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Effect of fortification of mango (*Mangifera indica)* kernel flour on nutritional, phytochemical and textural properties of biscuits

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Abstract
The feasibility of partially replacing refined wheat flour with Mango kernel flour in biscuit making was evaluated in several formulations, aiming to find a formulation for the production of Mango kernel flour incorporated biscuits with better nutritional, phytochemical quality and consumer acceptance and to utilize the waste obtained from mango processing. Kernels of Kesar Mango were separated from seeds and processed into flour through various processing steps. Mango kernel flour is a good source of starch, protein, dietary fibre, minerals and vitamins. Mango kernel flour was incorporated at levels of 10, 20, 30 and 40 percent in preparation of biscuits with maintaining all other ingredients constant. The prepared biscuits were subjected to organoleptic, nutritional and phytochemical analysis. Biscuits containing up to 30 % mango seed kernel flour were acceptable without adverse effect on sensory qualities. The prepared biscuits were feasible to be exploited in market

Keywords: Refined wheat flour, mango kernel flour, biscuits, phytochemical

Introduction
Bakery industry is the one of the largest food industries in India with an annual turnover about Rs. 3000 billion. The biscuit industry has been growing at an average rate of 15% during the past 3 years and this is expected to be maintaining in coming years (IBMA, 2010) [19]. Baking technique is probably the earliest and oldest of all other techniques and is still going steady over food processing field. Bakery products have played a key role in the development of mankind, being a principal source of convenience, variety and a healthy nutrition component to modern lifestyles. Biscuits, cookies and crackers represent the largest category of snack items among the baked foods in India.
Biscuit consumption per capita in India is 2.1 kg, compared to more than 10 kg in the USA, UK and West European countries and above 4.25 kg in South-east Asian countries, e.g. Singapore, Hong Kong, Thailand, Indonesia, etc. China has per capita consumption of 1.90kg, while in the case of Japan it is estimated at 7.5kg (Serivastava 2009) [32].
Biscuits constitute major component of human snacks in most part of the world. It is an unleavened crisp, sweet pastry made from wheat flour, shortening (hydrogenated fat) and sugar, and is usually made light by the addition of baking powder (a mixture of sodium carbonate, sodium bi-phosphate and cereal flour). Wheat flour constitutes the basic ingredient for biscuit production because of its gluten proteins, which are not present in flour of other cereals. Gluten protein forms elastic dough during baking and gives high organoleptic quality to the finished product.
The biscuits available in market are prepared from wheat flour (whole/refined) which lacks in good quality protein because of its deficiency in lysine; and dietary fibre contents. Mango kernel flour which is highly nutritious in protein, vitamin, minerals and phytochemical content has been found for its incorporation into preparation of biscuit. The study provides the information about a commercially viable application of increasing protein and nutraceutical content in biscuits and also these can be solve the problem of malnutrition and other essential macro and micro nutrients deficiency among the population. The objective of the present study was also to expand the utility mango kernel flour by value addition through incorporating with wheat flour to prepare the biscuit and their characterization. Present study was conducted to assess the effect of addition of mango kernel flour in biscuits also evaluate the consumer acceptability.
"Kesar" Mango is the queen of mangoes widely grown in the Gujarat state of India. It has a unique sweet taste. Kesar is characterized by its golden color with green overtones. This variety is slightly smaller in size as compared to the Alphonso mango variety but offers a

unique taste. Kesar variety of mangoes are considered to be one of the best mangoes, which are famous for their fruit quality, attractiveness fruit and pulp color, taste & good shelf life. This is a leading variety of Gujarat with a red blush on the shoulders. Fruit size is medium, shape oblong. The mango has high pulp content, and a saffron color (hence the Indian name- 'Kesar'). It has a very rich pulp and is therefore ideal for pulping and juice concentrates.

During processing of mango, peel and kernel are generated as waste (40-50% of total fruit weight). They are rich in various nutrients and many value added products can be obtained from them. Kernels take up about 17-22% of the fruit. The major components of mango seed are starch, fat and protein. The oil of mango seed kernel consists of about 44-88% saturated fatty acids. Mango seed kernel can be used as a potential source for functional food ingredients, animal feeds, antimicrobial compounds and cosmetic due to its high quality of fat and protein as wells as high levels of natural antioxidants. Food industry waste and byproducts are substances that originated during processing and can be further utilized in different ways. If we could produce valuable products from the industry by products through new scientific and technological methods, environmentally pollution by-product can be converted into products with a higher economic value.

