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Functional and antioxidant properties of novel snack crackers incorporated with *Hibiscus sabdariffa* by-product



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ABSTRACT

The Hibiscus sabdariffa calyxes' residue (HSR) remained after the extraction of beverage is discarded which contributes to environmental pollution. The objective of this study was to explore the suitability of incorporating different amount of HSR (0%, 1.25%, 2.5%, 3.75%, and 5.0%) in crackers to enhance dietary fiber and antioxidant content. Physicochemical properties, antioxidants activity, nutritional quality, sensory profile and microstructure properties of samples containing HSR were examined and compared with control crackers. Cracker protein and fat levels decreased as HSR increased from 0.0% to 5% while ash increased. The total dietary fiber DF increased from 3.36% to 8.17% where the highest DF was reached at 5% HSR. The content of phenols increased from 5.99 to 17.57 mg/g and total flavonoid content increased from 49.36 to 104.63 mg/g of crackers incorporated with 5% HSR. DPPH radical scavenging activity increased two fold by increasing HSR up to 5%. HSR containing crackers exhibited darker L values than none/less HSR containing ones. In sensory ranking tests, acceptable crackers with pleasant flavor were obtained by incorporating up to 3.75% HSR into the cracker's formula. Crackers prepared with 5% HSR received the poorest sensory rating compared to non/less HSR enriched cracker. Scanning electron microscopy (EM) images of the prepared crackers revealed marked changes caused by incorporating HSR as upon HSR addition the surface was observed to be scratched, cracker and rougher. Overall results suggest that HSR is a potential functional food ingredient high in fiber content and antioxidants activity that may be processed into flour and used in food applications, such as baked goods.

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Introduction

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The increasing demand by the modern consumer for health food that are quick sources of good nutrition has prompted the food industry to develop food like snacks that combine convenience and nutrition. Many attempts are being made to improve snacks' nutritive value and functionality by modifying their nutritive composition. Such effects are very often achieved by increasing the nutrient density in basic recipes [1-4]. Additionally, a very important aspect of food's function-

2090-1232 © 2014 Production and hosting by Elsevier B.V. on behalf of Cairo University. http://dx.doi.org/10.1016/j.jare.2014.07.002 ality is its antioxidative capacity since there is much scientific evidence indicating the important role of food antioxidants in the prevention of different types of cancer and coronary heart diseases [5]. Among snack foods, crackers remain a versatile food, which is highly consumed by a wide range of populations, due to their varied taste, long shelf life and relatively low cost. It serves as a proper vehicle to meet consumer demand for nutritious, convenient, and tasty snacks.

Hibiscus sabdariffa L. is one of the most common flower plants grown worldwide and is used to make jellies, jams and beverages. Recently, it has gained importance as a soft drink material in many parts of the world. It is a good source of major nutrients i.e. phytochemical and antioxidant compounds activity [6,7]. The remaining calyxes after preparing the Hibiscus drink are usually disposed as a by-product without effort to exploit its usefulness and benefits. Generally, agro-food processing waste creates huge environmental, economic and social problems. There is now a growing recognition that the twin problems of waste management and resource depletion can be solved together through the utilization of waste as a renewable resource.

Preliminary experiments conducted in our laboratory showed that *H. sabdariffa* calyxes remaining after drink preparation (HSR) are characterized by high dietary fiber content, low fat content and considerable proportion of other biologically active compounds mainly polyphenols (data not shown). Alternatively, the absence of gluten in HRS makes it a very interesting raw material for application in the baking industry.

However, to the best of our knowledge, there is no published data on the food utilization of the residue remained after the preparation of *H. Sabdariffa* beverage. Therefore, this study was undertaken to investigate the possibility of using aforementioned raw materials for production of nutritionally improved crackers with respect to minerals (K, Ca, Mg, Mn, Fe and Zn) content, dietary fiber content, phytochemical compounds and antioxidant activity. In this study HSR was incorporated in different amounts (0%, 1.25%, 2.5%, 3.75%, and 5.0%) in the snack cracker formula. Physicochemical measurements, microstructure, and sensory quality of the developed crackers were tested.

Material and methods

Materials

All purpose wheat flour, salt, sugar, sun flour oil and baker's yeast were procured from a local supermarket. *H. sabdariffa* L. calyxes remaining after preparing a beverage were washed with water, spread in trays and dried at 40 °C forced air oven for 18 h to the moisture level of around 10%. The dried calyxes were powdered using coffee grinder and were sieved through a 150 mm sieve.

Chemicals

2,2-Diphenyl-1-picrylhydrazyl (DPPH) isoflavone and phenolic acid standards were obtained from Sigma–Aldrich (Germany). All other chemicals and solvents were of analytical grade.

