GAE/g Total Phenolic content with minimal DPPH Radical Scavenging Activity  $[(12.03 \pm 0.25)$  %] (Table 2). Enhanced Total phenolic content along with improved antioxidant activity due to substitution of chickpea husk in the formulation of bread was also reported in a study (Niño-Medina et al. 2019). Many ongoing studies have recorded significant positive correlation ( $p \leq 0.05$ ) between the available phenolic contents and the antioxidant activity of food (Minatel et al. 2017; Dong et al. 2019; Xiang et al. 2019; Suleria et al. 2020). Wheat bran flour (Higuchi 2014) might also have significant involvement in the availability of antioxidant activity in the formulated cookies. Blanching, traditional oven or microwave heating showed an effect on the phenol activity of prepared foods in some previous studies (Minatel et al. 2017). The enhanced antioxidant content in MBC might contribute to the minimization of the risk of cardiovascular diseases, diabetes relatively than the consumption of wheat flour cookies (WFC) or 100% wheat flour based products. Incorporation of MBC as snacks can be a bonus to the diet upholding the taste with health.

#### *In vitro protein and starch digestibility*

Estimated in vitro starch digestibility of MBC was  $30.51 \pm 0.07$  mg maltose released/g of product starch and that of WFC was  $32.99 \pm 0.05$  mg maltose released/g of product starch. Although in vitro starch digestibility of WFC was better than that of MBC, in vitro protein digestibility of MBC was better than that of WFC. Lack of gluten in legume husk and small proportion of incorporated wheat bran in

composite flour development might attribute to the higher in vitro protein digestibility (%) of MBC  $[(69.89 \pm 0.15)$  %] (Table 2). Gluten of wheat creates a difficult condition for enzymatic hydrolysis due to presence of Proline fractions (Gulati et al. 2020). Gluten containing wheat flour cookies (Control) showed lesser in vitro protein digestibility which was  $(52.01 \pm 0.01)$  %. Improved in vitro digestibility of starch and protein was also observed in a study (Singh Sibiyan and Singh Riar 2020) due to incorporation of germinated legume grains lacking gluten protein. Market cookies are generally 100% wheat flour based products, whereas MBC is almost gluten-free product. Therefore, novel MBC cookies can be a nutritive baked snack to boost digestibility.

