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Effect of partial substitution of wheat flour with millet-based gluten free composite flour on the quality of cookies

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Abstract

Effects of partial replacement (20, 40, 60, 80 and 100%) of whole wheat flour by composite flour (based on foxtail millet, proso millet and buckwheat flours in equal proportions) on physical, nutritional, sensory and antioxidant properties of multi-millet cookies were investigated. An increase in the ratio of composite flour in the blend and formulated cookies resulted in an increase in ash, fiber, fat and protein and a decrease in moisture and carbohydrate contents. Increase in supplementation levels of composite flour added to the TPC (Total Phenolic Content), DPPH (2,2-diphenyl-1-picrylhydrazyl), reducing power and FRAP (Ferric Reducing Antioxidant Power) and decrease in metal chelating activity of cookies. Maximum gain in thickness and weight and loss in diameter, spread ratio and bake loss were observed with the highest composite flour substitution. Color values such as a* was found to be enhanced whereas L* and b* were found to be weakened, simultaneously NEB (Non-Enzymatic Browning) values got increased upon substitution. A 60% blend of composite flour with whole wheat flour was selected best used in the cookie's formulation based on of sensory evaluation. Overall, this study demonstrates that millet based composite flour can effectively improve the functional values of formulated cookies, in addition to an option for the developing of gluten free products for celiac patients.

Keywords: Foxtail millet, proso millet, buckwheat, multi-millet cookies, gluten free.

Introduction

It is evident from various researches that coarse cereals, pseudo-cereals and by-products are nutritionally superior to staple cereals such as rice and wheat which make them suitable for industrial-scale utilization for various food preparations like weaning food, snack food, composite dairy mix, and other dietary products. Possession of phenolic compounds ensures their potential health benefits. Foxtail millet (*Setaria italica*) is one of them that work to enrich prepared food

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products with its dietary fiber, divalent cation minerals (Ca^{2+}, Mg^{2+}, and Zn^{2+}) with low glycemic index attributes (Kim *et al*. 2011). It is high in polyphenolics which prevents cardiovascular disease, and elevates our immune system (Marak *et al*. 2019). Proso millet nutritive profile is also comparable to common staples even exceeding in regard to protein, crude fiber and phenolics (Amadou *et al*. 2013). It has attracted attention for its considerable health benefits, particularly anti-cancer, anti-diabetic, antiproliferative effects and anti-liver injury. Studies have shown that the extracts of proso millet possess antioxidant properties (Chandrasekara and Shahidi, 2012). Similarly, pseudo-millets are the key targets of researchers especially buckwheat *(Fagopyrum esculentum*) having prime importance owing to its high protein uniqueness with favorable amino acids, high dietary fiber and almost all vitamins, essential minerals, and trace elements. Buckwheat possesses health benefits like reduction of high blood pressure, blood sugar control, lower blood cholesterol, prevention of fat accumulation, constipation, colon carcinogenesis and mammary carcinogenesis, strengthen capillary blood vessels and suppresses plasma cholesterol and gallstone formation (Sedej *et al*. 2011). Another functional property of buckwheat is its gluten-free characteristics which make it a promising diet for patients suffering from celiac disease (Chopra *et al.* 2014).

Development of bakery products such as biscuits and cookies based upon the fortification of composite flour is the recent trend in the bakery industry (Hussain and Kaul, 2018: Hussain *et al.* (2018a). Research studies incorporating millets for the formulation of bakery products have been reported earlier. The foxtail millet (20, 30, and 40%) substituted wheat flours were studied and compared to the control (100% wheat flour) (Marak *et al*. 2019). The increase in incorporation levels enhanced the functional characteristics, phenolic contents and antioxidant capacities (DPPH and ABTS scavenging potentials) of the composite flour as well as the cookies developed. Sensory evaluation disclosed that cookies developed from 30% foxtail millet had comparable scores with that of control. Rice flour based gluten-free cookies prepared by blending malted foxtail millet, proso millet and buckwheat flours at 15 % and 30 % levels was investigated by Kumari *et al*. (2023). The protein, fat, ash, total dietry fiber and in vitro protein digestibility of developed cookies increased whereas glycemic and starch hydrolysis indices decreased upon increase in substitution levels of malted flours of all three grains. But till date studies having wheat based composite flour comprised of a blend of foxtail millet, proso millet, and buckwheat flours for cookies formulation are not reported. Since the selected crops are gluten-free, thus replacing wheat flour with composite flour-based on these crops will dilute the wheat gluten proteins, and thereby help in preventing celiac disorders. In addition to that, the developed product will impart enhanced nutritional and health benefits due to the presence of ample quantity of essential nutrients and health promoting constituents in the composite flour based on the selected raw materials. Various chronic diseases which are prevalent these days can be managed by the consumption of such products. Since the selected crops at this stage are under under-utilization, this study will help in their revival. The objective of the present study is to compare the functional properties of the millet-based composite flour with wheat flour, to utilize it at different levels for cookie making, and to assess the nutritional characteristics and organoleptic attributes of the formulated cookies.

Materials and methods

Foxtail millet, proso millet and buckwheat flour was procured from farmers of Garkhon and Skurbuchan villages of Ladakh, India. Whole wheat flour was procured from farmers of Matho village. The flours so obtained were sieved through a 40 mm sieve. Ghee, baking powder, refined sugar, and corn flour were procured from the local market (Leh, India). The experiments were conducted in the Food Processing Laboratory of HMAARI, SKUAST-K, Leh, India and part of the quality analysis was done in Division of Food Science and Technology, SKUAST-K, Kashmir, India.