The complete exploitation of these mango seed kernels not only eliminates the disposal problem but also results in valuable products production. Several studies have shown that mango seed kernels contain various phenolic compounds and can be a good source of natural antioxidants (Puravankara *et al*., 2000 [30]; Abdalla *et al*., 2007 [2]). Gallotannins and condensed tannin-related polyphenols were reported to be present in mango kernels (Arogba 1997) [8]. In addition, polyphenols from dry mango kernel meal were found to contain tannic acid, gallic acid, and epicatechin in the ratio 17:10: 1, respectively (Arogba 2000) [7].

Mango kernel was also shown to be a good source of phytosterols including campesterol, β-sitosterol, stigmasterol and it also contains tocopherols (Soong and Barlow 2004) [34]. Seeds of *Mangifera indica* contain phytochemicals such as tannins, polyphenol, alkaloid etc. A few reports on characterization of protein showed that it can be a good source for essential amino acids (Kittiphoom, 2012) [22].

Materials and Methods
Materials
The study was conducted in the laboratories of the Department of Food Engineering, College of Food Technoogy, VNMKV, Parbhani. The Mango fruits (*Mangifera indica*) and all other ingredients were purchased from the local market of Parbhani. The proposed research was carried out in Department of Food Engineering, College of Food Technoogy, VNMKV, Parbhani.

Methods
Preparation of Mango kernel flour
Evenly ripened disease free and sound mango fruits have been selected. The pulp and seed of mango fruit was separated manually. Mango seeds were washed and dried in hot air at 60 °C for 6 hours. Kernels were separated from stone manually using stainless steel knife and dried in hot air oven at 50 ºC for 4 hours and stored in air tight containers. During processing stored kernels were soaked (18-20 hr) in water, chopped into small pieces, blanched (1-2 min), dried (60 ºC for 5 hours) and ground into flour in electric blender, sieved and stored in air tight container (Yatnatti *et al*., 2014) [38].

Preparation of Biscuits
Firstly all dry ingredients were sieved and mixed together. Sugar and fat were creamed together. Soft dough was prepared in dough mixer with all the ingredients and an adequate amount of water. Dough was spread into a sheet and cut into suitable shapes and size. Cut pieces were gently placed on baking tray. Tray was placed in oven for approximately 15 minutes at 170 °C. Baked biscuits were removed from oven and cooled. Prepared Biscuits were packed in suitable packaging material. Biscuits were prepared by method described by Yadav *et al*., (2012) [36].

Mango kernel flour replaced the wheat flour to the extent of 0%, 10%, 20%, 30% and 40% levels without altering the total flour content of the preparation. Standardization of recipe for formulation of Biscuits with fortification of Mango Kernel Flour was as follows:

Table 1: Formulation of Mango Kernel Flour Biscuits

Ingredients	Amount (g)				
	K0	K1	K2	K3	K4
Refined wheat flour	100	90	80	70	60
Mango Kernel Flour	--	10	20	30	40
Fat	25	25	25	25	25
Sugar	50	50	50	50	50
Salt	1	1	1	1	1
Baking powder	2.5	2.5	2.5	2.5	2.5
Milk powder	2	2	2	2	2
Water (mL)	20	20	20	20	20

Where K_0- control sample
K_1- Biscuits with 10 percent Mango kernel flour
K_2- Biscuits with 20 percent Mango kernel flour
K_3- Biscuits with 30 percent Mango kernel flour
K_4- Biscuits with 40 percent Mango kernel flour

Proximate analysis
The proximate composition like moisture, protein, lipid, crude fibre and ash were measured as per standard method given in A.O.A.C. method (2005) [6].

Minerals Content
Mineral content of samples were estimated by method given by (Ranganna, 1986) [31].

Phytochemical Analysis of Mango Kernel Flour
Determination of Total Phenolic Content
Preparation of sample extracts
The extracts of samples were obtained as described by Bloor (2001) [10]. Half gram from samples was extracted with 20 ml of methanol: water (60:40 v/v). The mixture was centrifuged and the supernatant was adjusted to 25 ml. An aliquot of these extracts were used for the quantification of total phenolics content.