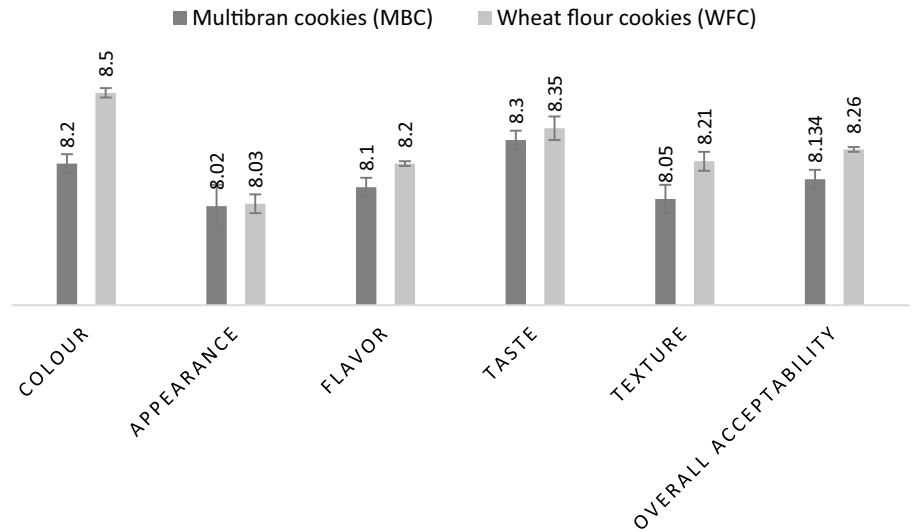
#### *Dietary fiber*

Substitution of composite flour in cookies formulation showed significant increase in soluble dietary fiber (g/ 100 g) of MBC  $(6.85 \pm 0.12)$  in comparison to WFC  $(0.98 \pm 0.01)$ . Insoluble dietary fiber (g/ 100 g) level was also elevated in MBC  $(3.53 \pm 0.11)$ . WFC contained  $1.04 \pm 0.02$  g/100 g insoluble dietary fiber. Total dietary fiber (g/ 100 g) of the formulated MBC and WFC was  $10.38 \pm 0.06$  g/ 100 g and  $2.02 \pm 0.05$  g/ 100 g respectively (Table 2). Similar enhanced level of dietary fiber was also observed in a study after substitution of chickpea husk fiber (Niño-Medino et al. 2019). Commercially available high fiber cookies are generally made utilizing wheat bran or wheat grain. Some ongoing studies are also there to prepare high fiber cookies with wheat bran and mango peel powder (Ghimire 2018). Due to a much higher level of fiber (83.45 g/100 g) in Chickpea husk, studies are going on to formulate high fiber food products using chickpea husk extract (Niño-Medino et al. 2019). Formulated MBC in this study could contribute to fiber requirement to much extent compare to WFC. Throughout the large intestine, if fiber remains intact without being affected by fermentation, it can provide a laxative effect. The available fiber can help maintaining metabolic health conditions viz., lower cholesterol, better glycemic control.

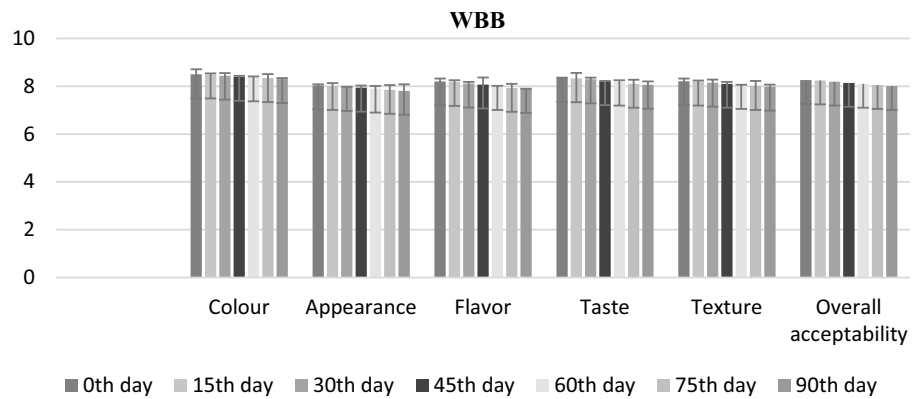
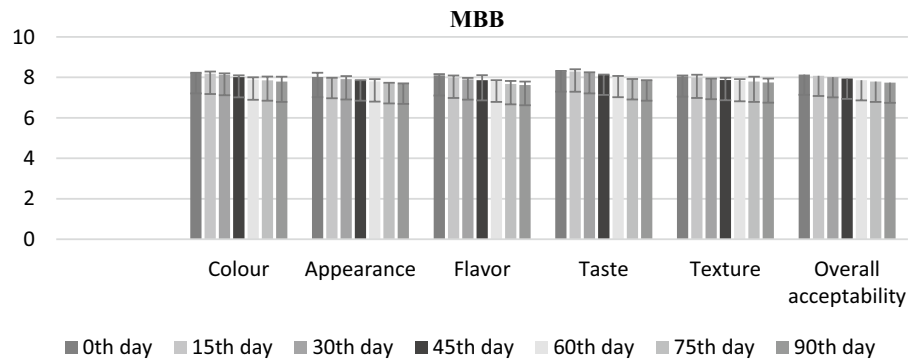
#### *Sensory evaluation of cookies*

Evaluation of sensory parameters of the formulated products is usually carried out among panelists to find out the possible acceptability of the novel products as a supplementary food product for the health benefits along with the minimization of the malnutrition. The overall acceptability was more in case of WFC  $(8.26 \pm 0.01)$  than MBC  $(8.134 \pm 0.04)$ , although MBC showed satisfactory acceptability (Fig. 2) in sensory profiling among panelists. Significant differences ( $p \leq 0.05$ ) between treatments was observed in the sensory