Preparation of flour blends

Blends based upon the composite flour were made by taking whole wheat flour as control. The composite flour was obtained by mixing buckwheat, foxtail millet and proso millet flours in equal proportions. Blends were prepared by mixing composite flour with wheat flour in the proportions of 100:0, 80:20, 60:40, 40:60, 20:80, and 0:100, which correspond to 0, 20, 40,60, 80, and 100 % blends.

Preparation of cookies

The multi-millet cookies formula based on flour weight was: 100 g (32.05%) flour, 20 g (6.41%) icing sugar, 40 g (12.82%) ghee, 1 g (0.32%) baking powder, 1 g (0.32%) corn flour and 150 ml (48.07%) water. The dough was sheeted to a thickness of 0.5 cm with a rolling pin. The cookies were cut round in shape with a cookie die of diameter 5 cm and transferred to a tray lined with aluminum foil and were baked at 180 °C for 15 min in an electric oven. The cookies were cooled to room temperature and packed in airtight containers.

Functional properties

Water Absorption Capacity (WAC) and Oil Absorption Capacity (OAC)

The WAC and OAC of flour samples were determined as per the method described by Jan *et al.* (2015) with slight modifications. The flour samples (1 g each) were mixed with 10 ml distilled water or refined mustard oil in test tubes and were kept at ambient temperature for 30 min and then centrifuged for 10 min at 2000 rpm. After centrifugation, the aqueous supernatant or clear oil obtained was decanted. The test tubes were allowed to drain for 5 min on a paper towel and followed by weighing of the residue. The WAC/OAC are expressed in g of water/oil per g of dry weight of the sample.

Foaming Capacity (FC) and Foam Stability (FS)

The determination of FC and FS was done by the method given by Jan *et al.* (2015). Exactly 2 g of flour samples were mixed with 50 ml of distilled water at 30±2 °C in a 100 ml graduated measuring cylinder. The suspension was shaken properly by whipping for 3 min in an electrical blender to form foam. The content was then transferred into the measuring cylinder and the volume of the foam was recorded after 30 seconds.

FC (%) = (Volume after whipping - Volume before whipping) x 100/ Volume before whipping

The foam volume was recorded 1 h after whipping to determine the FS as a percentage of the initial foam volume.

FS (%) = (Foam volume after standing time/Initial foam volume) x 100

Swelling Power (SP)

The method described by Gani *et al*. (2013) with slight modifications was followed to determine the SP. Briefly, 3 g of each sample was mixed with 30 ml distilled water in a centrifuge tube and heated at 70°C in a water bath for 30 minutes. The suspension after heating was centrifuged at 1000 rpm for 15 min. The supernatant was decanted and the residue was weighed.

SP = Weight of the residue - Weight of dry flour

Bulk Density (BD)

The flour samples (100 g each) after tare were gently filled in a 500 ml graduated cylinder. The bottom of the cylinder was tapped gently for several times until there was no further diminution of the sample level after filling to 500 ml mark.

BD (g/ml) = Weight of flour (g)/Volume taken by flour (ml)

Pasting properties

Pasting properties of flour were determined using a rapid visco analyser and calculated through the pasting curve using Thermocline version 2.4 software (RVA-4, Newport Scientific Pty Ltd., Warriewood, Australia). 3.5 g of flour sample (moisture = 14%) was transferred into a canister and approximately 25 ± 0.1 ml distilled water was added (corrected to compensate for 14% moisture basis). After heating the slurry at 50°C, it was stirred at 160 rpm for 10 seconds for thorough dispersion. The slurry was held at 50°C for up to 1 min, and then heated to 95°C over 7.3 min and held at 95°C for 5 min, and finally cooled to 50°C over 7.7 min. The parameters determined were peak viscosity, trough viscosity, break down viscosity, final viscosity, set back viscosity, pasting time and pasting temperature.

Proximate composition

The proximate composition of flours and blends were determined according to the methods of AOAC (2016). Crude protein was estimated according to Kjeldhal method (AOAC 992.23). The nitrogen conversion factor taken for crude protein was 5.83. Moisture was determined by hot air oven method (AOAC 952.08), ash by gravimetric method (AOAC 985.29), total fat by acid hydrolysis method (AOAC 948.15) and dietry fiber by enzymatic gravimetric method (AOAC 930.30). The carbohydrate (non-fiber) content was calculated using difference method by subtracting the contents of moisture, ash, fat, fiber and protein from 100 % of dry matter.

Antioxidant properties

Preparation of extracts

Each sample (0.3 g) was dissolved in 20 ml of 70% methanol, stirred for 2 hours on a magnetic stirrer followed by a centrifugation for 10 minutes a t3500 g. The supernatant was evaporated at 40 °C and the supernatant was stored at -18 °C.

Total Phenolic Content (TPC)

The Folin-Ciocateu spectrophotometric method given by Jan *et al.* (2015) with some modifications was followed for the determination of TPC. The results were expressed as Gallic acid equivalents (nmolGAE) per µg of dried weight of sample.

DPPH radical scavenging activity

DPPH (2,2-diphenyl-1-picrylhydrazyl) radical scavenging activity of the extract solutions (Methanolic) of each sample was determined according to the method described by Shah *et al.* **(**2015). After incubation in dark for 30 minutes, the absorbance at 517 nm was measured after each sample solution.

Inhibition (%) = $A_{control}$ - A_{sample} /$A_{control}$ x 100

Where $A_{control}$ indicates the absorbance of the control and A_{sample} indicates the absorbance of the extract.