Determination of Total Phenolic Content
Total phenolics content of the extracts were used for the quantification of total phenolics. The total phenolics content was determined colorimetrically, using the Folin-Ciocalteu method as described by Singleton *et al*., (1999) [33]. Aliquots of 1 ml of the extract were added to 1 ml of Folin-Ciocalteu reagent followed by addition of 1 ml of an aqueous 7.5% solution of sodium carbonate. The mixture was stirred and allowed to stand for 30 min. The absorbance at 765 nm was measured using a model UV/VIS-Spectrophotometer. A blank

sample consisting of water and reagents was used as a reference. The results were expressed as milligrams of gallic acid equivalents per gram flour (mg GAE/g).

$$C = c\ V/m$$

Where: C - Total content of phenolic compounds in gallic acid equivalent (GAE)
c - Concentration of gallic acid extrapolated from the calibration curve, mg/ml
V - Volume of methanolic sample extracts (ml)
m - Weight of the sample taken.

Determination of Total Tannin Content

The vanillin method was used to determine total tannin content as described by Sundang *et al*., (2012) [35]. Concentrations of 0, 5, 10, 20, 30, 40, 50 and 100 µg/ml of reference catechin were prepared to obtain a calibration curve, and 100 µg/ ml of each sample was prepared to determine total tannin content in catechin equivalent. Samples or reference (1.0 ml) was mixed with 3.0 ml of 4 % (w/v) vanillin followed by addition of 1.5 ml 1 M HCl, then incubated in the dark for 5 min and the absorbance measured at 500 nm using a UV-Vis spectrophotometer.

Determination of Total Flavonoid Content

The determination of the total flavonoid content of the plant extract was based on the aluminium chloride colorimetric method of Zhilen *et al*., (1999) [39] and modified by Miliauskas *et al*., (2004) [26]. Distilled water (0.4 ml) was added to 0.1 ml of sample extract or reference. This was followed by 0.1 ml of 5 % sodium nitrite. After 5 min of incubation, 0.1 ml of 10 % aluminium chloride and 0.2 ml of sodium hydroxide were added and the volume was made up to 2.5 ml with distilled water. The absorbance at 510 nm was measured against the blank. A stock solution of quercetin (1 mg/ml) was prepared. Aliquots of 0.2, 0.4, 0.6, 0.8, and 1 ml were taken. Each volume was made up to 2 ml with distilled water and treated as described above, in order to prepare a calibration curve. The total flavonoid content of the sample was then calculated as shown in the equation below and expressed as mg quercetin equivalents per gram sample. The analysis was conducted in triplicate.

$$X = q\ V/w$$

Where X - total content of flavonoid compounds in quercetin equivalent
q - Concentration of quercetin extrapolated from the reference curve
V - Volume of extract (ml)
w - Weight of the sample

Determination of Alkaloid Content

Five gram of the sample was weighed into a 250ml beaker and 200ml of 10% acetic acid in ethanol was added and allowed to stand for 4minutes, this was filtered and extract was concentrated on a water bath to one quarter of the original volume. Concentrated ammonium hydroxide added drop wise to the extract until the precipitation was completed. The whole solution was allowed to settle and the precipitate was collected and washed with dilute ammonium hydroxide and then filtered. The residue was alkaloid which was dried and weighed (Harbone, 1973) [18].

$$\%\ \text{Alkaloid} = \frac{W_3 - W_2}{W_1} \times 100$$

Where: W_1 =initial weight of sample,
W_2 =weight of the extract,
W_3 = final weight of the residue

Determination of Saponin content

Two gram of the finely grinded sample was weighed into a 250 ml beaker and 100 ml of Isobutyl alcohol was added. Shaker was used to shake the mixture for 5hours to ensure uniform mixing. The mixture was filtered using No. 1 Whatman filter paper into 100 ml beaker containing 20 ml of 40% saturated solution of magnesium carbonate. The mixture obtained again was filtered using Whatman filter paper No. 1 to obtain a clean colorless solution. One (1 ml) was added into 50 ml volumetric flask using pipette, 2 ml of 5% iron (iii) chloride ($FeCl_3$) solution was added and made up to the mark with distilled water. It was allowed to stand for 30 min for the colour to develop. The absorbance was read against the blank at 380 nm (Bruneton, 1999) [11].

$$\text{Saponin} = \frac{[\text{Absorbance of sample x concentration of standard}]}{\text{Absorbance of standard}} -1$$

Physical Analysis of Biscuits

Physical analysis of Biscuits was carried out to determine the Diameter, Thickness and Spread-Ratio as described by (A.A.C.C., 1976) [1].

a) Diameter: Diameter (W) of biscuits was measured by laying six biscuits edge-to-edge with the help of a scale. The same set of biscuits was rotated 90° and the diameter was remeasured. Average values were reported in millimeter.

b) Thickness: Thickness (T) of biscuits was measured by stacking six biscuits on top of one another and taking the average in millimeter.

c) Spread ratio (D/T): The spread ratio was calculated by dividing the average value of diameter (D) by the average value of thickness (T) of biscuits.