Methods

Cracker preparation

Cracker samples were prepared in a straight dough process according to the recipe of commercial all purpose wheat flour (100% flour basis, fb), sugar (2%), salt (2%), bakers' yeast (2%) and water (60–64%), depending upon the percent of HSR in the formula. HSR was replaced wheat flour at varied amount as follows: 0%, 1.25%, 2.5%, 3.75% and 5% (i.e. if 5% HSR was added then the flour amount was reduced to 95%). Ingredients were mixed into cohesive dough, rolled into a consistent, thin sheet using a pasta roller and cut with rectangular mold into pieces after proofing. The crackers were baked in a forced-air convection oven (MMM Einrichtung GMbH, Germany) at 210 °C for 15 min. Baked samples were then cooled at ambient temperature, grounded in a standard coffee grinder to pass through a 40-mesh sieve, and stored in sealed bags in desiccators at room temperature.

End-product evaluation

Stack height, stack weight, specific volume, color, moisture and pH of the end products were determined. The stack height and stack weight were measured using seven sample pieces. The stack height was measured by a vernier caliper (Sakara, electronic digital vernier caliper), measuring once and turning the crackers 90° and measuring again to obtain the average value. The cracker specific volume was determined by dividing the volume by weight. The pH of samples was measured according to method 943.02 AOAC [8] in a glass electrode pH-meter Jenway-3505. All measurements were conducted at least three times.

The color ($L a^* b^*$) values of the crackers were determined by Hunter, Lab Scan XE – Reston VA, USA. The crackers were ground prior to color analysis. The instrument was standardized each time with white tile of Hunter Lab Color Standard. The lightness (L^*), redness (a^*) and yellowness (b^*) of the samples were recorded. The value taken was an average of three readings. The total color difference (ΔE) between the control and the HSR containing crackers was calculated as follows:

$$\Delta E = \left[(Lc - Ls) + (ac - as) + (bc - bs) \right] \frac{1}{2}$$

where subscript c = control and subscript s = samples containing HSR.

Macro-nutritive composition of crackers

Protein, fat and ash contents were estimated using the standard methods of analysis AOAC [8]. Soluble, insoluble and total dietary fiber content was investigated by employing enzymatic treatment of the samples AOAC [8]. Available carbohydrate was obtained by difference, by subtracting the sum of grams of water, protein, fat, ash and dietary fiber from a 100 g basis mass. The method has been chosen due to its simplicity, since it has been proven to be as accurate as other commonly used methods for estimation of available carbohydrates in starchy foods [9]. Mineral analysis of crackers was done by the procedure described in AOAC [10] method No. 3.014-016. The mineral content i.e. K, Ca, Mg, Mn, Fe, and Zn and were estimated by using Atomic Absorption Spectrophotometer (A Analyst 100, Perkin Elmer, Norvalk, C.T., USA) in acetylene air flame at wavelengths: 422 nm, 248 nm, 325 nm, 214 nm and 279.5 nm, respectively.

Total phenolic and total flavonoids content

The total phenolic content of the crackers were determined based on the Folin–Ciocalteu (FC) method [11]. In brief, 50 μ L of the sample extract was mixed with 250 μ L of freshly prepared FC reagent. After incubation for 5 min at room temperature, 0.75 mL of (7.5%, w/v) sodium carbonate solution and 3 ml distilled water were added and the solution was mixed thoroughly and incubated for 60 min at room temperature. Followed by this, absorbance was measured using a UV–visible spectrophotometer (Jenway 6715) at 765 nm. A suitable calibration curve was prepared using standard Gallic acid solution. All the results were expressed as mg Gallic acid equivalents (GAE) per gram of sample.

Total flavonoids in extracts were determined using the aluminum chloride colorimetric method of Change et al. [12]. The appropriate dilution of extracts (0.5 ml) was mixed with 1.5 ml of 95% ethanol, followed by 0.1 ml of 10% aluminum chloride, 0.1 ml of 1 M potassium acetate and 2.8 ml of distilled water. After incubation at room temperature for 30 min, the absorbance of the reaction mixture was measured at 415 nm. The flavonoid content was calculated using a standard calibration of rutin solution and expressed as micrograms of rutin equivalent (RE) per gram of sample.

DPPH radical scavenging activity

The capacity of the crackers extracts to scavenge DPPH radicals (2, 2-diphenyl-1-picrylhydrazyl) was measured based on the method described by Sanchez-Moreno et al. [13]. The results obtained were expressed as the percentage inhibition of DPPH based on the following formula:

Percent inhibition of DPPH =
$$[(A_{\text{control}} - A_{\text{sample}})/A_{\text{control}}] \times 100$$

where A_{control} is the absorbance of the DPPH solution without sample extract and A_{sample} is the absorbance of the sample with DPPH solution.

