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Physico-chemical and Nutritional Evaluation of Cookies with Different Levels of Rosehip and Hibiscus Powder Substitution

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Authors' contributions

This work was carried out in collaboration among all authors. Author SA designed the study. Author SA performed the statistical analysis, wrote the protocol and wrote the first draft of the manuscript. Authors AS, AB, RT and SS managed the analyses of the study and the literature searches. All authors read and approved the final manuscript

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ABSTRACT

Aim: Food with high nutritional value is in great demand for proper functioning of body systems and potential health benefits. As a result, value-added foods or functional foods with higher level of dietary fiber and antioxidant have developed, especially in bakery products such as cookies. This study was aimed to develop and evaluate proximate composition, phenolic compounds, vitamin C content and sensory acceptance of cookies.
Study Design: The formulations were prepared (20% rosehip powder and 15% hibiscus powder in formulation CF1, 15% rosehip powder and 20% hibiscus powder in formulation CF2, 15% rosehip powder and 15 % hibiscus powder in formulation CF 3) in a standard cookie recipe.
Place and Duration of Study: Department Of Food Technology, ITM University Gwalior, Madhya Pradesh, (India), between June 2018 to February 2019.

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Methodology: The proximate analysis,phytochemical analysis and sensory analysis of cookies was done using standard AACC International procedures.
Results: Cookies prepared from formulation second (15% rose hips and 25% hibiscus powder) was more acceptable than other formulations on the basis of sensory evaluation.
Conclusion: The analysis revealed that the increment in substitution in flour has increased the health benefits of cookies when compared to control cookies. The substitution with Rose hips and Hibiscus powder has a significant share in enhancing the TPC content, Total antioxidant capacity and other bioactive compounds in cookies. For sensory quality the CF2 formulation (the overall acceptability) has found greatest in all formulations. Substitution with Rose hips and Hibiscus could bring much potential health benefits to the consumer by adding nutritional (crude fat, crude protein and crude fiber) as well as organoleptic properties.

Keywords: Cookies; proximate composition; bioactive compounds; rosehips and hibiscus.

1. INTRODUCTION

Food with high nutritional value is in great demand for proper functioning of body systems and potential health benefits. As a result, value-added foods or functional foods with higher level of dietary fiber and antioxidant have developed, especially in bakery products such as cookies [1]. Numerous studies have been carried out in order to replace wheat flour made from fruit residue, petal of flowers and food waste in preparation of bakery products such as cookies and biscuits, due to the new consumption trends and eating habits [2]. Cookies are widely consumed throughout the world. Cookies are appreciated for their taste, aroma, convenience, and long shelf stability due to low moisture content. In fact they represent the largest category of snack foods in most of the world.

Recent years have witnessed enhanced research work reported on plants and plant products. In this regard, plants with traditional therapeutic usage are being screened more efficiently to be considered as a substitution [3]. Hence in relation to good health demands, the nutritional value of cookies can be enhanced by supplementation with other nutrient sources [1]. The flour can be mixed with proportion of rosehips (*Rosa canina*) and China rose (*Hibiscus rosa-sinensis L.)*. Rosehip fruits (*Rosa canina*) grow wildly in various regions of India, having culinary and medicinal values. Rosa species have attracted the attention due to their antioxidant, antimicrobial and other properties. It is mostly used for prevention and treatment of common cold, gastrointestinal disorders, diabetes, kidney disorders and infections [4]. Rosa canina fruits are a valuable source for food and pharmaceutical industry. They contain a wide variety of biologically and physiologically active ingredients such as vitamin(C, B, P, E, K), flavonoids, carotenes, organic acids (tartaric, citric), carbohydrates (mono and oligosac-charides) and trace elements. Rosehips are also well known to have the high vitamin c content (300-4000 mg. /100 gm.) among fruits and vegetable [5].