Reducing power

The reducing power of methanol extracts were determined by the method described by Gani *et al.* (2013) with some modifications. Different volumes of extracts of six samples were mixed with 0.2 M sodium phosphate buffer pH 6.6 (2.5 ml) and 1 % (w/v) of aqueous potassium ferricyanide (2.5 ml). The mixture so obtained was incubated at 50 °C for 20 min. 10 % (w/v) trichloroacetic acid (2.5 ml) was added to the mixture, which was then centrifuged at 3000 g for 10 minutes. After centrifugation, 2.5 ml supernatant was collected and diluted with 2.5 ml of deionized water and 0.1 % (w/v) ferric chloride 0.5 ml was added. The absorbance was recorded at 700 nm against methanol (control) and compared with ascorbic acid which was used as a standard. A higher absorbance shows a higher reducing power.

Reduction (%) = (1−A_C - A_S) x 100

Where, A_C absorbance of standard

A_S absorbance of the sample

FRAP (Ferric Reducing Antioxidant Power) value

The method described by Jan *et al.* (2015) was followed for its estimation. 3 ml of the FRAP reagent [10 ml of acetate buffer (300 mM, PH 3.6), 1 ml TPTZ (10 mM) in hydrochloric acid solution (40 mM) and 1 ml of $FeCl_3$ solution (20 mM)] was added to 0.1 ml of each sample extract.

After incubation for 4 minutes at room temperature, absorbance was taken at 593 nm immediately and the results were expressed as μM Fe^{2+} /L

FRAP = Change in absorbance of sample from 0-4 minutes/ Change in absorbance of standard (ascorbic acid) from 0-4 minutes x 2

Where 2 indicated the FRAP value of ascorbic acid.

Metal chelating activity

The metal chelating activity of samples was determined as described by Jan *et al.* (2015) which was calculated as follows:

Iron (Fe^{2+}) chelating activity (%) = 1− (A_{S562}/A_{C562}) x 100

Where, A_{S562} Absorbance of the sample at 562 nm

A_{C562} Absorbance of the control at 562 nm

Physical characteristics

The thickness of the cookies was measured by a Vernier Caliper and the diameter was determined using a measuring scale placing them edge-to-edge. From the measurements taken, the spread ratio (W/T) was calculated, where W is the diameter and T is the thickness of the cookies.

Color analysis

The color was measured using a Hunter Lab Colorimeter, (Color Flex Reston, Virginia, USA). Color values were measured as L*, a*, and b*. Each value represents the average of four measurements at different points of the cookies. An L* value is the lightness, while as a* and b* values are the chromaticity values that indicate redness and yellowness, respectively.

Non-enzymatic browning index (NEB)

The NEB of the samples was determined as previously described by Sharma and Gujral (2014). The browning index (ΔA) was calculated as:

ΔA = Absorbance at 420 nm - Absorbance at 550 nm

Sensory evaluation

A panel of 20 judges (semi-trained panelists) from the scientific staff members of HMAARI, SKUAST-K, Leh, India and students of the FoA, SKUAST-K, Kashmir, India was selected after explaining different terminologies, characteristics of good quality cookies, score-sheet, and methods of scoring used in the sensory evaluation. The criteria for selection of judges include availability, capability and reliability. The samples were evaluated on the basis of color, texture, taste, flavour and overall acceptability using 9-point Hedonic Scale assigning scores 9-like extremely to 1-dislike extremely. A score of 5.5 and above was considered acceptable.

Texture analysis

The textural properties of the products were investigated using a Texture analyzer (TA XP plus Exponent Stable MicroSystem, Brookfield, USA). A 3-point Bending Rig and 5 kg load cell were used to measure the fracture strength. The distance between the two beams was 7 mm. Another identical beam was brought down from above at a pre-test speed of 1.0 mm/sec, test speed of 3.0 mm/sec, and post-test speed of 10.0 mm/sec which was continued until the cookie broke. The peak force was considered as the fracture strength.

Statistical analysis

The data obtained from experiments performed in triplicates were subjected to one-way ANOVA and Duncan test by SPSS (version 27.0.1).

Results and discussion

Although it was difficult to sheet the dough, cookies were developed using 100% composite flour with extra effort. However, this flour resulted in relatively fragile and darkest cookies among the other cookie samples studied (Fig. 1).

Functional properties of flour

The WAC of composite flour was significantly ($p \leq 0.05$) higher than that of wheat flour (Table-1). Variations in the WAC between both the flour might be responsible due to the difference in protein structure and the presence of different hydrophilic carbohydrates (Kaur *et al.* 2014). The OAC of composite flour was significantly lower than that of wheat flour and these variations may be explained by the variation in the presence of hydrophobic amino acids having an affinity for the hydrocarbon side chain of the oil. Similar results were observed by Mohamed *et al.* (2009). The SC of composite flour was lower than that of wheat flour which corroborated with the results of Poongodi Vijayakumar *et al.* (2010). The SC of the flour depends upon particle size, types of variety, and processing methods used to obtain flour. The difference may also be attributed to the high level of gluten content in wheat flour which helps it in the retention of water, and increase in volume by absorption of water. The FC and FS of 100% composite flour were found significantly higher than that of 100% wheat flour. The results of FC are in line with those found by Kaur *et al.* (2014). FC is considered to be dependent upon the configuration of protein molecules. Flexible proteins have a good FC but the reverse is true for highly ordered globular molecules (Baljeet *et al.* 2010). Composite flour having 100% mixed millet flour had the maximum whereas, the control reported minimum BD's. Present results are in line with the findings of Poongodi Vijayakumar *et al.* (2010).

