OPTIMIZATION OF MILLET-LEGUME BASED COMPOSITE FLOUR, ITS RHEOLOGY AND USE AS PRO-HEALTH INGREDIENT IN BAKERY INDUSTRY

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(Date of Receiving : 08-01-2021; Date of Acceptance :15-04-2021)

ABSTRACT

Modern day lifestyle and eating habits have significantly contributed to several health disorders and calls for inclusion of healthy ingredients in bakery industry. Dietary modification is suggestive of protecting against diabetes, cancer, cardiovascular disease, and obesity. Amongst them Diabetes Mellitus is one of five leading causes of deaths and debilitating disease in the world. With these serious health repercussions in mind, the present research intends to develop a functional composite flour comprising millets and legumes as a healthy bakery ingredient and also exploit its nutritional benefits in diabetes diet management. With extensive literature review, foxtail millet, pearl millet, finger millet and Bengal gram were chosen to understand the health benefits on diabetes patients over refined flour. Mixture Optimal (Custom) Design of Design Expert, v. 11 was used for the statistical analysis. For the experimental design, the composite flour components (Foxtail millet, Pearl millet, Finger millet and Bengal gram) were the variables while the functional properties i.e., Water absorption capacity (WAC), oil absorption capacity (OAC), swelling power (SP), foaming capacity (FC), foaming stability (FS) and bulk density (BD) were the responses. The optimised flour with 20 % wheat flour was tested for dough characteristics using Brabender. Optimization with mixture optimal custom design resulted in six different solutions. Amongst them the best solution with highest desirability was selected which was foxtail millet (49.65%), finger millet (10%), pearl millet (10%) and Bengal gram (10.35%) with desirability of 0.864. The ANOVA, R2 and R2 adjusted values for WAC, OAC, SP, FC and BD showed that the composite flours were statistically significant whereas FS model was not fit implying no significant effect of any composite flour ingredient on FS. Farinograph confirms that the optimised composite flour is high-quality strong flour. Composite flour with accepted functional properties can be obtained from composite blends of millets and legumes which could be helpful in substituting refined flour from bakery products and could also play a beneficial role in diabetes diet management.

Keywords: Composite flour, Millets, Functional properties, Bakery products, Diabetes Mellitus, Design expert.

INTRODUCTION

The globalisation has led to a paradigm shift in the basic food consumption from coarse grains to more refined cereals. The gradual acquisition of the food habits by these refined cereals has derailed our dietary intake of fibre (Oghbaei and Prakash, 2018). In parallel, there is a rising popularity in instant energy food resulting in number of lifestyle disorders and onset of metabolic diseases such as diabetes mellitus (Moreno-Fernández et al., 2018). According to International Diabetes Federation (IDF) Type 2 diabetes is the most common type of diabetes, characterized by insulin resistance and in some individuals might also result into hyperglycaemia. Over the years, researchers and clinicians has presented an insight to the etiopathogenesis of the disease, still growing into multitudes with 463 million in 2019 (20-79 years) and expected to be 700 million by 2045.
India is ranked second with the largest adult population of 77 million in 2019 and contributes highest to the mortality rate (IDF 2019). With several precursors such as family history, overweight, unhealthy diet, physical inactivity and ethnicity, the healthy lifestyle changes remain the primary intervention and cornerstone for diabetes management (Sami et al., 2017; Uusitupa et al., 2019).

Several epidemiological studies and RCT including (Allison et al., 2016; Nelson et al., 2016; Fontes- Villalba et al., 2016; Aguilera et al., 2016) has confirmed that dietary manipulation in particular intake of whole grain & fruit act as a protective against diabetes and associated comorbidities.

Healthy controlled diet based on cereals (wheat, rice, maize, millets etc), legumes (Bengal gram, green gram, black gram etc), fruit & vegetables (apple, carrot etc), fibres (soluble & insoluble) and antioxidants is the concurrent solution alongside medication for disease management and in controlling its on-set (Evert et al., 2014; Alkhathib et al., 2017).