Hibiscus *(Hibiscus rosa-sinensis L.)* is a profusely flowering, perennial, woody ornamental shrub distributed widely in tropical regions. Studies have indicated *H. rosa-sinensis* to possess bioactive properties and is recommended to be used as an herbal alternative to cure many diseases [3]. Hibiscus flower is also known to contain components that lower the blood pressure. A natural emollient, the leaves and flowers of hibiscus plant are used all over the world for softening or healing the skin and as a hair conditioner. The red flowers of hibiscus are high in Vitamin C content [6]. Edible flowers have been traditionally utilized for human consumption in various cultures. They enhance the taste, appearance and aesthetic value of food. “The edible flower is defined as Non-toxic, innocuous flowers with health benefits consumed in human diet” [7]. Hibiscus flowers are the natural source of Anthocyanins and Flavonoids. Anthocyanins are natural colorants that have extensive range of colors and occur widely in nature. Anthocyanins are the most important pigments ranging from orange, pink, red and violet to blue in the flowers and fruits of the vascular plants. They are harmless and water soluble which makes them interesting for their use as natural water soluble colorants.

For this research, rosehips and hibiscus flower petals were dried and processed in to powder before incorporated in to cookies formulation. rosehips and hibiscus flowers have been extensively used in food preservation and beverages but not in bakery products. Therefore the purpose of this paper was to prepare and evaluate the quality of cookies which can impart valuable source of nutrition and dietary fibers.

2. MATERIALS AND METHODS

2.1 Sample Collection and Preparation

The sample of mature rosehips and hibiscus flowers were procured from the campus of ITM University Gwalior (M.P). After the sample collection, rose hips were dried in hot air oven at low temperature (50°C) and petals of hibiscus flowers were dried naturally. The dried samples were secured in airtight jars to prevent degradation.

In the recipe rosehip and hibiscus powder was used with flour (20% rosehip powder and 15% hibiscus powder in formulation I, 15% rose hip powder and 20% hibiscus powder in formulation II, 15% rosehip powder and 15% hibiscus powder in formulation III) in a standard cookie recipe. In the procedure sugar, egg white, butter was creamed in a hand mixture. The measured amount of flour and other ingredients with rosehip and hibiscus powder was mixed with the liquid ingredients properly. The firm dough was then rolled and cut with a cookie cutter. Prepared cookies were then transferred to the greased trey and baked at 150 °C for 15 min. The baked cookies were removed from baking oven cooled to room temperature prior to analysis.

Proximate analysis was conducted on the control cookies and cookies prepared from the substituted flour. According to AACC standard methods, moisture content was carried out based on AACC method (44-19.01) [8]. The hot air oven was set at a temperature of 135°C.The crude fat content was determined by Soxhlet extraction method described in AACC (2000), Method [30-25.01] [9]. Crude fiber analysis was conducted based on AACC Method [32-10-01] [10]. The crude protein content was determined by the method described in AACC Method (46-10.01) [11]. The ash content was determined by the method described in AACC (2000), Method no. 08-01 [12]. Carbohydrate content was calculated using the following equation .

% Carbohydrate = *100% - (Moisture + Crude fat + Ash + crude protein) %*

For the analysis the samples were ground in to fine particles. The analysis was carried out in triplicates for all samples and expressed in a dry basis.

2.2 Analysis of Physical parameters

Spread ratio: The spread ratio was determined by using this formula. AACC Method 10-50.05 (AACC, 1999) [13].

Spreadratia(D/T)=diameter(mm)/thickness(mm)

Volume: Volume of biscuit is defined as the area of the cookie multiplied by thickness.

$$Volume\ (cm^3) = \frac{d^2 \prod T}{4}$$

t=Average thickness of biscuit (mm)
d=Diameter of biscuit (mm)

Density: After calculating volume, density was obtained by ratio of weight of volume (AACC 1983) [14].

$$Density\ (g/cm^3) = \frac{mass\ of\ sample\ (g)}{volume\ of\ sample\ (cm3)}$$

2.3 Analysis of Bioactive Compounds

2.3.1 Sample preparation for analysis

Sample (1 g) was mixed with 100 ml methanol 70% (v/v) in conical flask (250 ml) wrapped with aluminum foil. The mixture was then shaken in an orbital shaker for 2 hrs. at 160 rpm . Next, the mixture was poured into centrifuge tubes (wrapped with aluminum foil) and centrifuged in a centrifuge at 2500 rpm for 30 minutes to get a clear solution. The extract obtained was used for total phenolic content (TPC), total antioxidant value (phospho-molybdenum method) and other bioactive component analysis. For vitamin c analysis the sample was mixed with 100 ml water, and then the above mentioned procedure has been repeated for vitamin C estimation.