Pasting properties of flour

Pasting properties are important factors in determining the applications of flours for baking purposes. The results obtained (Table-2) revealed significant differences in the pasting profiles of flour blends of millets. As the proportion of composite flour increased, the peak, trough, breakdown final viscosity, pasting time and pasting temperature decreased while as setback viscosity increased in flour blends of composite flour from F_1 (0% composite flour) to F_6 (60% composite flour). The pasting properties depend upon the amount of starch content, amylase activity, amylose/amylopectin ratio, protein and lipid content present in the flours. Anberbir *et al*. (2024) reported significant differences between the different types of viscosities in teff, pearl millet and buckwheat composite flour.

Proximate composition of flour and cookies

The proximate composition as given in Table-3. The moisture content is partially affected by the fiber content in a sample. However, in our study blended flour had lower moisture content as compared to wheat flour despite having higher fiber content with a similar trend in its products. This might be due to the higher fat content which reduces moisture in the blended flour (Sreerama *et al*. 2010). Hussain *et al.* (2018b) also found similar results during multigrain biscuit formulation. Ash, fiber, fat and fiber of flour increased while carbohydrate decreased upon blending. Cookies with composite flour showed higher ash as compared to that with wheat flour. The higher ash content may be due to the high mineral content in the former. The analyzed formulated cookies had dietary fiber ranging from and 3.26 to 7.84% with cookies formulated from composite flour had higher fiber content. Chhavi and Savita (2012) observed similar trend in finger millet incorporated bread. All the cookies formulated contained high fat content ranging from 19.81% to 25.27% which is mainly due to the use of ghee in the formulation of these cookies. Increased total fat content in flour decreases its shelf life but improves binding ability, flavor retention capacity and reduces moisture in the final product (Sreerama *et al*. 2010). The formulated cookies had crude protein content ranging from 8.19 to 9.79% which may be explained by the fact that the composite flour contain comparatively higher protein. The carbohydrate content of cookies ranged between 51.34 and 63.59%. The carbohydrate levels decreased upon the supplementation which could be attributed to the higher fraction of other constituents (moisture, protein, ash, fiber and fat content) present in the composite flour. This trend is also probably because wheat flour contains endosperm as major portion which is a good source of carbohydrates.

Antioxidant properties of flour and cookies

The blended flour as well as the cookies formulated by replacing whole wheat flour with composite flour exhibited significantly ($p<0.05$) high antioxidant activities for most of the investigated properties when compared with the control samples (Table-3).

Total phenolic content

An increasing trend in the formulated cookies was observed upon increase in the ratio of composite flour. Higher TPC was observed in 100% composite flour (12.07nmolGAE/µg) and cookies prepared with 100% composite flour (7.65 nmol GAE/µg) as compared to their controls. Other researchers (Chen *et al.* 2021) revealed increasing trend in TPC upon the substitution of finger millet flour in rice and wheat-based food products. Sharma and Gujral (2014) suggested that a continuous baking process could modify the chemical structure of a phenolic compound, specifically due to polymerization, and consequently reduce extractable polyphenols in baked products.

DPPH scavenging potential

DPPH radical scavenging activity was higher in composite blends than in wheat flour and their formulated products as consequence of higher polyphenolic content in composite flour. The examined extracts of composite blends possess radical inhibitors or scavengers with the possibility to act as primary antioxidants. Similar results were found by Sedej *et al.* (2011) for milling fractions of wheat and buckwheat. The extracts might react with free radicals (peroxy radicals) which are the major contributors of the auto-oxidation chain of fat, thereby culminating the chain reaction (Sharma and Gujral, 2014). Baking led to an increase in the scavenging activity of radicals in cookies which has earlier been reported by Sharma and Gujral (2014). The possible reason behind this might be the formation of brown pigments melanoidins, the products of maillard reaction, which takes place during the process of baking.

Reducing power

Better reducing power was found in composite flour as well as cookies with 100% composite flour than their respective controls thus showing increasing trend on increase in blending ratios. Incorporation of buckwheat and rye flour to wheat flour for bread preparation led to the increase in reducing power on baking (Filipcev *et al*. 2011). The reducing power of an antioxidant compound is because of the presence of reductones and their antioxidant capacity is due to the breaking of the free radical chain reaction by donating a hydrogen atom and preventing peroxide formation. Baking of cookies led to an increase in reducing power. The products which were generated during the baking of cookies due to maillard reaction, might resulted in increasing the reducing power (Sharma and Gujral, 2014).

FRAP value

FRAP value of flour blends increased with the incorporation of composite flour. FRAP was found to be lowest for wheat flour-based cookies (control) and it increased as the proportion of composite flour increased in the blends. These results are in accordance with that of Jayawardana *et al*. (2022) in finger millet blended biscuits. The polyphenolic content of composite flour as well as supplemented cookies is responsible for their ferric ions reducing ability.

Metal chelating activity

The metal chelating activity was significantly higher for wheat flour extracts than for composite flour extracts as well as their respective cookies. Although the methanolic extracts of wheat flour exhibited more potent chelating activity on Fe^{2+} than the millet extracts despite rutin content of millets possesses more structural features than ferulic acid for complexing metal ions. Lower antioxidative capacity of rutin as metal chelator in comparison to ferulic acid (and other hydroxycinnamic acids from wheat flour) might be due to steric hindrance of the sugar moiety of rutin (Sedej *et al*. 2011). The presence of ferulic acid, a high potent chelating component in wheat flour extract might be responsible for its higher chelating capacity than composite flour extract.