Microstructure properties

The crackers were cut and mounted on aluminum stubs using double adhesive tape. The samples were sputter-coated with gold-palladium to render thermoelectrically conductive by using (Edwards S150A Sputter Coating Device) and then scanned using JXA-840A scanning electron microscope (JEOL, Tokyo-Japan). The micrographs were taken at magnification of 50× for the surface and cross section parts of the crackers.

Sensory evaluation

The sensory evaluation of different treatments of crackers for various attributes including color, taste, crispness, odor, appearance and overall acceptability was carried out by 14 trained taste panel using seven hedonic score system as described by Meilgaard et al. [14]. On the day of evaluation, crackers from all the treatment were placed in transparent plates, labeled with three digit random codes. Panelists were given distilled water to neutralize their mouth between the samples. The samples were presented in random order and judges were asked to rate their acceptance by giving a score for all the parameters.

This study has been assessed and approved by the National Research Centre Ethics Committee. Consent was sought from panelists participating in this study. Samples were prepared according to good hygiene and manufacturing practices. Participants were informed about the study and explained that their participation was entirely voluntary, that they could stop the interview at any point and that the responses would be anonymous.

Statistical analysis

Statistical analyses were conducted with the SPSS software 17 (SPSS Institute, Chicago, USA). The mean and standard deviation of parameters from proximate analysis, physical properties, and color analyses were calculated and differences between the formulations were evaluated by analysis of variance (ANOVA) with significant level being considered at P < 0.05. Mean comparisons were assessed by Duncan's multiple range test, with the values expressed as means \pm standard deviations.

Results and discussion

End product evaluation

Physical characteristics of crackers were measured to determine the effect of supplementation of HSR on stack weight, stack height, and specific volume Table 1. The stack weight, stack height, and specific volume of crackers decreased as the addition level of HSR increased. In other words, HSR containing crackers were denser compared to the control samples. The presence of gluten protein in wheat flour was presumed responsible for the increased volume and height. Gluten develops when wheat flour is mixed with water and it forms a matrix that retains more gas. As the dough is baked, it expands more, thereby increasing loaf volume and height. However, addition of HSR to wheat flour lowered the amount of gluten in the blends, causing poor gas retention and thus reduced loaf volume and height of the blended crackers. The presence of relatively large particle fibers in HSR might also have played significant roles in reducing crackers volume by puncturing gas cells as the dough was expanding. The results are in agreement with findings by Wu et al. [15].

Moisture contents of crackers were assessed to determine storability of the products. The results indicated that the moisture contents among samples containing HSR were statistically different from each other, and that HSR crackers had a significantly lower amount of moisture compared to control samples. The low moisture content of the products is important for prolonging their shelf life. In addition, water content of baked products is of interest in the degree of crunchiness as well as stability of phenolic compounds. It has been suggested that hydrolysis may have a role in phenolics degradation, and cleavage of isoflavones esters to glucosides occurs via

Table 1 End-product quality evaluation on dry basis.						
Samples	Stack height* (mm)	Stack weight (g)	Specific volume (mm/g)	Moisture	pH	ΔE^{**}
Control	$69.67^{\rm a} \pm 3.01$	$65.66^{a} \pm 1.09$	$1.06^{\rm bc} \pm 0.03$	$8.30^{a} \pm 0.51$	$5.82^{a}\pm0.06$	0.0
HSR-1.25%	$59.33^{b} \pm 4.37$	$53.04^{b} \pm 4.32$	$1.12^{ab} \pm 0.07$	$6.07^{\rm b} \pm 0.62$	$4.88^{b} \pm 0.02$	2.52
HSR-2.50%	$56.33^{bc} \pm 1.76$	$49.53^{\rm bc} \pm 0.47$	$1.14^{ m ab}\pm0.07$	$6.06^{\rm b} \pm 0.16$	$4.78^{\circ} \pm 0.02$	4.46
HSR-3.75%	$56.67^{bc} \pm 2.08$	$48.96^{bc} \pm 0.55$	$1.16^{\rm a}$ \pm 0.06	$6.64^{\rm b} \pm 0.06$	$4.44^{d} \pm 0.00$	4.40
HSR-5.0%	$53.50^{\rm c} \pm 1.00$	$48.44^{c} \pm 1.38$	$1.10^{ m abc}\pm0.03$	$6.28^{b} \pm 0.03$	$4.09^{e}\pm0.03$	4.88

 Table 1
 End-product quality evaluation on dry basis.

Data were the mean value \pm S.D.

Values in the same column followed by the same letters are not significantly different (P > 0.05).

Standard deviation was at least three replicate experiments.

* Stack of 7 crackers.

** ΔE : total color difference between control and treatment.

hydrolysis [16]. The loss of water in the form of steam may have consequences in the ability of oxygen to intercept and oxidize phenolics.