Table 1. Formulation of Cookies

Ingredients	Control	CF1	CF2	CF3
Flour (g)	100	65	65	70
Rosehip powder (%)	-	20	15	15
Hibiscus powder (%)	-	15	20	15
Butter (g)	20	2	20	20
Sugar (g)	25	25	25	25
Baking powder (g)	1	1	1	1
Beaten whole egg (No's)	1	1	1	1

2.3.2 Total phenolic content

Total phenolic contents in extract were determined by the folin-ciocalteu reagent method. 1 ml. of the aqueous extract of leaves was mixed with 5 ml.folin-ciocalteu reagent (CDH) (diluted with water 1/10 v/v) and 4 ml.(7.5%) of sodium carbonate. The solution was allowed to stand for 30 min. at 20°C.Absorbance of the sample and standard were measured at 765 nm. by using spectrophotometer (ELICO Double Beam, SL164, UV-VIS Spectroscopy). The total phenolic content in plant extract in gallic acid equivalents (GLE) was calculated using the following equation [15].

$$C = (c \times V) / m$$

Where,

C = total content of phenolic compounds mg/gm plant extract
c = the concentration of gallic acid established from the calibration curve (mg/ml)
V = the volume of extract in ml.
m = weight of crude plant extract in gm.

Vitamin C content: The amount of ascorbic acid of the rose hips was determined according to the methods of AOAC 967.21.1 [16]

2.3.3 Total anthocyanin content

Anthocyanins were extracted from samples by homogenizing the sample of cookies with acidified 70 % ethanol solution. The mixture was centrifuged after 10 min. The pH was adjusted by HCl. The mixture was brought to the volume of 25 ml and the absorbance was measured 515 nm, using ELICO Double Beam, SL164 UV-VIS Spectrophotometer [4].

$$Anthocyanin\ (mg\ per\ 100g) = absorbance\ at\ 515nm \times volume\ of\ extracrtion\ solution \times 100 / wt\ of\ sample\ (gm) \times 98.2$$

2.3.4 Total flavonoid content

The flavonoids content was measured following a spectrophotometric method (ELICO Double Beam, SL164 UV-VIS Spectrophotometer) .The sample extract were appropriately diluted with distilled water. Initially, 5% sodium nitrite solution was added to each test tube; after five minutes, (0.3 ml.)10% AlCl3 solution was added and then at 6thminutes 1.0 M NaOH was added. Finally, water was then added to the test tube and mixed well. Absorbance of resulting pink-colored solution was read at 510 nm against the blank (distilled water) [4]. The concentration of flavonoid was read on calibration line, the content of flavonoid was expressed in terms of quercetin (mg/100 g).

2.3.5 The total antioxidant capacity

The total antioxidant capacity was evaluated by the phospho-molybdenum method according to the procedure described by [17]. A 0.3 ml of extract was combined with 3 ml of reagent solution (0.6 M sulfuric acid, 28 mM sodium phosphate and 4 mM ammonium molybdate).The tubes containing the reaction solution were incubated in a water bath at 95°C for 90 min. Then, the absorbance of the solution was measured at 695 nm using a UV-VIS spectrophotometer (ELICO Double Beam, SL164 UV-VIS Spectrophotometer) against blank after cooling to room temperature. Methanol (0.3 ml) in the place of extract was used as the blank. The total antioxidant activity is expressed as the percent inhibition of the phosphor-molybdenum complex. The calibration curve was prepared by mixing ascorbic acid with methanol.

$$Total\ antioxidant\ capacity\ (\%) = [(Abs.\ of\ control - Abs.\ of\ sample)/(Abs.\ of\ control)] \times 100$$

2.3.6 Sensory evaluation of cookies

The sensory analysis was carried out using 10 members including student and staff members. The final sample has been analyzed on the basis of color, taste, appearance and sweetness. The sample was packed in a transparent package and presented with specific code. Water has been provided to every member to neutralize the previous effect of sample. For sample analysis 9-Point Hedonic scale showing least acceptable to most acceptable on selected parameters (9 is Excellent and 1 for Very poor).

2.4 Statistical Analysis

The experimental data were analyzed statistically. Analysis of variance and Duncan's multiple range tests were used to determine and compare the statistical differences of each data in this study. p-Value of less than 0.05 was regarded as the significance of result.