Physical properties of cookies

Since appearance is used to evaluate a food product before the taste, physical characteristics are equally important determinants of consumer acceptability and marketability. The physical properties of cookies prepared from blended flour and wheat flour are shown in Table-4. The diameters of cookies made from composite flour were found significantly lower than 100% wheat flour. The thickness of cookies ranged between 0.75 to 0.81 cm, which increased with increase in the level of composite flour incorporations. Increase in thickness might be due to the decrease in diameter on addition of composite flour content in cookie dough. The changes in diameter and thickness were reflected in spread ratio. Spread ratio decreased with the addition of composite flour. Other workers, Sharma and Gujral (2014) and Kaur *et al.* (2014) obtained similar results in barley and buckwheat blended cookies, respectively. Reduced spread ratios of composite flour fortified cookies were attributed to the fact that composite flours form aggregates with increased numbers of hydrophilic sites available that compete for the limited free water in biscuit dough. The thicknesses and spread ratios of the cookies were inversely proportional to the level of millet based composite flour. The weight of cookies increased as the concentration of composite flour increased in the blends. The range of cookie weight was10.30 g to 11.35 g with maximum value in 100% composite flour cookies. The results are in agreement with the that of Chauhan *et al*. (2016) which they have attributed to the high water-holding capacity of amaranth flour when compared to wheat flour. The decrease in bake loss of cookies with the increase in the level of composite flour can be attributed to the ability of their dietary fiber to retain more water.

Color analysis of cookies

Composite flour incorporation significantly ($p<0.05$) affected the colors of the cookies. The surface color of the cookie decreased for L* and b* as the composite content increased in the blends (Table-4). While as, increase in a* was observed with an increase in composite flour content in the cookies. The decrease of L* and b* values as well as the increase of a* values indicated that the darkness of incorporated biscuits was proportional to the level of composite flour. The crumb color of barnyard millet-wheat composite flour bread (Singh *et al*. 2012) and finger millet supplemented wheat biscuits Jayawardana *et al*. (2022) was reported to be lower as compared to their controls. Maillard browning and caramelization of sugar in addition to oxidation of phenolic compounds considered to produce brown pigments during baking (Sharma and Gujral, 2014).

Non-enzymatic browning index of cookies

The NEB index of pure wheat and pure blended cookies was observed to be 0.008 and 0.024, respectively (Table-4). Incorporation led to a significant increase in non-enzymatic browning index which might be due to possible dilution of the sugars and proteins of the wheat flour upon incorporation of composite flour. Sharma and Gujral (2014) and Jan *et al*. (2015) reported a significant increase in browning upon baking of *chapati* and cookies, respectively, which corroborate with our results. Increase in browning is associated with Maillard browning, caramelization of sugar occurring during baking of cookies. (Sharma and Gujral, 2014).

Texture profile analysis of cookies

The hardness of the cookies is determined from curve shown in Fig. 2(a). Once the trigger force is attained, the force is seen to increase until the cookie fractures and falls into two pieces. This is observed as the maximum force and can be referred to as the 'hardness' of the sample. The distance at the point of break is the resistance of the sample to bend and so relates to the 'fracturability' of the sample i.e., a sample that breaks at a very short distance has a high fracturability. The cookies made from 100% composite flour were found to be the least hard and most fragile thus hardness of cookies decreased with the incorporation of composite flour. The hardness of wheat flour cookies (100%) and composite flour cookies (100%) were 6952.48 g and 2987.25 g, respectively. The order of the hardness was found to be as follows; $C_1 > C_2 > C_3 > C_4 = C_5 < C_6$. The values of hardness and fracturability are given in Table 4. Baljeet *et al*. (2010) found similar results for buckwheat biscuits. The addition of gluten free component (composite flour) to flour blends may be responsible for the softer cookies.

Sensory attributes of cookies

From the results, Fig. 2(b), it was observed that most of the sensory score of the blended flour cookies displayed the decreasing trend from C_1 to C_6 samples because of increasing substitution levels of composite flour. The sensory scores for color, texture and appearance saw a decline, simultaneously a rise in scores for taste and flavor in cookies formulated using composite flour was observed. Our results in terms of appearance and texture are also in accordance with that of Poongodi Vijayakumar *et al.* (2010). The composite flour had a dark colour, due to its high phenolics, ash content and sandiness, which might have contributed to the reduction in panel sensory perceptions and hence decreased the scores for sensory attributes like colour, texture and appearance. Other studies of millet have also found that, as the amount of millet addition increases, sensory scores decrease (Shimray *et al*. 2012). Most of the sensory attributes were significantly affected however the decline in the scores was not very high due to supplementation. The sensory results indicated that the inclusion of 60% millet based composite flour seems to be the most preferred amount to add to a cookie.

Conclusions

From our study, it can be concluded that addition of blended flour (foxtail millet, proso millet and buckwheat) to whole wheat flour enhances the protein, fiber, fat, ash, total phenolic content and antioxidant properties of the multi-millet cookies. The Overall acceptability of cookies was highest at the 60% level of blending. This study may provide ample scope for utilization of these under-utilized gluten free crops in other food products. This will also be contributing step to the celebration of International Year of Millets recognized by FAO and UNO for the year 2023.

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Table-1: Functional properties of flour.

Sample	WAC (g/g)	OAC (g/g)	SC (%)	FC (%)	FS (%)	BD (g/mL)
F_1	1.20^{a}±1.76	1.88^{b}±1.43	5.78^{f}±0.09	09.42^{a}±0.29	52.12^{a}±0.94	0.72^{a}±0.05
F_2	1.34^{a}±2.19	1.67^{b}±1.45	5.31^{e}±1.07	09.39^{a}±0.11	54.61^{b}±0.63	0.75^{b}±0.02
F_3	1.53^{a}±1.08	1.34^{a}±2.76	5.13^{d}±0.31	09.16^{a}±0.09	60.51^{c}±1.43	0.80^{c}±0.11
F_4	1.61^{a}±0.76	1.19^{a}±2.04	4.93^{c}±0.04	13.39^{b}±0.45	62.13^{d}±0.32	0.87^{d}±0.07
F_5	1.67^{a}±1.45	1.14^{a}±0.09	4.38^{b}±2.06	21.64^{c}±0.37	64.41^{e}±0.41	0.94^{d}±0.09
F_6	1.70^{a}±0.37	1.09^{a}±0.98	3.89^{a}±0.09	30.38^{d}±0.34	70.62^{f}±0.14	1.02^{d}±0.05

The values with different superscript in a column differ significantly ($p \leq 0.05$). F_1, F_2, F_3, F_4, F_5 and F_6 refer to 0, 20, 40, 60, 80, 100% composite flour, respectively.