The pH of the end products ranged from 5.82 for control to 4.09 for 5% HSR containing crackers which is lower than commercial product. With the increasing level of HSR, more acids were exist contributed to higher value bulk density/lower volume as more acids modify and decrease the spread of the dough. This in turn resulted in decreased stack height and stack weight of crackers. The preservative properties of organic acids also enhance crackers' microbiological and physicochemical stability.

Fig. 1 shows the effect of HSR incorporation on the color of finely grinded cracker snacks. The L^* 'lightness' a^* 'redness' and b^* 'yellowness' values varied significantly (p < 0.05) with inclusion of HSR into crackers. L^* values that correspond to whiteness or lightness decreased with increase in HSR to the crackers and ranged from 74.62 for control to 51.48 for 5% HSR crackers as expected, the color of the end products gradually became browner and darker with the increased ratio of HSR. This result is in accordance with the sensory test where control perceived the lighter color among all treatments and 5% HSR the darkest one.

The a^* values which correspond to red–green profile ranged from 6.73 for control to 10.42 for 5% HSR crackers, highlighting the obvious redness in the color profile. The b^* values which represents yellowness were decreased by the increase of HSR from 1.25% to 5%. Higher b^* values which corresponded to yellow–blue profile ranged from 27.44 to 23.08 for control and 5% HSR respectively. Higher b^* values comparatively indicate the samples exhibited more yellowish color.

In terms of the total color difference (ΔE) between the control and crackers containing the HSR, all sample exhibited ΔE value higher than control sample Table 2. This means that all of them were darker than the control, having lower values of L, higher values of b and a. Consequently, there was a reduction in the typical golden or very light brown color of the commercial crackers.

Macronutrients composition of crackers

Macro-nutritive evaluation of the formulated crackers including total protein, crude fat, ash, and dietary fiber varied significantly (p < 0.05) Table 2. The addition of varying amounts of HSR in wheat flour changes the biochemical composition of the resulting crackers. Total protein content ranged from 10.43 g/100 g for 1.25% HSR crackers to 9.70 for 5% HSR



Fig. 1 L^* : lightness; a^* : redness; b^* : yellowness. (%)^{*} denotes percent *Hibiscus sabdariffa* residue addition rate of 0%, 1.25%, 2.5%, 3.75%, 5% to snack crackers.

crackers. By increasing the level of HSR incorporation, protein content in the final product decreased accordingly. Same trend was reported for cured fat content of crackers where fat was 7.45% for control and 5.30% for 5HSR crackers. The ash content of samples was high, ranging from 2.41% in control to

Table 2 Macronutrients composition of crackers.					
	Control	HSR-1.25%	HSR-2.50%	HSR-3.75%	HSR-5.0%
Protein %	$10.27^{\rm a} \pm 0.34$	$10.43^{\rm a}$ \pm 0.35	$9.80^{b} \pm 0.33$	$9.81^{b} \pm 0.33$	$9.70^{\circ} \pm 0.33$
Ash %	$2.41^{\circ} \pm 0.18$	$2.47^{c} \pm 0.20$	$2.41^{\circ} \pm 0.14$	3.01 ± 0.20^{b}	$3.38~^{a}~\pm~0.09$
Fat %	$7.45^{ m a}\pm0.45$	$6.66^{\rm b} \pm 0.40$	$6.15^{b} \pm 0.02$	$5.35 \pm 0.04^{\circ}$	$5.30^{\rm c} \pm 0.20$
TDF %	$3.36^{d} \pm 0.49$	$3.69^{cd} \pm 0.11$	$4.25^{\rm c} \pm 0.11$	$6.08^{b} \pm 0.17$	$8.17^{a} \pm 0.19$
Carbohydrate ^a	76.51	76.75	77.39	75.75	73.45
Energy (kcal)	420.89	418.60	412.61	402.55	396.64
Macro minerals mg/g					
K mg/g	$45.75^{a} \pm 3.18$	$50.70^{\rm a}$ \pm 5.23	$54.75^{a} \pm 14.49$	$58.50^{a} \pm 15.55$	$57.75^{a} \pm 23.68$
Ca	$30.25^{a}\pm0.35$	$30.75^{a} \pm 2.47$	$31.00^{a} \pm 6.36$	$34.25^{a} \pm 3.18$	$38.25^{a} \pm 1.76$
Mg	$10.50^{\rm b} \pm 3.53$	$11.0^{\rm b} \pm 5.65$	$11.00^{\rm b} \pm 1.41$	$12.00^{\rm b} \pm 6.36$	$28.75^{a} \pm 7.42$
Mn	0.36 $^{\rm a}$ \pm 0.21	$0.44^{a} \pm 0.16$	$0.42^{a} \pm 0.25$	$0.58~^{a}~\pm~0.31$	$0.67^{a}\pm0.06$
Micro minerals mg/g					
Fe	$1.46^{\rm c} \pm 0.48$	$1.68^{b} \pm 0.21$	$2.99^{a} \pm 1.53$	$3.39^{\rm a}$ \pm 2.55	$3.43^{a} \pm 0.50$
Zn	$1.76^{a} \pm 0.27$	$1.84^{a} \pm 0.37$	$1.72^{a} \pm 0.22$	$1.91^{a} \pm 0.47$	$2.29^{a} \pm 1.02$

Values are presented as means \pm SD.