3. RESULTS AND DISCUSSION

3.1 Nutritional Analysis

The nutritional analysis shown in Table: 2, moisture content obtained from all samples was

compared with the moisture content of regular cookies (2.5-3%) [1]. The incorporation of rosehips and hibiscus affects the moisture content of each formulation (Fig. 1). Due to the presence of fiber the water binding ability increases. More hydroxyl groups of cellulose in fiber were able to bind with free water molecules through hydrogen bonding (1). The ash content differs in each formulation as the level of rosehips and hibiscus substituted, but it has no significant difference. Crude fiber content for control cookies was found to be significantly low when compared to the rosehips and hibiscus powder substituted cookies. It shows that incorporation of rosehips and hibiscus powder has definitely increased the fiber content, but it also changes with the percent of incorporation.

From the results in Table: 2, Each formulation has specific crude protein content but has no significant difference among them.

3.2 Analysis of Bioactive components and Antioxidant activity

High vitamin C content (300-4000 mg/100 g) of fruit of rosehip and other substances ensure the normal functioning of the endocrine glands, brain, heart and liver [15]. In the study we have found different levels of ascorbic acid in each formulation of cookies. Due to baking the concentration of ascorbic acid has decreased. After processing as shown in Table 3, the levels of ascorbic acid analyzed ranging from 564.2±0.12 (CFI), 521.7±0.47 (CFII), 477.4±0.14 mg/100 gm (CFIII). The ascorbic acid present in the cookies has incorporated little tartness to cookies. The total polyphenolic content calculated as mg Gallic acid equivalent (GAE) of cookies in aqueous solution. The total phenol content ranging from 151.25±0.02 (Formulation: I), 155.00±0.04 (Formulation: II),131.10±0.49 mg GAE/100gm (Formulation:III). The results of total antioxidant capacity can be observed in Table: 3 (observations in percent inhibition), showed a significantly lower for control cookies compared to the substituted cookies. The antioxidant activity could possibly because of the higher amount of vitamin C and the phenolic compounds present in rose hips and hibiscus powder (Fig. 2).

Anthocyanins water-soluble compounds having a great interest in nutrition and medicine because of their potent antioxidant capacity. Generally, environmental factors (temperature and light, latitude and altitude) have an impact on anthocyanin formation. Therefore, when studying the influence of latitudinal, altitudinal or temporal variations on anthocyanin production, environmental factors such as light and temperature are of prime importance. (2). Our findings sh 24.41±0.01 (CF1), 24.30±0.11 (CF2), 23.14±0.06 (CF3) mg/100 gm Anthocyanin content in cookies.The most common flavonoids in plants are quercetin and kaempferol and usually occur as glycosides. In rose hips there are mainly glycoside derivatives of quercetin: quercitrin (quercetin-3-O-rhamnoside), isoquercitrin (quercetin-3-O-glucoside) and hyperoside (quercetin-3-O-galactoside) [2]. As mentioned in the Table 3, the flavonoid content in different formulations is 9.5±0.16 (CF1) 8.0±0.17 (CF2) 6.46±0.20 (CF3). The total antioxidant (% inhibition) was detected in different formulations is 24.63±0.31 (CF1) 24.50±0.59 (CF2) 21.71±0.32 (CF3), which can be due to the presence of ascorbic acid and poyphenols present in the rosehips and hibiscus.

3.3 Analysis of Physical Parameters

Diameter of cookies has long been used to determine the quality of flour for producing cookies Table 4, shows that there was a significant decrease in the diameter of cookies (with different formulations) (CF1,CF2,CF3) after incorporating cookies with Rose hips and hibiscus powder. There was a slight decrease in the thickness, spread ratio, volume when compared to control (C) (Fig. 3).

3.4 Sensory Analysis

Cookies supplemented by different levels of substitutions of rose hip and hibiscus powder were sensory evaluated and compared with control biscuits 100% wheat flour. Data indicated that the percent score of cookies containing 15% Rose hip, 20% Hibiscus were found to be the most acceptable (over all acceptability). One-way analysis of variance (ANOVA) was performed to evaluate the significant differences between sample means for sensory analysis of cookies, with significant level being considered at P =0.05 Mean comparisons were assessed by Duncan's test, with the values expressed as means ± standard deviations. All data presented are mean values of (n= 10).

