



Physicochemical and nutritional characteristics of nutrition bar fortified with cowpea protein

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Abstract

Cooked and oven-dried cowpea in different four concentrations of 0%, 10%, 20%, and 30% was incorporated into nutrition bars (CNB0, CNB10, CNB20, and CNB30 respectively). The nutrition bar contained other ingredients including roasted crushed peanuts, almonds, cashew, oatmeal, raisins, date syrup, hazelnut butter, glucose syrup, sorbitol, and glycerol. The bars were packed and stored at room temperature for 180 days and experiments were conducted at interwall of 30 days. Nutritional value, moisture content, hardness, organoleptic properties, and microbial test of the bars were evaluated. Nutritional values showed that CNB30 is generally more nutritious than other treatments with less energy value. Decrease in moisture content directly affected the hardness of nutrition bar during 6 months storage. However, water–protein interaction modified the textural properties of the samples. Addition of cowpea as a protein source resulted in greater organoleptic properties so that CNB30 received the highest score in overall acceptability even after 180 days with score 6. Microbial count of sample CNB30 was within the standard acceptable range showing the product remained stable and safe for consumption until the end of 180 day. Consequently, the use of cowpea protein in nutrition bars had significant impact on physicochemical properties and overall acceptability of the bars.

Keywords High-protein nutrition bars · Cowpea protein · Hardness · Nutritional value

Introduction

Cowpea (*Vigna unguiculata*) is an annual herbaceous legume, which belongs to the Fabaceae family and is mostly cultivated for human consumption and animal-feed in African countries [1]. Cowpeas might be used to provide wide variety of meals and snacks [2]. Cowpeas are rich in protein, fiber, and have a low glycemic index, making them very nutritious and potentially health-beneficial [3–5]. Besides containing a high proportion of carbohydrates (68% to 73%) [6], cowpea seeds are a potential novel source of proteins,

since they contain 18.2–30.4% protein [7]. Most of the cowpea grain proteins consist of globulins with lower levels of albumins, glutelin, and prolamins [8, 9]. The amino acid composition of the cowpea is rich in lysine, leucine, arginine and other essential amino acids; thus, can largely fulfil the essential amino acid requirements of a human diet. However, cowpeas are low in the sulfur amino acids (methionine and cysteine) compared to cereals and animal products and thus, for a balanced diet, cowpeas need to be supplemented with cereals or vegetables, meat and/or dairy products [10]. The lipid content of cowpea whole grain ranges from 0.5 to 3.9% [8]. With respect to fatty acids, linoleic acid and palmitic acid predominate followed by oleic acid, stearic acid and linolenic acid [8, 11]. Cowpeas are a source of essential minerals, calcium, magnesium, potassium, iron, zinc and phosphorus [7].

Snack bars are ready-to-eat convenient products in the shape of a bar occupying larger space in the consumer market which not only satisfy the hunger, but prove as a quality source of nutrients and a convenient means of replacement of a meal. They are made up of proteins, carbohydrates, fats, vitamins, and minerals [12, 13] and provide different

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functions such as energy boosting [14]. Employing a variety of components to appeal to a wide range of consumer groups [15], nutrition bars are fascinating healthy snacks among a wide range of people namely the youth, athletes, vegans, people who are on diet etc. [16]. These healthy snacks may contain cereals, nuts, nut paste, seeds, and dried fruits replacing junk foods. In this study, cowpea seed is offered as an ingredient in such bars with the concept of boosting energy. Nutritional and physicochemical properties as well as consumer preference during shelf-life were analyzed to determine the impact of protein addition to the nutrition bars.

Materials and methods

Materials

To improve the acceptability of the product for vegetarians, the food ingredients were preferentially chosen from plant origin; nevertheless, the presence of all main food categories was kept in mind. Cowpea grains, Almonds, Cashew, Peanuts, Oatmeal, Raisins, Date syrup, and Glucose syrup were purchased from the local markets in Tehran, Iran.