Values in the same column followed by the same letters are not significantly different (P > 0.05).

^a By difference.

3.38% in sample enriched with 5HSR. Dietary fiber content was significantly increased in all cracker samples and ranged from 3.36% in the control samples to 8.17% in 5% HSR enriched sample. These results would be expected because HSR contains higher level of dietary fiber compared to all-purpose wheat flour [17].

Dietary fiber is a collective term for a variety of plant substances that are resistance to digestion process of human gastrointestinal enzymes. Several studies revealed that fiber-rich foods have important effect of serum cholesterol [5,18]. Hence, enrichment of cereal bakery products with HSR would enhance the nutritional quality of the product and diversify the sources of fiber other than cereal bran sources.

The contents of protein, carbohydrate, fat and fiber were converted to food energy using an Atwater general factor system according to FAO recommendations [19]. Conversion factors used were 4.0 kcal g^{-1} for proteins and carbohydrates; 9.0 kcal g^{-1} for fats; 2.0 kcal g^{-1} for dietary fiber. A stepwise decrease in energy value was observed among the blends from 420.89 kcal/100 g of dry matter for control crackers against 396.64 kcal/100 g of dry matter for 5% HSR containing crackers. According to the acceptable macronutrient distribution ranges for energy given by dietary reference intakes (DRIs) according to FNB Food [20] consumption of examined crackers provides a balanced intake of energy derived from proteins as well as carbohydrate and low energy derived from fat. In contrast to these nutrients profile, partial replacement of crackers with HSR result in significant increases of dietary fiber content in the products which in turn decrease energy compared to the control (Table 2).

Supplementation of crackers with HSR significantly (p < 0.05) increased the levels of magnesium from 10.50 for control to 28.75 for 5% HSR crackers; and iron increased from 1.64 for control to 3.43 for 5% HSR crackers respectively Table 2. Crackers contents of potassium, calcium, magnesium and zinc were not significantly affected by the addition of HSR. The relatively higher iron content demonstrated the potential health benefit of the HSR supplemented crackers. Based on the results, supplementation with HSR in the product may satisfy the nutritional needs of the consumers.

Phytochemical contents and antioxidant capacity of snack crackers

Since HSR represents a multiple sources of antioxidants, including total phenolic content and flavonoids, the following work focuses on how the incorporation of HSR in the prepared crackers influences the antioxidant capacity of the final products. Total phenolic compounds TPC were quantified by the Folin–Ciocalteu method which is an electron transfer based assay and measures reducing capacity [11]. The TPC values reported varied among the different HSR containing crackers as shown in Table 3. Incorporation of up to 5 g of HSR/100 g of formulated product significantly increased TPC when compared to the control.

This trend was particularly evident across the different concentration and total flavonoid content TFC ranged from 49.36 for control to 104.63 µg rutin/g for 5% HSR. TFC of the crackers increased by increasing the HSR percent and the respective TFC were ranked in the following order as: HSR-5.0 > HSR-3.75 > HSR-2.5 > HSR-1.25 > control.

The antioxidant activity of the crackers was measured by DPPH radical scavenging activity (RSA) assay. Prepared crackers contained remarkably different antioxidant properties in terms of DPPH radical scavenging activity (RSA). With the highest level of HSR incorporation i.e. 5% a two fold increase in DPPH-RSA was observed. The increase in TPC and DPPH-RSA was also reported when crackers were formulated with other food based by-products such as mango peel powder [21].

HSR enriched crackers were characterized with higher antioxidant potential in comparison to control due to the incorporation of phenolic compounds, which had been shown to possess antioxidant activity [22,23]. Accordingly, HSR crackers could be developed as a functional food with more effective antioxidant properties.

Microstructure properties of snack crackers

Figs. 2 and 3 show surface as well as cross sectional morphologies of the formulated crackers. With regard to surface section, the control crackers displayed a smooth surface with no

Table 5 Antioxidant properties of disease entended with tiger hat nour.					
Samples	TPC ¹ mg Gallic acid equiva lents/g	TFC ² mg Quercetin equivalents/g	RSA ³ (%)		
Control	$5.99^{ m c}\pm0.96$	$49.36^{\circ} \pm 2.13$	$12.68^{d} \pm 0.25$		
HSR-1.25%	$8.75^{b} \pm 0.21$	$58.56^{bc} \pm 0.17$	$15.01^{\rm c} \pm 1.09$		
HSR-2.50%	$9.95^{\rm b} \pm 0.21$	$60.04^{\rm b} \pm 0.70$	$15.93^{\rm c} \pm 0.06$		
HSR-3.75%	$9.89^{\rm b} \pm 0.27$	$62.92^{b} \pm 7.99$	$22.77^{\rm b} \pm 1.09$		
HSR-5.0%	$17.57^{\rm a} \pm 1.52$	$104.63^{\rm a} \pm 1.06$	$30.52^{a} \pm 0.19$		

 Table 3
 Antioxidant properties of biscuit enriched with tiger nut flour.