**Fig. 2** Sensory evaluation of the formulated food products (a, b)



**(a)** Freshly prepared Multi-Bran Cookies and Wheat Flour Cookies



**(b)** The effect of storage on sensory quality characteristics of Products

score of WFC and MBC. Colour ( $8.2 \pm 0.04$ ) and texture ( $8.05 \pm 0.06$ ) were slightly affected in MBC. Whereas, WFC showed better colour ( $8.5 \pm 0.02$ ) and texture ( $8.21 \pm 0.04$ ). Appearance, flavor, and taste of the products were in the same range. From the obtained data, it seemed that the use

of byproducts in cookies production can break the stigma associated with edible agricultural byproduct consumption and might be easier to grow attention among consumers worldwide to accept the product. Niño-Medino et al. (2019) and Bora and Kulshrestha (2015) also concluded the same

**Table 3** Shelf life determination of novel value added products

Day intervals	0th day	15th day	30th day	45th day	60th day	75th day	90th day
<i>MBC</i>							
Peroxide value (meq peroxide/1000 g)	2.24 ± 0.01 <sup>e</sup>	2.65 ± 0.08 <sup>b</sup>	4.98 ± 0.04 <sup>c</sup>	6.78 ± 0.11 <sup>a</sup>	8.91 ± 0.01 <sup>e</sup>	11.05 ± 0.04 <sup>c</sup>	12.98 ± 0.03 <sup>d</sup>
Free fatty acid (mg KOH/100 g)	0.52 ± 0.08 <sup>b</sup>	0.61 ± 0.10 <sup>a</sup>	0.69 ± 0.12 <sup>a</sup>	0.75 ± 0.11 <sup>a</sup>	0.79 ± 0.16 <sup>c</sup>	0.82 ± 0.08 <sup>b</sup>	0.90 ± 0.02 <sup>d</sup>
<i>WFC</i>							
Peroxide value (meq peroxide/1000 g)	2.01 ± 0.01 <sup>e</sup>	3.05 ± 0.08 <sup>b</sup>	4.89 ± 0.04 <sup>c</sup>	6.94 ± 0.11 <sup>a</sup>	8.89 ± 0.01 <sup>e</sup>	10.87 ± 0.04 <sup>c</sup>	12.12 ± 0.03 <sup>d</sup>
Free fatty acid (mg KOH/100 g)	0.31 ± 0.08 <sup>b</sup>	0.42 ± 0.10 <sup>a</sup>	0.55 ± 0.12 <sup>a</sup>	0.61 ± 0.11 <sup>a</sup>	0.73 ± 0.16 <sup>c</sup>	0.81 ± 0.08 <sup>b</sup>	0.85 ± 0.02 <sup>d</sup>

Data presented are significantly different,  $p < 0.05$  (Statistical analysis has been done row wise). The products were stored for 90 days at room temperature

observation regarding the use of chickpea husk and moong bean husk about product formulations.

#### *Shelf life of the prepared food products*

Formulated products were further stored to check their shelf life. Fig. 2 showed that the sensory attributes of the stored products were intact until 90th day that is 3 months from the manufactured date.

Peroxide value and free fatty acid value of both the products was observed to increase moderately until 90th day. They showed almost 2 meq peroxide/1000 g and 12 meq peroxide/1000 g peroxide value on initial day and 90th day of the assessment of storage stability. Peroxide value of freshly prepared MBC i.e., on 0th day was  $2.24 \pm 0.01$  meq peroxide/1000 g. On 90th day, the value increased to  $12.98 \pm 0.03$  meq peroxide/1000 g. However, the control (WFC) showed  $2.01 \pm 0.01$  meq peroxide/1000 g peroxide value on 0th day and  $12.12 \pm 0.03$  meq peroxide/1000 g on 90th day of the storage. Free fatty acid value was recorded in the range 0.3 to 0.8 mg KOH/100 g in these novel formulations during the assessment. The obtained peroxide value was moderate. Freshly prepared MBC and WFC showed  $0.52 \pm 0.08$  and  $0.31 \pm 0.08$  mg KOH/100 g Free fatty acid values respectively. On 90th day, the values increased to  $0.90 \pm 0.02$  and  $0.85 \pm 0.02$  mg KOH/100 g for MBC and WFC respectively (Table 3). Data recorded on 90th day were also acceptable as the peroxide value should not be above 10–20 meq/kg fat (Connell 1975) to avoid rancidity flavor. During the storage, the overall peroxide values and free fatty acid values of WFC were lower than MBC as MBC contained milling byproducts and these byproducts have tendency to generate rancidity within short period of storage (Mohammadi et al. 2021). However, the obtained data indicated higher storage stability of MBC as ready-to-eat food.