WAC: Water Absorption Capacity, OAC: Oil Absorption Capacity, SC: Swelling Capacity, FC: Foaming Capacity, FS: Foaming Stability, BD: Bulk Density.

Table 2: Pasting properties of flour.

Sample	Peak Viscosity (cp.)	Trough Viscosity (cp.)	Breakdown Viscosity (cp.)	Final Viscosity (cp.)	Setback Viscosity (cp.)	Pasting time (min)	Pasting Temperature (°C)
F_1	2341^{e}±35.14	1636^{d}±41.88	971^{f}±0.09	2982^{e}±24.63	1352^{a}±0.94	6.702^{d}±0.08	85.72^{c}±0.61
F_2	2218^{d}±28.38	1670^{d}±9.64	834^{e}±1.07	2911^{e}±41.39	1461^{c}±2.63	5.175^{b}±0.07	85.56^{c} ±0.88
F_3	1975^{c}±10.50	1541^{c}±1.15	619^{d}±49.31	2816^{d}±31.09	1405^{b}±7.43	5.380^{c}±0.11	84.28^{b} ±1.91
F_4	1580^{b}±31.95	1216^{b}±26.02	493^{c}±10.04	2539^{c}±0.45	1413^{b}±0.32	5.287^{c}±0.07	84.62^{b} ±5.73
F_5	1406^{b}±16.18	1288^{b}±33.19	339^{b}±2.06	2464^{b}±22.17	1644^{d}±82.41	5.094^{a}±0.09	83.42^{a} ±1.19
F_6	1153^{a}±12.27	1024^{a}±19.16	173^{a}±19.89	2309^{a}±1.34	1770^{e}±11.14	5.028^{a}±0.05	84.19^{b}±0.48

The values with different superscript in a column differ significantly ($p \leq 0.05$). F_1, F_2, F_3, F_4, F_5 and F_6 refer to 0, 20, 40, 60, 80, 100% composite flour, respectively.

Table 3: Proximate composition and antioxidant properties of flour and cookies.

Sample	Proximate composition (%)						Antioxidant properties				
	Moisture content	Ash	Fiber	Fat	Protein	Non-fiber carbohydrate	TPC (nmolGAE/ μg)	DPPH Inhibition (%)	Reducing power (%)	FRAP (μM Fe^{2+}/L)	Metal chelating activity (%)
F_1	$13.08^{a}\pm0.09$	$1.69^{e}\pm0.06$	$2.62^{f}\pm0.06$	$01.75^{d}\pm0.01$	$10.14^{c}\pm0.15$	$70.72^{a}\pm0.07$	$07.67^{f}\pm0.01$	$38.19^{e}\pm0.01$	$33.15^{f}\pm1.03$	$2.08^{d}\pm0.10$	$52.51^{a}\pm8.5$
F_2	$12.77^{b}\pm0.02$	$1.93^{d}\pm0.01$	$3.13^{e}\pm0.01$	$01.96^{d}\pm0.07$	$10.15^{c}\pm0.15$	$70.06^{a}\pm0.11$	$08.08^{e}\pm0.01$	$44.26^{d}\pm0.05$	$37.30^{e}\pm1.60$	$2.08^{d}\pm0.24$	$30.06^{b}\pm1.63$
F_3	$12.26^{c}\pm0.08$	$2.34^{c}\pm0.02$	$3.66^{d}\pm0.01$	$02.37^{c}\pm0.03$	$11.23^{b}\pm0.04$	$68.14^{b}\pm0.27$	$08.67^{d}\pm0.02$	$48.31^{c}\pm0.04$	$41.07^{d}\pm1.48$	$2.23^{c}\pm0.13$	$28.61^{c}\pm7.20$
F_4	$11.88^{d}\pm0.04$	$2.63^{b}\pm0.07$	$4.34^{c}\pm0.01$	$02.78^{c}\pm0.04$	$11.25^{b}\pm0.22$	$67.12^{c}\pm0.08$	$09.75^{c}\pm0.01$	$52.16^{b}\pm0.07$	$43.18^{c}\pm1.56$	$2.30^{c}\pm0.09$	$13.23^{d}\pm3.00$
F_5	$10.38^{e}\pm0.25$	$3.39^{a}\pm0.02$	$5.72^{b}\pm0.05$	$03.79^{b}\pm0.05$	$11.57^{a}\pm0.03$	$64.11^{d}\pm0.01$	$10.64^{b}\pm0.04$	$56.87^{a}\pm0.02$	$44.48^{b}\pm3.97$	$2.56^{b}\pm0.19$	$05.34^{e}\pm0.45$
F_6	$10.13^{e}\pm0.05$	$3.40^{a}\pm0.08$	$6.71^{a}\pm0.01$	$04.81^{a}\pm0.01$	$11.68^{a}\pm0.07$	$62.27^{e}\pm0.07$	$12.07^{a}\pm0.14$	$56.91^{a}\pm0.03$	$45.40^{a}\pm1.14$	$2.86^{a}\pm0.01$	$03.35^{f}\pm1.92$
C_1	$06.76^{e}\pm1.14$	$1.26^{a}\pm0.08$	$3.26^{a}\pm0.07$	$19.81^{a}\pm0.32$	$08.19^{a}\pm0.10$	$63.59^{e}\pm0.03$	$06.58^{a}\pm0.09$	$40.61^{a}\pm0.09$	$30.08^{a}\pm0.91$	$2.09^{a}\pm0.06$	$60.09^{e}\pm1.51$
C_2	$06.23^{d}\pm0.68$	$1.55^{b}\pm0.05$	$4.29^{b}\pm0.03$	$20.27^{b}\pm0.02$	$08.30^{a}\pm0.07$	$61.94^{d}\pm0.06$	$06.71^{a}\pm0.10$	$55.53^{b}\pm0.08$	$37.16^{b}\pm0.77$	$2.16^{b}\pm0.01$	$52.54^{d}\pm8.52$
C_3	$05.67^{c}\pm0.03$	$1.63^{c}\pm0.01$	$5.94^{c}\pm0.04$	$21.86^{c}\pm0.19$	$08.62^{b}\pm0.16$	$58.52^{c}\pm0.16$	$06.85^{a}\pm0.01$	$57.18^{c}\pm0.11$	$45.50^{c}\pm0.91$	$2.18^{b}\pm0.06$	$49.08^{c}\pm1.45$
C_4	$04.19^{b}\pm0.05$	$1.69^{d}\pm0.01$	$6.26^{d}\pm0.04$	$23.17^{d}\pm0.21$	$08.71^{b}\pm0.10$	$57.01^{c}\pm0.27$	$07.21^{b}\pm0.33$	$61.65^{d}\pm0.09$	$49.82^{d}\pm4.14$	$2.61^{d}\pm0.05$	$32.47^{b}\pm1.87$
C_5	$03.56^{a}\pm0.07$	$1.73^{e}\pm0.02$	$7.51^{e}\pm0.02$	$24.98^{e}\pm0.09$	$09.20^{c}\pm0.19$	$53.51^{b}\pm0.17$	$07.53^{c}\pm0.19$	$64.79^{e}\pm0.02$	$63.41^{e}\pm2.87$	$2.64^{d}\pm0.04$	$24.68^{a}\pm1.19$
C_6	$03.09^{a}\pm0.52$	$2.72^{i}\pm0.03$	$7.84^{e}\pm0.08$	$25.27^{f}\pm0.04$	$09.79^{d}\pm0.15$	$51.34^{a}\pm0.04$	$07.65^{c}\pm0.06$	$67.81^{f}\pm0.10$	$65.09^{e}\pm0.39$	$3.76^{f}\pm0.40$	$23.01^{a}\pm2.00$