All data are the mean + SD of three replicates.

Means in a column with the same letter are not significantly different p > 0.05.

¹ Total phenolic content.

² Total flavonoids content.

³ Radical scavenging activities.



Fig. 2 SEM micrographs (50×) for surface structure (crust) of snack crackers ((A) control, (B) 1.25% HSR, (C) 2.5% HSR, (D) 3.750% HSR, (E) 5% HSR).



Fig. 3 SEM micrographs (50×) for inner structure (crumb) of snack crackers ((a) control, (b) 1.25% HSR, (c) 2.5% HSR, (d) 3.750% HSR, (e) 5% HSR).

fissures and few surface markings. The control crackers were also characterized as having a continuous structure that appeared disaggregated. Marked changes were observed by incorporating HSR where surface became scratched, cracked, and rougher. With the increase in the HSR incorporation the surface structure became more distorted surrounding structures.

The internal structures of the snacks were affected by the percent of HSR addition where large air cells obtained in cracker snacks with more HSR (Fig. 3). Control crackers were characterized by a number of large and intermediate air cells, in addition to many smaller cells. The control crackers, contrasted with the HSR incorporated crackers by having many

small, and few intermediate, air cells. In most of the HSR containing samples showed numerous fiber fragments adhering to surrounding granules. Increase in the addition of HSR up to 5% resulted in the formation of larger pore size and unbroken fibers while, less dense structure was revealed with less HSR. This result coincide with study of Bhattacharya and coworkers [24] whom reported that increasing fiber content into baked snack resulted in compact cell structure.

Crackers differ from cookies by being higher in moisture, and lower in sugar and fat. Both the formulations and baking processes used for crackers contribute to their characteristic flaky and open structure that distinguishes them from cookies. The lower amount of protein in the HSR incorporated crackers might results in the less developed gluten-based matrix, while the additional water in control crackers may enable gelatinization of a proportion of the starch within crackers during baking.

Sensory evaluation

Snack crackers incorporated with varied level of HSR were evaluated for their sensory qualities and general acceptability. Overall likeability of the different prepared crackers was compared using a 7-point hedonic scale. Sensory tests were performed at the National Research Centre taste panel facility using 14 panelists in each test. 57% of panelists were females and 43% male, in the 25–47 age range. The analysis of the sensory profiles of the five recipes of crackers indicated significant differences between the evaluated crackers at p < 0.05 (Oneway ANOVA) Table 4. Generally all crackers were scored higher than control except HSR 5% which was rated lower than control. Meanwhile, this crackers still acceptable since the mean scores were greater than a score of 2.5 (neither like nor dislike). The order of product acceptability based on 7point hedonic scale was 1.25 HSR > 2.50 HSR > 3.75 HSR > control > 5% HSR.

As the color differences among the prototype crackers were distinct, difference in mean color score was significant. Increase in the HSR content from 1.25% to 5% in the mixture decreased the color score. About 77% of color acceptability scores were in the "liking" range (5.0–7.0) of the 7-point hedonic scale, indicating that product color within the range of samples evaluated in this test is not likely to be a hindrance to product acceptability.

Difference for taste was significant with mean scores ranging from 5.61 to 3.5 across the treatments. Where, samples incorporated with 1.25 as well as 2.5% HSR had better effects on taste compared with other treatments and control formula. Crackers crispness score ranged from 3.43 for control to 5.32 for 2.5% HSR crackers. The incorporation of HSR improves the crispiness of HSR containing crackers compared to the control where 1.25% and 2.50% HSR crackers scored with higher intense values followed by 3.75% and 5% HSR crackers.

A marked similarity was reported for odor where cracker recipes containing 1.25% and 2.5% HSR scored higher than crackers with increased amount of HSR and control as well. For appearance cracker snacks which contained HSR received lower scores compared to control crackers. The surface characteristics of such crackers were negatively affected by the HSR addition. Panelists were also asked to indicate the one sample they most preferred. It was clearly evident that crackers containing 1.25% HRS was the most preferred one followed by crackers containing 2.5% HSR. In other words 1.25–2.5% is considered the desirable range for HSR incorporation. Although several panelists specifically commented on good crispiness of the crackers containing HSR, there were several comments regarding crackers incorporated with higher amount of HSR that indicated further investigation of other textural attributes. The high fiber nature of these products may have inhibited development of a texture resembling the commercial crackers familiar to many general consumers.