Millets and legumes are nutritionally rich in terms of protein, carbohydrate, minerals, dietary fibre and antioxidant. Magnesium present in millets help in the prevention of Type II Diabetes by increasing the efficiency of Insulin and glucose receptors (Kostov, 2019). Finger millet has the highest calcium content among all cereals (344 mg/100 g) (Devi et al., 2014), with an important role in insulin secretion (Ahn et al., 2017). The dietary fibre i.e., soluble and insoluble present in millets and legumes helps in maintaining post prandial blood glucose level and low glycaemic index of food (Kumar et al., 2018; Bresciani and Marti, 2019). Millets and legumes have abundant bioactive compounds with hypoglycaemic and antioxidant activity (Singhal et al., 2014; Ambati and Sucharitha, 2019). The millet proteins are a good source of essential amino acids, in particular sulphur containing amino acids methionine and cysteine but deficit of lysine and threonine. However, combining legumes with millets will overcome the deficiency of lysine (Anitha et al., 2020). This balanced nutritional blend can be an alternative for type 2 diabetic patients.

Bakery products are easy and readily available food however is detrimental for diabetics owing to its ingredients. An approach to change or replace some of the ingredient such as refined flour from whole grain flour in bakery products can make a healthy food alternative for type 2 diabetic patients.

The response surface methodology is a statistical tool which create design of experiments using input variables influencing different response variables. Estimated response variables of the experiment are affected by moderation in input variables level (Montgomery, 2013). Limited scientific literature regarding the combination of millets and legumes for formulation of nutritionally rich composite flour necessitates the use of RSM. Therefore, the present research is aimed at optimizing the millet-legume based composite flour using foxtail, finger, pearl millet and Bengal gram and analyse its effect on the functional and rheological properties to achieve a composite flour suitable for diabetic patients and also for bakery industry.

### MATERIALS AND METHODS

#### Source of materials

Bengal gram was procured from local market and milled at a local mill. Foxtail millet flour was purchased from Muthu Organics (Tamil Naidu, India). Finger millet and pearl millet flour was collected from Crop-Connect (New Delhi, India). All the Chemicals used in the present study were of analytical grade from Sigma- Aldrich Co. (Steinheim, Germany).

#### Experimental design for composite flour

The mixture optimal (custom) model of response surface methodology (RSM) (Design Expert 11 version) was used to investigate the effect of different flour including foxtail millet(10-46.67%), finger millet(10-30%), pearl millet(10-30%) and Bengal gram(10-15%) at varying levels of its weight percentage on the functional characteristics of the composite flour (Table 1), with 20 random treatments (Table 2). The range of the independent variables of each flour was based upon the previously available literature (Saha et al., 2011; Awolu, 2017; Bresciani and Marti, 2019; Marak et al., 2019) and on preliminary trials. The dependent variables included the functional characteristics i.e., water absorption capacity (WAC), oil absorption capacity (OAC), swelling power (SP), foaming capacity (FC) & foaming stability (FS) and bulk density (BD).

#### Water absorption capacity

Water absorption capacity (WAC) of each sample treatment was measured using the method described by (Diniz and Martin, 1997) with slight modifications. About 0.5 g of each flour treatment was mixed with 10 ml of distilled water in centrifuge tubes and spined for 30 s. The dispersals were allowed to stand at room temperature for 30 min and centrifuged for 25 minutes at 3000 rpm. Whatman No.1 filter paper was used to filter the supernatant and the final volume was noted.

\[
\text{water absorption capacity (ml/g)} = \frac{\text{amount of water absorbed}}{\text{weight of sample}}
\]

#### Oil absorption capacity

Oil absorption capacity (OAC) was measured using method of (Narayana and Rao, 1982) with certain modifications. Approximately, 1 g of each flour treatment was mixed with 10 ml of refined oil in 15ml centrifuge tube and was mixed using vortex. Suspensions were kept aside for 30 minutes. The mixture was then centrifuged at 3000 rpm for 20 minutes and thereafter supernatant was transferred to graduated cylinder to record the volume.

\[
\text{oil absorption capacity (ml/g)} = \frac{\text{volume of oil added} - \text{volume after absorption}}{\text{weight of sample}}
\]

#### Swelling power

Swelling power (SP) was estimated with slight modifications in method proposed by Leach, 1962. One gram of each sample treatment is amalgamated with 10 ml distilled water and was heated for 30 minutes at 80° C. The suspensions were kept aside for 30 minutes. After heating, suspensions were centrifuged at 1000 rpm for 15 minutes. Supernatant was poured and weight of paste was taken.
Swelling power (g/g) = \frac{\text{weight of paste}}{\text{weight of flour}}