Preparation of nutrition bars

Granulated, steamed, and oven-dried cowpea, roasted crushed peanuts (8.5 g), almonds (10.5 g), cashew (8.5 g), and oatmeal (39.5 g), raisins (5.0 g), date syrup (13.5 g), sorbitol (2.5 g), glucose syrup (3.0 g), and hazelnut butter (9.0 g) were combined in a mixing bowl, poured into rectangle molds, and chilled for 2 h. The bars were then cooked for 30 min at 180 °C. The size of the bars was 30 × 11 × 120 mm and weighed approximately 35 g. They were packed individually in cellophane and aluminum foil and stored in fridge until further study. Four variants of the cowpea nutrition bars (CNB) were formulated: CNB0, CNB10, CNB20, and CNB30 with 0%, 10%, 20%, and 30% incorporation of cowpea, respectively. Samples were stored for 180 days and experiments were conducted at interwall of 30 days.

Proximate analysis

The fat content of samples was evaluated according to AOAC Method [17]. Sadasivam and Manickam's Anthrone technique was used to evaluate the carbohydrate content of the samples [18]. The protein content and total fiber were determined according to the methods described in AACC [19]. The moisture content of the samples was evaluated using oven method [20]. The Atwater factor method was used to calculate the energy value of each sample [21]. The standard procedure outlined by Sadasivam and Manickam

was used to determine sodium, potassium, and iron in each sample [18]. Calcium and zinc were measured using the titrimetric method and atomic absorption spectrophotometry, respectively [19]. Employing the phosphovanado-molybdate colorimetric technique, the phosphorus was calculated by generating color with ammonium molybdate solution [17].

Texture hardness

Hardness of cowpea nutrition bars was studied by the texture analyzer (Texture Analyzer, TA-XT2, UK) at room temperature. This was done to test the products' hardness. The probe height was adjusted to 5.0 mm from the sample and the measurement mode settings compression was set to a constant speed of 1 mm/s. The samples were compressed to 50% of their original size. All of the measurements were carried out in triplicate on each sample.

Sensory evaluation

Sensory evaluation was carried out to know the level of acceptance by consumers. It is used to analyze and interpret the responses to products that are perceived by sense of smell, color, nature, texture, taste, and touch. A panel of twenty untrained judges evaluated and assessed the products on a 9-point hedonic rating scale in this research. It was done on a scale of likes and dislikes; starting with 'dislike extremely' (1) and 'like extremely' (9). Five denotes the neutral choice (neither like nor dislike). The panelists were asked to rate the nutrition bars on their appearance, color, flavor, taste, texture, and overall acceptability.

Microbial analysis

The total mesophilic count, as well as the yeast and mold count, was examined bimonthly. Total fecal coliforms and *Bacillus cereus* were also tested for safety for the first time, as stated in the AOAC [20].

Statistical analysis

Statistical analyses were conducted using SPSS software version 16. Tukey's adjusted p-value was used to determine significance ($p < 0.05$) after applying ANOVA to check the significant differences on different parameters.

Result and discussion

Proximate analysis

Since the nutritious profile of food products reflects the quality and proportion of components used, the relationship

between the proportion of integrated cowpea and the nutritional value of the bar was evaluated (Table 1). The proximate analysis of the cowpea nutrition bars revealed that the nutrition bar with 30% cowpea (CNB30) had less fat and more protein and carbohydrate than others. It is important to highlight the fact that the findings of the analysis prove that the cowpea nutrition bars are high protein bars with a protein content of 15.39–17.20% [22–24]. Calorie values in cowpea nutrition bars showed that the gaining energy from carbohydrates and protein increased since in samples containing cowpea fat was reduced and protein as well as carbohydrates were increased. However, in protein bars, calorie from protein and fat is preferred [22, 25]. Crude fiber in all samples was between 8.43% and 9.08%. The mineral composition of the cowpea nutrition bars was significantly different among the samples ($p < 0.05$) (Table 1). The CNB30 is rich in zinc and phosphorous. CNB0 has a higher concentration of iron and calcium.

The moisture content in cowpea nutrition bars revealed that, like most of other snack bars, they are in the category of intermediate moisture foods (10–40% moisture) [26]. During 6 months of storage, all samples exhibited a decrease in moisture content from (15.65–15.84%) to (15.30–15.35%) and no significant difference was observed at ambient

temperature ($p < 0.05$) (Table 2). This phenomenon has also been reported by Silva et al. for cassava flour-based food bars [27].