Sensory Analysis

The prepared Mango kernel based biscuits were organoleptically evaluated on the basis of 9-point hedonic scale. The level of addition of Mango kernel flour in biscuits was optimized on the basis of sensory score.

Textural Analysis Profile of Biscuits

Stable Micro System *TAXT2 plus* Texture Analyzer was used for texture profile analysis (TPA) of Biscuits prepared by different treatments. Each biscuit was placed on a heavy duty platform table with a holed plate, and the penetration test was performed using P/5 element. The probe penetrated completely through the biscuits. The heavy duty platform was repositioned so that there was no contact between the blade and slot surfaces and a 'blank' test run as a check. The blade was then raised to place of the sample. Samples were removed from their place of storage just prior to testing and allowed them to fit centrally on the platform under the knife edge. The blade was then allowed to shear through the sample. For comparison purposes, sample dimensions were kept constant. A typical textural profile curve for biscuit done by cutting test with one complete run. The TA settings selected were pre-test speed: 1.5 mm/s, test speed: 2 mm/s, post-test speed: 10 mm/s and distance: 5 mm. The absolute peak force of the resulting curve was considered as cutting strength of the biscuit. Hardness, defined as the maximum peak force during the first compression cycle (first bite) (Figure 1). The results of the experiments were given as the average of three replicates.

Results and Discussion

Proximate Composition of Mango kernel flour

The chemical composition is a simple and convenient way of illustrating the amount of nutrients was highly desirable and which could be noticed in present study. Proximate composition of mango kernel flour moisture, protein, fat, ash, crude fiber and carbohydrate and is as shown in table 2.

Table 2: Proximate Composition of Mango kernel flour

Parameters (%)	Mango kernel Flour
Moisture	7.79
Protein	9.36
Fat	9.59
Ash	1.31
Crude Fiber	1.76
Carbohydrate	70.69

* Each value is an average of three determinations

The data presented in table 2 shows that the moisture content of mango kernel flour was 7.79 per cent. The moisture content, protein, fat, ash, crude fiber and carbohydrate content mango kernel flour were determined as 7.79, 9.36, 9.59, 1.31, 1.76 and 70.69 percent. Ash content is an indication of the level of minerals present in food material this suggests that mango kernel can help in boosting the mineral content of prepared product. The carbohydrate level in mango kernel flour meaning they can be exploited as energy source foods.

Findings of present investigation related to the values of mango kernel were in close conformity with values described in literature with slight differences by Fowomola (2010) [15]. He reported that Mango kernel flour contains crude protein (10.06%), crude oil (14.80), ash (2.62), crude fibre (2.40) and carbohydrate (70.12%). Other Researchers, Yatnatti *et al*., (2014) [38] showed that processed mango kernel flour has 7.05 per cent moisture. Macro nutrient composition is as follows, protein: 7.53g, fat: 11.45 g, crude fiber: 2.20 g and carbohydrate: 69.77 g.

The observed differences may be due to varietal variations, environmental factors like climate and location and method of preparation etc.

Mineral Content of Mango kernel

The data presented in table 3 revealed that mango kernel flour is rich in mineral content. Among all minerals mango kernel flour contained potassium in high amount and observed as 645mg/100g. Potassium records an important nutritious role in any organism. Second abundant mineral in mango kernel flour was calcium and recorded as 325 mg/100g.

Table 3: Mineral composition of Mango kernel flour

Parameters (mg/1oo g)	Mango kernel Flour
Potassium	645
Calcium	325
Magnesium	178
Sodium	22

* Each value is an average of three determinations

The magnesium content of mango kernel flour was 178 mg/100g. While sodium content of mango kernel flour was 22 mg/100g. The high mineral content of the mango kernel flour justifies its role for utilizing it as an ingredient in the preparation of various foods. The results obtained during present investigations are more or less similar with obtained by Nzikou *et al*., (2010) [28] and Fowomola (2010) [15].