**Foaming capacity and Foaming stability**

The foaming capacity (FC) and foaming stability (FS) was estimated using method of (Narayana and Rao, 1982) with certain modifications. Each flour treatment of approximately 2 g was blended with 50 ml distilled water and was homogenised using a hand blender for 1 minute to form foam. The solution was then poured into a 100ml graduated cylinder and the volume was documented.

\[
\text{Foaming capacity (\%)} = \frac{\text{volume after whipping} - \text{volume before whipping}}{\text{volume before whipping}} \times 100
\]

All the treatments were left undisturbed for 1 hour at ambient temperature and FS was calculated.

\[
\text{Foaming stability (\%)} = \frac{\text{foam volume after stand three minutes}}{\text{initial foam volume}} \times 100
\]

**Bulk density**

Bulk density (BD) was calculated using the method of (Okaka and Potter, 1979). Each flour treatment of 50g was weighed and transferred to graduated cylinder. The graduated cylinder was then tapped multiple times thereafter volume of flour was recorded.

\[
\text{Bulk density (g/mL)} = \frac{\text{weight of sample}}{\text{volume of sample after tapping}}
\]

**Statistical analysis**

The obtained data was examined for analysis of variance (ANOVA) and regression models using the Response Surface Methodology (Design-Expert software version 11, Stat-EaseInc. Minneapolis, U.S.A) to investigate the effect of foxtail millet (A), finger millet (B), pearl millet (C) and Bengal gram (D) on the functional characteristics i.e., WAC, OAC, SP, FC, FS and BD of the composite flour. Second order mixture polynomial was fitted to the data to get the regression equations. The statistical significance of the model was evaluated using model analysis, lack of fit F-value and model F-value and coefficient of determination (R²) (Weng et al., 2001). The linear, quadratic and interactive effect of variables on the response was described at 1% and 5% level of confidence.

**Optimization**

The optimization process was performed using numerical method with the Design-Expert software (version 11, Stat-EaseInc. Minneapolis, U.S.A). The objective of the optimization was to find the best combination of composite flour with desired functional properties. In the optimization process, all the independent variables and the dependent variables namely WAC, FC, FS, BD were kept in range whereas OAC was minimized and SP was maximized. These parameters are key index to measure the desirability function, a technique mostly employed in process optimization with multiple response where desirability varies from 0 (lowest) to 1 (highest)(Myers and Montgomery, 1995). The solution with the highest desirability was selected for further experiments. Response surface graphs were generated showing the effect of different independent variables on the variables of responses.

**RESULT AND DISCUSSION**

**Water absorption capacity**

Water absorption capacity (WAC) is substance’s ability to associate with water under a limited water condition (Singh et al., 2000). WAC ranges from 4.4 to 6.2 ml/g for different runs of the experimental design. WAC is highest for run 5 (FX 10%, FM 30%, PM 25%, BG 15%) which is higher than the WAC of individual flour in the blend (Table 2). This increase of WAC in the blend might be attributed to the high protein and carbohydrate having hydrophilic parts such as polar or charged side chain (Lawal and Adebowale, 2004; Edemery, 2011). The increased WAC was also observed in previous studies of composite flour (Bhupender et al., 2013; Baranwal and Sankhla, 2019; Tortoe et al., 2019). The use of legume in the composite flour might be another reason for a higher WAC (Awolu et al., 2015, 2016). The coefficient of estimation of WAC showed that as the level of individual flour is increasing WAC is also increasing but the interactive effect of foxtail millet, finger millet, pearl millet, Bengal gram with WAC is negative (Table 3). The disparity in WAC is because of the variation in protein content of the flours, their conformational characteristics and degree of interaction with water (Abah et al., 2020). Water absorption capacity is crucial in bulking and consistency of product as well as in baking applications (Edema et al., 2005). The ANOVA shows that the model (quadratic) and model terms (linear mixture, AB, AC, AD, BC, BD, CD) are significant (p < 0.05). This indicates that all the flours present in the blend have positive effect on WAC. The 3D plots showing the effect of variables on water absorption capacity is shown in figure 1(a). The R- squared and adjusted R squared values are 0.8258 and 0.6690 respectively.