The acceptability of food products always rely on the food texture and crispiness was highlighted as the importance parameter especially in cracker or snacks product [25]. In addition, Siaw et al. [26] claimed that linear expansion which determines the crispiness had the least degree of tolerance in the acceptability of crackers.

In general the overall acceptability of the various crackers indicated that product within the range of samples evaluated in this test, is not likely to be a hindrance to product acceptability. Where at least 70% of scores in a sensory taste panel are > 5 on a 7-point scale, the potential for product success once modifications are made based on panelists' feedback is considered good. Both the 1.25HSR% and 2.5HR%, garnered over 80% of scores in this range for sensory attributes table.

Conclusions

Significant amount of potentially bioactive polyphenols remain in the residues of *H. sabdariffa* calvxes which is usually discarded. The goal of our research was to examine the possibilities of improving the quality of wheat flour based cracker by supplementing the basic recipe with different amounts HSR. It was found that as the amount of HSR increases; stack weight, stack height, and specific volume, moisture and pH of crackers decreased. Lower moisture as well as pH favors improved shelf life of the crackers. Crackers prepared with HSR exhibited lower protein, fat content and higher content of dietary fiber compared with control crackers. As dietary fiber has outstanding beneficial effects on human health, more nutritive and healthier crackers can be produced without affecting quality parameters negatively which also reduces the calorie with intake. Phenolic content has also a positive contribution on nutritional excellence of the developed cracker. Partial addition of up to 5% HSR produced light brownish crackers.

With respect to sensory quality, the product that had high crispness score was highly accepted by the panelists. Sensory

Table 4Sensory evaluation for Crackers at different supplementation ratio.						
Sample	Color	Taste	Crispiness	Odor	Appearance	Overall acceptability
Control	$6.32^{a} \pm 0.91$	$4.04^{b} \pm 1.52$	$3.43^{c} \pm 1.22$	$4.29^{ab} \pm 1.64$	$5.93^{a}\pm0.92$	$4.57^{bc} \pm 1.45$
HSR-1.25%	$5.32^{ab} \pm 1.07$	$5.61^{a} \pm 1.24$	$5.50^{\rm a}$ \pm 1.09	$5.32^{\rm a}$ \pm 1.56	$5.21^{ab} \pm 0.97$	$5.89^{\rm a} \pm 0.56$
HSR-2.5%	$5.07^{ab} \pm 1.69$	$5.57^{\rm a}\pm0.94$	$5.54^{a} \pm 1.42$	$4.57^{ab} \pm 1.87$	$4.93^{ab} \pm 1.59$	$5.21^{ab} \pm 1.42$
HSR-3.75%	$4.50^{\circ} \pm 1.51$	$4.25^{b} \pm 1.63$	$4.00^{\rm bc} \pm 1.57$	$4.35^{ab} \pm 1.65$	$4.43^{b} \pm 1.16$	$4.64^{\rm bc} \pm 1.00$
HSR-5.0%	$4.86^{\circ} \pm 2.38$	$3.50^{\rm b} \pm 1.99$	$4.97^{ab} \pm 1.57$	$3.85^{b} \pm 1.91$	$4.29^{b} \pm 2.23$	$3.82^{c} \pm 1.98$

Means for groups in homogeneous subsets are displayed.

Mean sample size = 24.

Values in the same column followed by the same letters are not significantly different (P > 0.05).

ratings for crackers containing 1.25% and 2.50% HSR replacement of wheat flour were positive, as evidenced by mean ratings that averaged 5.89 and 5.21 respectively. Specifically, taste, crispness odor and overall acceptability ratings for these crackers were superior compared with control cracker. Crackers with incorporated with up to 3.75% w/w are considered as acceptable as control crackers. However, when incorporation of HSR reached 5% w/w, the acceptability of all sensory traits was significantly lower than the control (p < 0.05). With the addition of marketing context factors (e.g. packaging and product information), consumers would likely be able to accept products with higher HSR percent.

This work indicated that DF, bioactive compounds, and antioxidant capacity are good reasons to foster the use of HSR as a source of antioxidant dietary fiber and it may be suitable for use as an ingredient in functional foods or nutritional supplements. In addition, utilization of HSR may surely minimize the production of waste from Hibiscus processing and contribute to the beneficial outcome of food industry.

Conflict of interest

The authors have declared no conflict of interest.