The values with different superscript in a column differ significantly ($p\leq 0.05$). F_1, F_2, F_3, F_4, F_5 and F_6 refer to 0, 20, 40, 60, 80, 100% composite flour, respectively and C_1, C_2, C_3, C_4, C_5 and C_6 refer to cookies made from 0, 20, 40, 60, 80, 100% composite flour, respectively.

Table 4: Physical properties, color values, NEB and textural properties of cookies.

Sample	Diameter (cm)	Thickness (cm)	Spread ratio	Weight (g)	Bake loss (%)	L*	a*	b*	NEB	Hardness (g) force^{-1}	Fracturability (mm) distance^{-1}
C_1	$06.32^{d}\pm0.16$	$0.75^{a}\pm1.89$	$08.42^{d}\pm1.15$	$10.30^{a}\pm2.10$	$11.39^{f}\pm1.39$	$62.55^{e}\pm0.71$	$03.00^{a}\pm0.94$	$25.39^{e}\pm0.75$	$0.008^{a}\pm0.17$	$6952^{e}\pm0.29$	$1.22^{d}\pm0.06$
C_2	$06.32^{d}\pm0.09$	$0.75^{a}\pm0.15$	$08.42^{d}\pm0.63$	$10.68^{b}\pm0.49$	$11.05^{e}\pm1.47$	$51.92^{d}\pm0.88$	$03.49^{b}\pm1.83$	$23.19^{d}\pm0.43$	$0.014^{b}\pm0.13$	$5551^{d}\pm0.19$	$1.22^{d}\pm0.17$
C_3	$06.29^{c}\pm2.27$	$0.78^{b}\pm0.67$	$08.06^{c}\pm049$	$10.75^{c}\pm1.27$	$10.53^{d}\pm1.60$	$48.56^{c}\pm0.61$	$03.83^{c}\pm1.73$	$21.04^{c}\pm0.86$	$0.015^{b}\pm0.14$	$4700^{c}\pm0.27$	$1.18^{d}\pm0.34$
C_4	$06.27^{c}\pm1.15$	$0.79^{b}\pm1.33$	$07.93^{b}\pm1.28$	$10.84^{d}\pm0.82$	$10.00^{c}\pm0.68$	$46.94^{b}\pm0.35$	$04.71^{d}\pm1.29$	$18.56^{b}\pm0.51$	$0.016^{b}\pm0.18$	$4625^{b}\pm0.36$	$1.11^{c}\pm0.08$
C_5	$06.23^{b}\pm0.62$	$0.80^{b}\pm3.15$	$07.78^{b}\pm1.23$	$11.03^{e}\pm0.21$	$09.24^{b}\pm0.91$	$46.51^{b}\pm0.40$	$04.91^{e}\pm1.45$	$17.44^{b}\pm0.37$	$0.019^{c}\pm0.13$	$4614^{b}\pm0.05$	$1.01^{b}\pm0.43$
C_6	$06.17^{a}\pm1.39$	$0.81^{b}\pm0.96$	$07.61^{a}\pm2.31$	$11.35^{f}\pm0.58$	$08.49^{a}\pm1.97$	$38.13^{a}\pm0.44$	$06.10^{f}\pm0.51$	$13.54^{a}\pm0.38$	$0.024^{d}\pm0.10$	$2987^{a}\pm0.64$	$0.92^{a}\pm0.09$

The values with different superscript in a column differ significantly ($p\leq 0.05$). C_1, C_2, C_3, C_4, C_5 and C_6 refer to cookies made from 0, 20, 40, 60, 80, 100% composite flour, respectively.