**Oil absorption capacity**

Oil absorption capacity (OAC) is the property of flour which aids in oil retention. It enhances the mouth feel and holds on the flavour of food products (Aremu et al., 2007). Oil absorption capacity values ranges from 0.6 to 1.6 (ml/g) for different runs of the experimental design. OAC is highest for run 1 (FX 35%, FM 20%, PM 10%, BG 15%) and run 20 (FX 23%, FM 21%, PM 21%, BG 15%) which is higher than the individual flour (Table 2). High OAC is due to the presence of several non-polar amino acids that may bind the hydrocarbon chains of fats (Sathe et al., 1982). OAC is demonstrated by the flour protein which leads to oil
absorption in flour. These proteins expose more non-polar amino acids to the fat and enhance hydrophobicity as a result of which flours absorb oil (Awolu et al., 2016). The coefficient of estimation of OAC show that the increasing level of flour is directly proportional to OAC whereas the interactive effect of foxtail millet, finger millet, pearl millet, Bengal gram flour is negative (Table 3). The antagonistic blending is disrupted by the protein concentration, number of non-polar sites and protein-lipid-carbohydrate interactions (Zayas, 1997). The ANOVA shows that the model (quadratic) and model terms (linear mixture, AB, AC, AD, BC, BD, CD) are significant (p < 0.01). This indicates that all the flours have positive effect on OAC. The 3D plots showing the effect of variables on oil absorption capacity is depicted in figure 1(b). The R-squared and adjusted R squared values are 0.8258 and 0.6690 respectively.

Swelling power

Swelling power (SP) is attributed to the capacity of starch molecules to hold water within its structure through hydrogen bonding (Ahmad et al., 2016). SP is vital for manufacturing and structure retention of different bakery products. SP of flour is associated to presence of flexible protein molecule that tends to decrease the surface tension of water (Sathe et al., 1982). Swelling power values varies from 2.31 to 2.79 (g/g) for different runs of the experimental design. The highest Swelling power is for run 17 (FX 46.66694%, FM 10%, PM 10%, BG 13.33306%) which is higher than the swelling power of the individual flours (Table 2). Increase in swelling power has also been reported by (Ahmad et al., 2016) in their experiment on carrot pomace powder in wheat flour. The coefficient of analysis of SP shows that on increasing the level of individual flour SP increases whereas the interactive effect of foxtail millet, finger millet and Bengal gram is showing negative effect on SP (Table 3). The ANOVA shows that the model (special cubic) and model terms (linear mixture, AB, AC, AD, BC, BD, ABC, ABD, ACD, BCD) are significant (p < 0.01). This indicates that millets and Bengal gram have positive effect on swelling power. The 3D plots showing the effect of variables on swelling power is shown in figure 1(c). The R-squared and adjusted R squared values are 0.9716 and 0.9102 respectively.

Foaming capacity

Foaming capacity (FC) is attributed to the presence of proteins, which form a continuous cohesive film around the air bubbles in the foam. Proteins in the dispersion lower the surface tension at the air–water interface, thus creating foaming capacity. Foaming capacity values are from 12 to 38 (%) for different runs of the experimental design. The highest foaming capacity is for run 13 (FX 30%, FM 10%, PM 30%, BG 11.67%) which is lower than the foaming capacity of pearl millet and Bengal gram flour is higher than the finger and foxtail millet (Table 2). Similar decrease in foaming capacity was observed by (Ahmad et al., 2016). The coefficient of analysis of FC shows linear effect of each factor (Table 3). The ANOVA shows that the model (linear) and model terms (linear mixture) are significant (p < 0.01). The 3D plots showing the effect of variables on foaming capacity is represented in figure 1(d). The R-squared and adjusted R squared values are 0.6424 and 0.5753 respectively.

Foaming stability

Foaming stability (FS) values ranges from 95.38 to 100 (%). The ANOVA shows that the model (quadratic) and model terms (linear mixture, AB, AC, AD, BC, BD, CD) are not significant (p > 0.05). This indicated that there is no significant effect of any flour on foaming stability and the model is not fit.