Phytochemical Content of Mango kernel flour

Phytochemical content of mango kernel flour was estimated and results pertaining to same are presented in Table 4.

Table 4: Phytochemical composition of Mango kernel flour

Parameters (g/1oo g)	Mango kernel Flour
Total Phenolic a Content	53.85
Flavonoid Content	0.87
Tannin Content	0.21
Alkaloid Content	4.1
Saponin content	0.12

a – mg of Gallic Acid Equivalent/g dw basis

It is seen from table 4 that mango kernel flour is excellent source of phytonutrients. Mango kernel flour was extracted with methanol and total phenolic content in extracts was determined. In mango kernel flour total phenolic content was found to be 53.85mg GAE per g. Higher values were reported by Soong and Barlow (2004) [34] i.e. 117 mg GAE per g this variation may be attributed to varietal difference. This result is in close agreement with Yatnatti and Vijaylaxmi (2017) [37].

The flavonoid content of mango kernel flour was found to be 0.87 g/100g. The alkaloid content of mango kernel and mango kernel flour was found to be 4.1g/100g. The presence of alkaloids in the mango kernel investigated showed that it has medicinal benefits.

While the tannin content of mango kernel flour was found to be 0.21 g/100g respectively. Saponin content of mango kernel and mango kernel flour was found to be 0.12 g/100g. Agroba (2014) [3] presented more or less similar findings for the flavonoids, tannins content in mango kernel. Variations may be due to varietal difference and climate.

Physical properties of biscuits with fortification of mango kernel flour

The data regarding Physical properties of biscuits with fortification of mango kernel flour is presented in Table-5.

Table 5: Effect of mango kernel flour on physical parameters of biscuits

Parameter	Samples				
	K_0	K_1	K_2	K_3	K_4
Diameter	5.63	5.71	5.78	5.82	5.86
Thickness	0.851	0.806	0.762	0.711	0.670
Spread Ratio	6.615	7.084	7.585	8.185	8.748

It could be observed from the Table-5 that the Diameter of biscuits significantly increased with increasing incorporation of mango kernel flour as compared with 5.63 cm for control while the thickness of biscuits is found to be decreased as compared with 0.851cm for control. The changes in diameter and thickness were reflected in spread ratio of biscuits. Concerning the spread ratio, it was observed that replacing of 40% wheat flour by Mango Kernel Flour recorded the highest value 8.748 compared with 6.615 for control. The results obtained in the present study are in agreement with those reported by Ashoush and Gadallah (2011) [9] and Elgindy (2017) [14].

Organoleptic evaluation of biscuits with fortification of mango kernel flour

Standard method was followed for preparation of Biscuits and different samples were prepared by incorporating varying levels of prepared mango kernel flour at the concentration of 0, 10, 20, 30 and 40 percent. The effects of mango kernel flour incorporation on organoleptic characteristics of Biscuits are presented in Table-6.

Table 6: Organoleptic Evaluation of biscuits with fortification of mango kernel flour

Samples	Appearance	Color	Flavor	Taste	Texture	Overall Acceptability
K_0	8.51	8.46	8.63	8.71	8.65	8.59
K_1	8.34	8.21	8.25	8.31	8.56	8.33
K_2	8.28	8.14	8.0	8.29	8.31	8.20
K_3	8.11	8.09	8.12	8.23	8.24	8.15
K_4	7.93	7.94	7.24	7.40	7.63	7.62

It is learnt from the results that mango kernel flour in the biscuits formulation significantly ($p \leq 0.05$) affected the acceptance of the product.

No significant change in observed in biscuits up to 10% (K1) of concentration of mango kernel flour. In case of all attributes K_1 sample with 10% mango kernel flour scored almost nearer to control samples, when compared to K_2, K_3 and K_4. Darkness in the samples increased with increase in polyphenol content of biscuits. Flavor increased as Mango kernel flour increased and bitterness was noticed as after taste in K_4 with 40 per cent. The texture acceptability of with respect to hardness increased with increase in concentration slightly up to 40%. Overall scores indicated highly acceptable for K_1-10%, K2-20% and K_3- 30% comparable to control (K_0). However, about 40 per cent of Mango kernel flour, the biscuits had a slight bitter which may be due to high polyphenols content activity. The results obtained are with agreement with those obtained by Kakali *et al.*, (2014) [21]. Thus, considering the acceptance, biscuits with 30 % mango kernel flour incorporation stood as the optimized product.