Table 2. Proximate analysis of cookies

Types of cookie sample (Formulation)	Composition %					
	Moisture	Ash	Crude fiber	Crude fat	Crude protein	Carbohydrate
Control (C)	1.041±0.002^{c}	0.973±0.004	0.211±0.01^{c}	20.366±0.449^{a}	4.32±0.03^{a}	73.08±0.08^{a}
20%Rosehip, 15%Hibiscus (CF1)	3.890±0.169^{a}	1.29±0.054^{a}	0.717±0.05^{b}	20.033±0.047^{a}	4.42±0.08^{a}	68.94±0.77^{d}
15%Rosehip, 20%Hibiscus (CF2)	3.223±0.301^{b}	1.28±0.051^{a}	0.710±0.05^{b}	20.100±0.081^{a}	4.27±0.229^{a}	71.12±0.02^{b}
15%Rosehip, 15%Hibiscus (CF3)	3.823±0.147^{a}	1.28±0.047^{a}	1.418±0.04^{a}	20.133±0.047^{a}	4.52±0.047^{a}	70.24±0.01^{c}

Data represented are mean ± standard deviation for (n =3); Means with different letters within a row are significantly different from each other at p=0.05 as determined by Duncan's multiple range tests

Table 3. Analysis of bioactive compounds and vitamin C in cookies

Formulations of cookies	Total phenolic content mg GAE/gm	Flavonoid content mg /100 gm	Anthocyanin content mg/100 gm	Vitamin C content mg/100 gm	Total antioxidant % inhibition phosphor-molybedenum (ascorbic acid)
Control (C)	105.00±1.54^{d}	0.5±0.12^{d}	00±00^{c}	00±00^{d}	10.80±0.75^{c}
20% Rosehip, 15% Hibiscus (CF1)	155.00±0.04^{a}	9.5±0.16^{a}	24.41±0.01^{a}	564.2±0.12^{a}	24.63±0.31^{a}
15% Rosehip, 20% Hibiscus (CF2)	151.25±0.02^{b}	8.0±0.17^{b}	24.30±0.11^{a}	521.7±0.47^{b}	24.50±0.59^{a}
15% Rosehip, 15% Hibiscus (CF3)	131.10±0.49^{c}	6.46±0.20^{c}	23.14±0.06^{b}	477.4±0.14^{c}	21.71±0.32^{b}

Data represented are mean ± standard deviation for (n =3); Means with different letters within a row are significantly different from each other at p=0.05 as determined by Duncan's multiple range tests

Table 4. Physical parameters of developed cookies

Formulations	Diameter (cm)	Thickness (cm)	Volume (cm3)	Density (g/cm3)	Spread ratio(D/T)
Control (C)	5.2±0.08 [a]	1.63±0.04 [a]	34.63±0.02 [a]	0.38±0.01 [b]	3.19±0.004 [a]
20%Rosehip,15% Hibiscus (CF1)	4.43±0.16 [b]	1.59±0.004 [ab]	24.49±0.01 [b]	0.47±0.01 [a]	2.77±0.009 [c]
15%Rosehip,20% Hibiscus (CF2)	4.41±0.014 [b]	1.53±0.04 [bc]	23.35±0.02 [c]	0.46±0.02 [a]	2.89±0.004 [b]
15%Rosehip,15% Hibiscus (CF3)	4.40±0.29 [b]	1.52±0.04[c]	23.10±0.02[d]	0.45±0.02 [a]	2.89±0.004 [b]

Data represented are mean ± standard deviation for (n =3); Means with different letters within a row are significantly different from each other at p=0.05 as determined by Duncan's multiple range tests

Table 5. Sensory analysis of final cookies

Formulations of Cookies	Sensory parameters				
	Color	Texture	Appearance	Taste	Overall acceptability
Control (C)	7.6±0.54 [a]	5.2±0.44 [b]	7.2±0.83 [b]	5.8±1.64 [b]	6.0±0.44 [c]
20% Rose hip, 15% Hibiscus (CF1)	7.2±0.83[a]	6.8±1.09[a]	7.1±0.54 [b]	7.4±0.89[a]	7.0±0.63[b]
15% Rose hip, 20% Hibiscus (CF2)	7.8±0.83[a]	7.4±0.54[a]	8.0±0.70[a]	6.2±1.30[b]	8.0±0.94[a]
15% Rosehip, 15% Hibiscus (CF3)	7.2±0.78 [a]	6.9±0.73 [a]	7.1±0.94 [b]	6.7±0.78 [ab]	6.8±1.09 [b]