Bulk density

The bulk density (BD) of food materials is affected by its particle size and the density of the food. Bulk density is an important factor in food packaging (Plaami, 1997). It has been shown that high BD is desirable for greater ease of dispensability and reduction of paste thickness (Amandikwa, 2012). It also determines the mixing quality of food materials. Low BD has an advantage in formulating complementary foods as it enhances nutrient and calorie density per feed of child (Akpata and Akubor, 1999). Bulk density values vary from 0.552 to 0.588 (g/ml) for the different runs of the experimental design. The highest bulk density is for run 4 (FX 28.33%, FM 30%, PM 10%, BG 11.67%), run 5 (FX 10%, FM 30%, PM 30%, BG 17%), run 13 (FX 30%, FM 10%, PM 30%, BG 10%), run 18 (FX 16.67%, FM 21.67%, PM 30%, BG 11.67%) which is higher than than the bulk density of foxtail and Bengal gram and lower than the pearl and finger millet flour (Table 2). The coefficient of analysis of BD showed linear effect of individual flour on BD (Table 3). The ANOVA shows that the model (linear) and model terms (linear mixture) are significant (p < 0.01). The 3D plots showing the effect of variables on foaming stability is represented in figure 1(f). The R-squared and adjusted R squared values are 0.5337 and 0.4463 respectively.

Optimisation of the composite flour

A desirability value of composite flour optimisation is 0.864. This desirability value corresponds to the conditions: FXM 49.65 g, FM 10 g, PM 10 g, BG 10.35 g with predicted value by the design for WAC 5.563 (ml/g), OAC 0.370 (ml/g), SP 2.722 (g/g), FC 17.563 (%), FS 97.065 (%) and BD 0.561 (g/ml) (Table 4). According to Carrera (1998), desirability value > 0.7 is considered to be excellent. Experiment is carried out with the optimal conditions of flours. The functional properties were evaluated and compared with the predicted values for the validation of the model obtained. There is no significant difference between the values predicted by the model and the real values obtained in the validation (Table 4). Therefore, the model used in each response except foaming stability resonates with the optimal level of foxtail millet, pearl millet, finger millet and Bengal gram for formulation of millet based composite flour for bakery products.

Dough characteristics

The farinograph curve obtained using Brabender is shown in figure 2. From the farinograph curve the values of farinograph parameters have been extracted characterizing the rheology of dough formed for water absorption capacity (53 %), dough development time (18 min) and dough stability (15 min) of the composite flour. The water absorption capacity of the composite flour is less than the water absorption capacity of fine wheat flour (Kohajdová et al. 2014; Mohammed et al., 2014). It is well known that gluten is responsible for water absorption in the wheat flour
and the 80% ingredients used in the composite flour are gluten-free. Hence, the reason for higher absorption ability might be a higher protein content in all flour. Composite flour reported a higher dough development time and dough stability than wheat flour (Kohajdová et al., 2014; Mohammed et al., 2014). Such a high value characterized the composite flour into the category of very high-quality flour (Munteanu et al., 2016). Dough development time and stability value are indicators of the flour strength, with higher values suggesting stronger doughs (Wang et al., 2002). Wheat flours described by bakers as “weak” reach the 500 BU mark quickly and show no stability before undergoing a considerable decline in viscosity. The “strong” flours take longer to develop, before reaching the 500 BU mark, where they remain for some time, at a good stability and then, show a minor decline in viscosity. Therefore, the composite dough made from millet, legume and wheat flour took longer time to develop and show greater stability unlike previous literature on composite flour (Nindjin et al., 2011).

**CONCLUSION**

Healthy ingredients are staircase for healthy living. The Nutritionally beneficial millet-legume based composite flour formulated in the present work could be a potential healthy prospect against the unhealthy bakery refined flour. The millet-legume based composite flour was developed with acceptable functional properties including water absorption capacity, oil absorption capacity, swelling power, foaming capacity and bulk density. Our observation for functional properties comprehends that the formulated composite flour is rich in both hydrophobic and hydrophilic protein and could fulfill the dietary requirements of diabetics and would be a healthy ingredient for food industry, in general. The dough characteristics categorise the optimized composite flour with 20% whole wheat as a high-quality strong flour.