Textural analysis profile of biscuits with fortification of mango kernel flour

Texture is very important to the consumer in the determination of food quality. The most important textural characteristic in biscuits is its hardness which can be evaluated by the force required to puncture or fracture a sample. The Textural Profile of Biscuits is determined by using TA-XT Texture Analyzer and the results obtained are depicted in Table 7.

Table 7: Effect of incorporation of mango kernel flour on hardness of biscuits

S. No.	Sample Code	Hardness (Kg)
1	K_0 – Control sample	7.948
2	K_1- Containing 10% Mango kernel flour	8.765
3	K_2- Containing 20% Mango kernel flour	9.026
4	K_3- Containing 30% Mango kernel flour	10.299
5	K_4- Containing 40% Mango kernel flour	11.022

This clearly brings out the fact that, the hardness of the product depends on the quantity of mango kernel flour incorporated. The increase in mango kernel flour from 0 to 40 per cent simultaneously increased the hardness of the product from 7.948 to 11.022kg. Thus, the breaking strength of Biscuits increased significantly ($p \leq 0.05$) with incorporation of mango kernel flour and the results correlated with the hardness profile observed during sensory analysis, wherein incorporation of more than 30 % mango kernel flour in biscuits, drastically increased the hardness of biscuits up to an unacceptable level. The high protein content of mango kernel flour and its interaction may be the contributory factor for harder texture of the mango kernel flour fortified biscuits. McWatters *et al.*, (2003) [25] reported that incorporation of cowpea flour in cookies increased the hardness due to high percent of protein content, which may interact with other ingredients and contribute to harder texture. Ajila *et al.*, (2008) [4] also reported that the breaking strength of the biscuits also increased with increase in mango peel fiber level. The hardness of Biscuit samples was as shown in Figure 1.

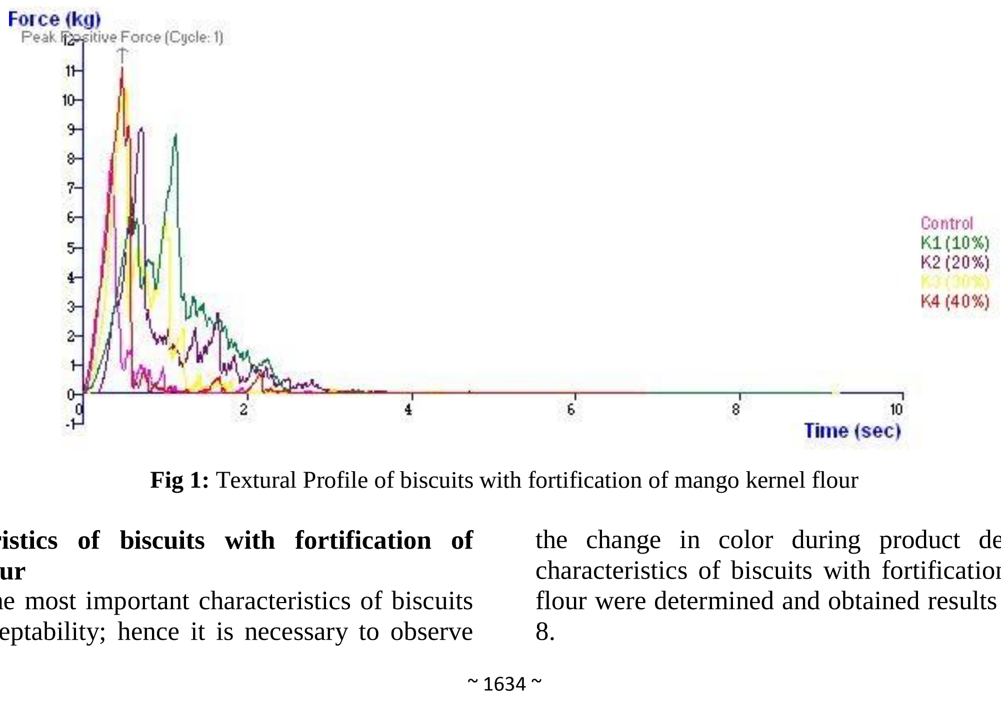

Fig 1: Textural Profile of biscuits with fortification of mango kernel flour

Color characteristics of biscuits with fortification of mango kernel flour

Color is one of the most important characteristics of biscuits for consumer acceptability; hence it is necessary to observe the change in color during product development. Color characteristics of biscuits with fortification of mango kernel flour were determined and obtained results are given in Table 8.