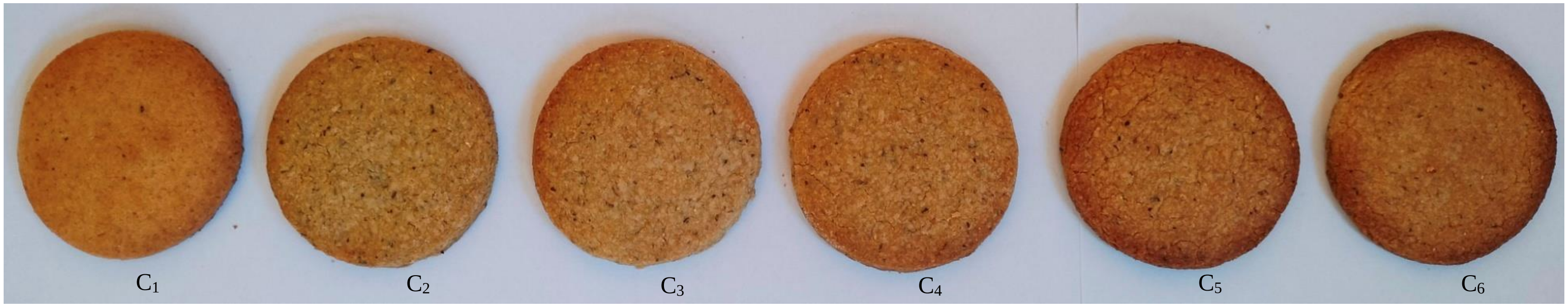

Fig. 1. Multi-millet cookies.

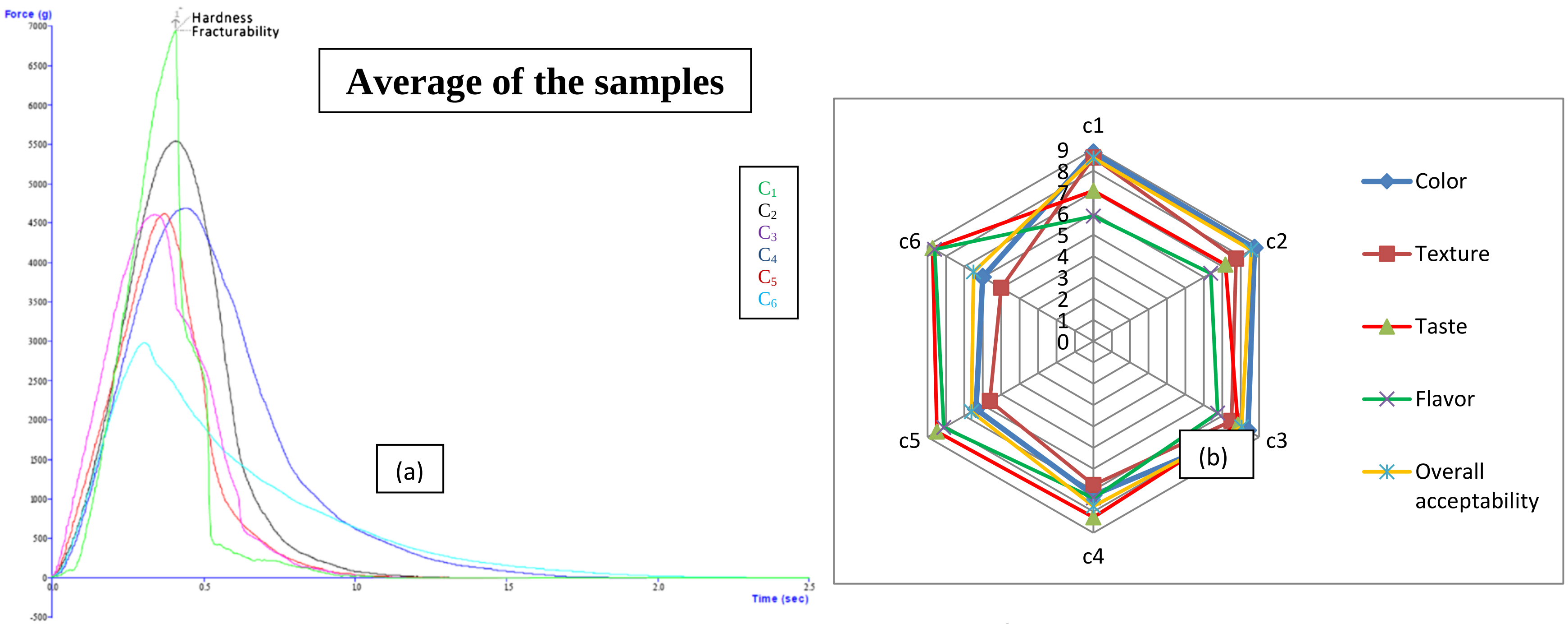

Fig. 2. (a) Hardness, and (b) Sensory scores of cookies.