Data represented are mean ± standard deviation for (n =3); Means with different letters within a row are significantly different from each other at p=0.05 as determined by Duncan's multiple range tests

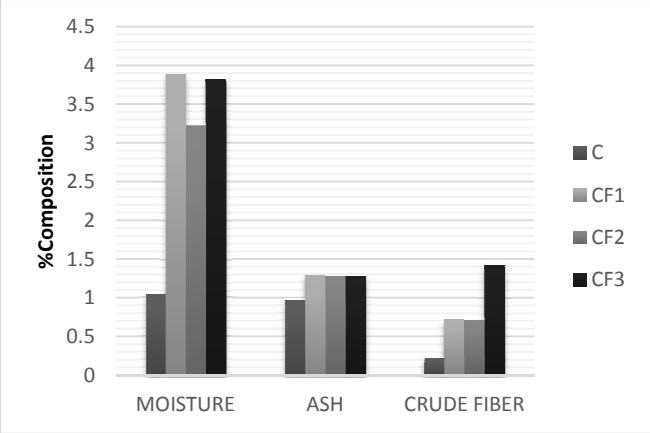

Fig.1. Physicochemical properties on different concentrations
**Data represented are mean ± standard deviation for (n =3)*

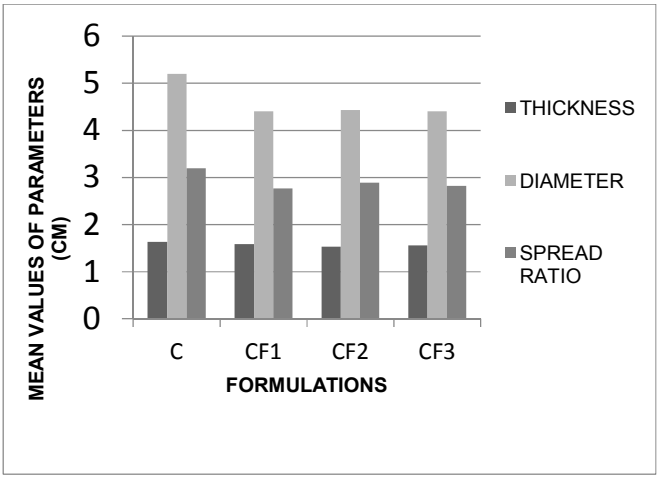

Fig. 2. Effects of treatments on thickness, diameter and spread rate
**Data represented are mean ± standard deviation for (n =3)*

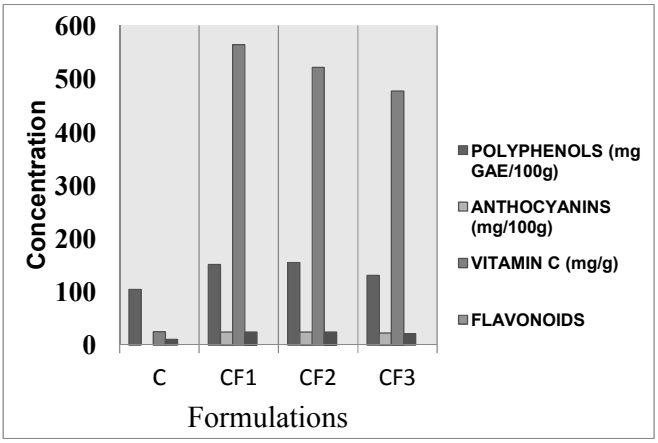

Fig. 3. Concentration of bioactive compounds for different formulations
**Data represented are mean ± standard deviation for (n =3)*

4. CONCLUSION

Numerous studies have been carried out in order to replace wheat flour made from fruit residue, petal of flowers and food waste in preparation of bakery products such as cookies and biscuits, due to the new consumption trends and eating habits. Food with high nutritional value is in great demand for proper functioning of body systems and potential health benefits. The analysis revealed that the increment in substitution in flour has increased the nutritive value of cookies when compared to control cookies. The substitution with Rose hips and Hibiscus powder has a significant share in enhancing the TPC content, Total antioxidant capacity and other bioactive compounds in cookies. Cookies prepared from formulation second (15% rose hips and 25% hibiscus powder) was more acceptable than other formulations on the basis of sensory evaluation. Substitution with Rose hips and Hibiscus could bring much potential health benefits to the consumers by adding nutritional as well as organoleptic properties.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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