Table 8: Effect of incorporation of mango kernel flour on color characteristics of biscuits

Sample Code	Color characteristics			Chroma	Hue
	L*	***a****	***b****		
K_0	78.22	3.73	25.86	26.13	81.81
K_1	65.26	7.47	27.19	28.19	74.60
K_2	61.93	7.88	25.57	26.75	72.84
K_3	57.75	8.55	25.92	27.30	71.74
K_4	56.25	9.30	23.43	27.06	69.89

Darkness in the samples increased with increase in total phenolic content of biscuits. Based on the observations of organoleptic evaluation, physical properties and color characteristics, it is concluded that 30% of mango kernel flour could be successfully incorporated in biscuits and hence considered optimum level for further analysis.

Proximate composition of Organoleptically accepted Biscuits

Proximate composition of food products gives the picture on the major nutrients in the product. The proximate compositions 30% mango kernel flour incorporated biscuits is compared with the samples prepared with control samples and the results are presented in Table-9.

Table 9: Proximate composition of Biscuits (Control Biscuits and Biscuits with 20% Mango kernel flour)

Parameter (%)	Control (K_0)	Biscuits with 30% Mango kernel flour (K_3)
Moisture	2.32	2.56
Protein	5.58	5.21
Fat	12.53	14.06
Ash	1.56	1.92
Carbohydrate	75.89	74.68

* Each value is an average of three determinations

The moisture content of Control biscuit sample and mango kernel incorporated biscuit sample was 2.32 and 2.56 per cent respectively. Whereas Protein, Fat, Ash and carbohydrate content of Control biscuit sample was 5.58, 12.53, 1.56 and 75.89 percent and mango kernel incorporated biscuit sample was 5.21, 14.06, 1.92 and 74.68 per cent respectively.

Data showed that with increasing level of Mango kernel flour increased the moisture content from 2.32 for control to 2.56 percent for 30 percent mango kernel flour. The results showed that there was increase in moisture content of biscuits as increasing level of mango kernel flour. Fat content also increased from 12.53 for control to 14.06 percent for 30 percent mango kernel flour due to high fat content of mango kernel flour whereas protein content of biscuits decreased from 5.58 for control to 5.21 percent for 30 percent mango kernel flour. Ash content of Control biscuit sample and mango kernel incorporated biscuit sample was 1.56 and 1.82 per cent, respectively. Similar results were observed by Kumar *et al.*, (2010) [23] studied the effect of addition of millet flours in soy based biscuits.

Phytochemical Content of Organoleptically accepted Biscuits

Phytochemical content of organoleptically accepted Biscuits was estimated and results pertaining to same are presented in Table 10.

Table 10: Phytochemical Content of Organoleptically accepted Biscuits

Parameters (g/1oo g)	Control (K_0)	Biscuits with 30% Mango kernel flour (K_3)
Total Phenolic a Content (mgGAE/g)	1.42	18.154
Flavonoid Content	0.035	0.194
Tannin Content	0.041	0.064
Alkaloid Content	0.06	0.82
Saponin content	0.005	0.038

a – mg of Gallic Acid Equivalent/g dw basis

It is clear from the table 10 that the Biscuit with 30% Mango kernel flour (K_3) is high in phytochemical content as compared to control sample. Total Phenolic a Content of K_3 was 18.154 mgGAE/g. In human phenolic compounds have been reported to exhibit a wide range of biological effects including anti bacterial, anti inflammatory and antioxidant property (Han *et al.*, 2007) [17]. The bioactive compounds such as phenolic compounds are responsible for valuable antioxidant potential of extracts from different plant materials such as fruits, seed, peels, leaves and stem and these are regarded as health beneficial constituents (Ghafoor, 2011) [16]. The use of phenolic is also reported for lowering and preventing obesity, effecting secretion of a dipokine and prevention of oxidative stress (Dalar *et al.*, 2014) [13].

Flavonoids content of the K_3 was observed to be 0.194 g/100g. The biological function of flavonoids includes protection against allergies, inflammation, free radicals platelet aggregation, microbes, ulcers, hepatoxins, viruses and tumors (Okwu, 2004) [29]. The alkaloids content of K_3 sample was 0.82 g/100g. Pure isolated plant alkaloids and their synthetic derivatives are used as basic medicinal agent for analgesic, anti pasmiodic and bacterial effects (Okwu, 2004) [29]. The presence of alkaloids in the mango kernel investigated showed that it has medicinal benefits.