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References

- Ainsworth P, Ibanoglu S, Plunkett A, Ibanoglu E, Stojceska V. Effect of brewers spent grain addition and screw speed on the selected physical and nutritional properties of an extruded snack. J Food Eng 2007;81(4):702–9.
- [2] Ajila CM, Leelavathi K, Prasada Rao UJS. Improvement of dietary fiber content and antioxidant properties in soft dough biscuits with the incorporation of mango peel powder. J Cereal Sci 2008;48(2):319–26.
- [3] Stojceska V, Ainsworth P, Plunkett A, Ibanoglu S. The recycling of brewer's processing by-product into ready-to-eat snacks using extrusion technology. J Cereal Sci 2008;47(3):469–79.
- [4] Sun-Waterhouse D, Teoh A, Massarotto C, Wibisono R, Wadhwa S. Comparative analysis of fruit-based functional snack bars. Food Chem 2010;119(4):1369–79.
- [5] JMarlett JA. Dietary fiber and cardiovascular disease. In: Cho SS, Dreher ML, editors. Handbook of dietary fiber. New York (USA): Merkel Dekker publisher; 2001. p. 17–30.
- [6] Mahadevan N, Shivali KP. *Hibiscus sabdariffa* Linn: an overview. Nat Prod Rad 2009;8:77–83.
- [7] Chen CC, Hsu JD, Wang SF, Chiang HC, Yang MY, Kao ES, Ho YC, Wang CJ. *Hibiscus sabdariffa* extract inhibits the

development of atherosclerosis in cholesterol-fed rabbits. J Agric Food Chem 2003;51:5472–7.

- [8] AOAC. Official methods of analysis. 15th Eds. Association of Official Analytical Chemists, Inc., Washington, USA; 1990.
- [9] Menzes EW, de Melo AT, Lima GH, Lajolo FM. Measurement of carbohydrate components and their impact on energy value of foods. J Food Comp Anal 2004;17:331–8.
- [10] AOAC. Official methods of analysis. 17th Eds. Association of Official Analytical Chemists, Inc., Washington, USA; 2003.
- [11] Singleton VL, Rossi JA. Colorimetry of total phenolics with phosphomolybdic-phosphotungstic acid reagents. Am J Enol Viticult 1965;16:144–58.
- [12] Change C, Yang M, Wen H, Chern J. Estimation of total flavonoid content in propolis by two complementary colorimetric methods. J Food Drug Anal 2002;10:178–82.
- [13] Sanchez-Moreno C, Laurrari JA, Saura-Calixto F. A procedure to measure the antiradical efficiency of polyphenols. J Sci Food Agric 1998;76:270–6.
- [14] Meilgaard M, Civile GV, Carr BT. Sensory evaluation techniques. 4th ed. Florida: CRC Press; 2007.
- [15] Wu KL, Sung WC, Yang CH. Characteristics of dough and bread as affected by the incorporation of sweet potato paste in the formulation. J Mar Sci Technol 2009;17(1):13–22.
- [16] Patras A, Brunton NP, O'Donnell C, Tiwari BK. Effect of thermal processing on anthocyanin stability in foods; mechanisms of kinetics and degradation. Trends Food Sci Technol 2010;21:3–11.
- [17] Nelson AL. High fiber ingredients. Eagen Press; 2001.
- [18] Hainida E, Amin I, Normah H, Mohd-Esa N, Ainul ZAB. Effects of defatted deride Roselle (*Hibiscus sabdariffa* L.) seeds powder on lipid profiles of hypercholestrolemic rats. J Sci Food Agric 2008;88:1043–50.
- [19] FAO Food and Nutrition Paper 77. Food energy methods of analysis and conversion factors, Food and Agriculture Organization of the United Nation, Rome; 2003.
- [20] FNB Food and Nutrition Board. Dietary reference intakes for energy, carbohydrate, fiber, fat, fatty acids, cholesterol, protein, and amino acids (Macronutrients); 2005.
- [21] Ajila CM, Aalami M, Leelavathi K, Rao UJSP. Mango peel powder: a potential source of antioxidant and dietary fiber in macaroni preparations. Innov Food Sci Emerg 2010;11(1):219–24.
- [22] Ramirez-Rodrigues MM, Plaza ML, Azeredo A, Balaban MO, Marshall MR. Phytochemical, sensory attributes and aroma stability of dense phase carbon dioxide processed *Hibiscus sabdariffa* beverage during storage. Food Chem 2012;134:1425–31.
- [23] Nman NM, Onyeke NG. Chemical composition of two varieties of Sorrel (*Hibiscus sabdariffa* L.), calyces and the drink made from them. Plant Foods Hum Nutr 2003;58:1–7.
- [24] Bhattacharya S, Das H, Bose AN. Effect of extrusion process variables on microstructure of blends of minced fish and wheat flour. J Food Sci Technol 1990;27(1):22–8.
- [25] Roudaut G, Dacremont C, Vallès Pàmies B, Colas B, LE Meste M. Crispness: a critical review on sensory and material science approaches. Trends Food Sci Technol 2002;13:217–27.
- [26] Siaw CL, Idrus AZ, Yu SY. Intermediate technology for fish cracker (Keropok) production. J Food Technol 1985;20:17–21.