#### *Microbiological safety of food*

The assessment of the microbial count of the products is essential to evaluate food safety. The *Total Plate counts*

**Table 4** Microbial load (Log CFU/g) analysis of freshly formulated value added products (MBC and WFC)

No. of days	MBC	WFC
<i>Total plate count</i>		
0th day	2.11 ± 0.01	2.07 ± 0.12
15th day	2.19 ± 0.05	2.13 ± 0.05
30th day	2.38 ± 0.12	2.25 ± 0.02
45th day	2.52 ± 0.01	2.30 ± 0.22
60th day	2.66 ± 0.09	2.35 ± 0.19
75th day	2.81 ± 0.21	2.42 ± 0.20
90th day	3.07 ± 0.08	2.49 ± 0.04
<i>Yeast and Mold Count</i>		
0th day	–ve	–ve
15th day	–ve	–ve
30th day	–ve	–ve
45th day	–ve	–ve
60th day	–ve	–ve
75th day	2.07 ± 0.05	2.11 ± 0.04
90th day	2.15 ± 0.14	2.19 ± 0.07

–ve: not detected

(Log CFU/g) were  $2.11 \pm 0.01$  and  $2.07 \pm 0.12$  found to be in freshly prepared MBC and WFC respectively under evaluation (Table 4). Then successively the total plate count (Log CFU/g) increased up to the mark of 3. Yeast and mold counts in freshly prepared formulations were negative. The yeast counts (Log CFU/g) increased to  $2.07 \pm 0.05$  and  $2.11 \pm 0.04$  after 75th day in MBC and WFC respectively (stored room temperature).

All formulations of prepared foods from 0th day to 90th day were within the parameters, and can be said microbiologically safe. Based upon another studies formulating food recipes using milling byproducts viz., bran of cereals and legumes were found microbiologically safe and healthy enough to incorporate in diet (Aktaş and Akın 2020).

## Conclusions

Based upon the evaluated scores and the overall analysis, it can be concluded that MBC exhibited an ample amount of nutrients, better-quality with satisfactory sensory acceptance. A notable change in nutrient composition, antioxidant activity, and in vitro digestibility of MBC explored probable utilization of milling byproducts, especially the byproduct of chickpea (Chickpea husk) and moong bean (Moong bean husk) as alternative food sources. In the cookies available antinutrient was lesser (within the tolerable level) due to processing of byproducts before product formulation. MBC showed better nutrition composition than the control sample i.e., WFC (formulated using only wheat flour). Although peroxide value and free fatty acid value of MBC were higher than that of WFC during the storage period at room temperature, the values of the same in case of MBC were within the acceptable range. The formulated product, MBC retained its sensory quality until the 90th day. This current study discloses a novel potential research area of value-added product formulation by using processed cereal-legume byproducts.

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**Authors' contribution** Manali Chakraborty- Investigation, Data analysis, Drafting, Writing, Editing. Savita Budhwar (Corresponding Author)- Supervising, Proposal drafting, Methodology, Validation of Data analysis, Proof reading, Editing. Suneel Kumar- Supervising, Validation of Data analysis, Proof reading, Editing.

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**Availability of data and materials** The data that support the findings of this study are available from the corresponding author upon reasonable request. Code availability Not applicable.

## Declarations

**Conflict of interest** Authors disclose no conflict of interest.

**Consent to participate** All authors gave their consent to participate in this study.

**Consent for publication** All authors gave their consent for the publication of this manuscript article.

**Ethics approval** This research article does not contain any studies with human participants or animals performed by any of the authors.

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