**Table 1:** Experimental Design using Optimal Mixture Model of RSM for the Composite Flour

<table>
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<tr>
<th>S. No.</th>
<th>Dependent variables</th>
<th>Symbol</th>
<th>Minimum (%)</th>
<th>Maximum (%)</th>
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<tr>
<td>2.</td>
<td>Finger millet</td>
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<tr>
<td>3.</td>
<td>Pearl millet</td>
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</tr>
<tr>
<td>4.</td>
<td>Bengal gram</td>
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<td>10</td>
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**Table 2:** Functional Properties of the Composite Blends from Foxtail Millet, Finger Millet, Pearl Millet and Bengal Gram

<table>
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<tr>
<th>Run</th>
<th>A (%)</th>
<th>B (%)</th>
<th>C (%)</th>
<th>D (%)</th>
<th>WAC (ml/g)</th>
<th>OAC (ml/g)</th>
<th>SP (g/g)</th>
<th>FC (%)</th>
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**FLOURS**

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<th>D</th>
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Foxtail Millet (A), Finger Millet (B), Pearl Millet (C), Bengal gram (D), Water absorption capacity (WAC), Oil absorption capacity (OAC), Swelling power (SP), Foaming capacity (FC), Foaming stability (FS), Bulk density (BD)
Optimization of millet-legume based composite flour, its rheology and use as pro-health ingredient in bakery industry

Table 3: Coefficient of Analysis and ANOVA Model

<table>
<thead>
<tr>
<th>Factor</th>
<th>WAC</th>
<th>OAC</th>
<th>SP</th>
<th>FC</th>
<th>FS</th>
<th>BD</th>
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<tbody>
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<td>A</td>
<td>+5.64</td>
<td>+0.7404</td>
<td>+2.72</td>
<td>+17.54</td>
<td>+75.48</td>
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<tr>
<td>B</td>
<td>+6.26</td>
<td>+1.75</td>
<td>+3.46</td>
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<td>C</td>
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<tr>
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<td>-82.89</td>
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<tr>
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Model

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<th>Special cubic</th>
<th>Linear</th>
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<th>Linear</th>
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<td>F-value</td>
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<td>15.82</td>
<td>9.58</td>
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<tr>
<td>p-value</td>
<td>0.0268*</td>
<td>0.0080**</td>
<td>0.0014***</td>
<td>0.0007**</td>
<td>0.177</td>
<td>0.0057**</td>
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<td>Lack of fit</td>
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<tr>
<td>R²</td>
<td>0.7689</td>
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<td>0.9716</td>
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<td>0.624</td>
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</table>

ns: non-significant; * significant at 5% (p<0.05); ** significant at 1 % (p<0.01)

Table 4: Comparison of Observed and Predicted Values of Optimized Composite Flour

<table>
<thead>
<tr>
<th>Response</th>
<th>Predicted value</th>
<th>Actual value ± SD</th>
<th>Standard error</th>
<th>Mean difference</th>
<th>Sig (two tailed)</th>
</tr>
</thead>
<tbody>
<tr>
<td>WAC</td>
<td>4.712a</td>
<td>4.382 ± 0.231a</td>
<td>0.133</td>
<td>-0.33</td>
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<tr>
<td>OAC</td>
<td>0.997a</td>
<td>0.9497 ± 0.027a</td>
<td>0.0157</td>
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<tr>
<td>SP</td>
<td>2.369a</td>
<td>2.265 ± 0.256a</td>
<td>0.148</td>
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<td>FC</td>
<td>29.419a</td>
<td>29.452 ± 0.381a</td>
<td>0.219</td>
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</tr>
<tr>
<td>BD</td>
<td>0.575a</td>
<td>0.546 ± 0.039a</td>
<td>0.0229</td>
<td>-0.028</td>
<td>0.342</td>
</tr>
</tbody>
</table>

Means ± standard deviation; means followed by same superscript in each row are not significantly different (p > 0.05)
WAC water absorption capacity; OAC oil absorption capacity; SP swelling power; FC foaming capacity; BD bulk density

Fig. 1: Response surface plots showing the effect of Foxtail millet, Finger millet, Pearl Millet and Bengal gram on functional properties of flour. (a) Effect of composite flour on water absorption capacity (WAC) (b) Effect of composite flour on oil absorption capacity (OAC) (c) Effect of composite flour on swelling power (SP) (d) Effect of composite flour on foaming capacity (FC) (e) Effect of composite flour on foaming stability (FS) (f) Effect of composite flour on bulk density (BD).
Acknowledgement

The authors duly acknowledge Er. Saumya Choudhary, Scientist B, ICMR-National Institute of Pathology, New Delhi for editing the manuscript draft.

REFERENCES