Tannins and saponin content of K_3 Biscuits sample were observed to be 0.064 and 0.038 g/100g respectively. The presence of tannin could be responsible for the bitter principle and sour taste. Tannin has astringent properties, hastens the healing of wounds, and inflamed mucous membranes (Okwu, 2004) [29]. Saponin have natural tendency to ward of microbes which makes them good for treating fungal and yeast infections. These compounds serve as natural antibiotics, helping the body to fight infection and microbial invasion (Okwu, 2004) [29]. Saponin therefore binds cholesterol and thus interferes with cell growth and division. The increase in phytochemical content of the Organoleptically accepted Biscuits is due to the incorporation of mango waste that is mango kernel flour.

Mineral Content of Organoleptically accepted Biscuits

Mineral content of organoleptically accepted Biscuits was estimated and results pertaining to same are presented in Table 11. It is revealed from table that organoleptically accepted Biscuits is rich in mineral content.

Among all minerals organoleptically accepted Biscuits contained potassium in high amount as compared to control and observed as 402mg/100g which significantly increased as control biscuits. Potassium records an important nutritious role in any organism.

Second abundant mineral in present mango kernel was calcium and recorded as 118 mg/100g in K_3. One of the main benefits from calcium is related to interactions between cells walls. Therefore, it ensures the cells structure by hard cementing them. Calcium is a cellular component and regulator of the nervous excitability (Marschner, 1986) [24]. It's also a factor of ethylene synthesis during the fruits ripening (Morard, 1996) [27].

Table 11: Mineral Content of Organoleptically accepted Biscuits

Parameters (mg/1oo g)	Control (K_0)	Biscuits with 30% Mango kernel flour (K_3)
Potassium	274	402
Calcium	41	118
Magnesium	103	141
Sodium	291	320

* Each value is an average of three determinations

The magnesium content of control and K_3 Biscuits was 103 and 141 mg/100g. Magnesium has major nutritional and therapeutic actions. Indeed, magnesium is an essential mineral for the cells functions. A daily sufficient intake is necessary for the energy production of the organism, the keeping of good cardiac rate and the fight against stress. Magnesium intervenes in several metabolic reactions like activator or enzymatic regulator. Thus, it improves the good functioning of the digestive tract, the maintenance of the structures of bones, teeth and the proteins synthesis. It takes part in the regulation of some minerals such as calcium, potassium, copper or zinc. Numerous nutritional investigations revealed that the magnesium intakes are generally below the securities. Thus, the European health authorities recommend daily contributions of 150 to 500 mg of magnesium for the adults (Ilja and Hollman, 2005) [20].

While sodium content of control and K_3 Biscuits was 291 and 320 mg/100g. Slight increase in sodium content was observed. Intake of higher potassium and less sodium content could prevent the hypertension, source of the cerebral vascular damages and the heart diseases (Cook and Obarzanek, 2009) [12]. It is the main intracellular mineral and takes part in the muscular activity and to the heart muscular. A diet with high potassium content is favourable to the healthy bone due to its alkaline effect (Anonyme, 2010) [5]. The high nutritional and phytochemical content of the mango kernel flour justifies its role for utilizing it as an ingredient in the preparation of various foods.

Conclusion

The present investigation aimed in exploration and quality evaluation of Mango (*Mangifera indica*) waste (kernel) as a nutraceuticals in biscuit. In the light of scientific results of the present investigation, it can be concluded that Mango kernel which are discarded as waste from processing industries, is having excellent phytochemical profile and is a mine of nutraceutical components. Various phytonutrients viz total phenolic contents, alkaloids, flavonoids, Saponin and tannin and minerals are present in abundant quantity. The variety selected in this study is locally available Kesar Mango. Mango stone was separated and processed to prepare Mango kernel flour. Biscuits were formulated from refined wheat flour incorporated with Mango kernel flour. Based on sensory evaluation biscuit sample was selected. The addition of 30 per cent Mango kernel flour yielded a nutritious product with nutraceutical properties with good sensory attributes.

Hence, it is finally concluded that developed processing technology for preparation of Biscuits with incorporation of Mango kernel flour, is having the nutraceutical and therapeutic values and therefore can be commercially exploited to address the problem of malnutrition in the developing countries.

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