Texture hardness

The hardness is the most key parameter of textural characteristics of nutrition bars which is directly related to consumer acceptance. The hardness of samples during storage is shown in Fig. 1. The instrumental results were consistent with the organoleptic evaluation of the bars ($r = -0.978$). As mentioned earlier, the samples harden as time passes by. The hardness of CNB0 and CNB30 increased from 28.90 and 32.27 N on the first day to 70.09 and 79.45 N, respectively at the end of 180 days of storage at ambient temperature. The differences in hardness levels between samples during storage were significant ($p < 0.05$). This demonstrates that a change in formulation has a significant effect on the product's storage stability in terms of hardness. The water–protein interaction modifies the textural properties of the samples. Samuel et al. reported that moisture migrates to protein sources causing protein aggregation and sugar crystallization, which may enhance the hardness of the bars [24]. Previous researches have attempted to determine the influence that different protein sources have on the texture of nutrition bars [22, 26, 28]. This enhancement in hardness may also be attributed to the decrease in moisture content with an increase in storage time [27]. Hardening of protein buffer solutions (40% moisture) during storage at 34 °C and an increase in bound water from 64 to 68% of the total water has been reported. Moreover, there are some synergistic effects on preventing bar hardness between different proteins but the mechanism for this is unclear [29]. Similar trends were reported for increasing the hardness value of high protein bar and choco-quinoa bar, respectively during ambient storage [28–30].

Sensory evaluation

The changes in sensory properties of samples stored at ambient temperature are shown in Fig. 2. The organoleptic

Table 1 Proximate and mineral analysis of cowpea nutrition bars

Nutritional composition	Sample's code			
	CNB0	CNB10	CNB20	CNB30
Fat (g/100 g)	21.93	19.87	17.80	15.73
Carbohydrate (g/100 g)	48.18	49.36	50.55	51.73
Protein (g/100 g)	14.49	15.39	16.30	17.20
Crude fiber (g/100 g)	8.43	8.65	8.87	9.08
Energy (kcal/100 g)	417.81	409.63	401.45	393.27
Sodium (mg/kg)	247.37	222.63	197.89	173.15
Potassium (mg/kg)	2693.50	2702.15	2710.80	2719.45
Iron (mg/kg)	131.13	120.52	109.90	99.29
Calcium (mg/kg)	718.17	646.35	574.54	502.72
Zinc (mg/kg)	0.11	1.39	2.68	3.97
Phosphorus (mg/kg)	133.93	276.53	419.14	561.75

Table 2 Moisture content (g/100 g) of nutrition bar formulations prepared from different percentages of cowpea during storage

Sample's code	Storage time (days)						
	0	30	60	90	120	150	180
CNB0	15.79 ± 0.04 ^A	15.68 ± 0.07 ^{AB}	15.60 ± 0.03 ^{ABC}	15.54 ± 0.07 ^{ABCD}	15.46 ± 0.12 ^{CD}	15.40 ± 0.08 ^{CD}	15.33 ± 0.07 ^D
CNB10	15.84 ± 0.03 ^A	15.76 ± 0.05 ^A	15.68 ± 0.04 ^{AB}	15.59 ± 0.09 ^{AB}	15.50 ± 0.08 ^{AB}	15.43 ± 0.10 ^{ABC}	15.35 ± 0.08 ^C
CNB20	15.65 ± 0.06 ^A	15.57 ± 0.05 ^A	15.51 ± 0.08 ^{AB}	15.45 ± 0.11 ^{AB}	15.39 ± 0.09 ^{ABC}	15.35 ± 0.09 ^{ABC}	15.30 ± 0.04 ^C
CNB30	15.74 ± 0.08 ^A	15.66 ± 0.09 ^{AB}	15.57 ± 0.09 ^{AB}	15.50 ± 0.07 ^{AB}	15.41 ± 0.13 ^{ABC}	15.35 ± 0.12 ^{ABC}	15.30 ± 0.04 ^C

Data are expressed as mean ± standard deviation. The significance of the values in the columns with different lowercase letters and in the rows with different superscripts uppercase letters are indicated ($p \leq 0.05$)

Fig. 1 Hardness of nutrition bar formulations prepared by different content of cowpea

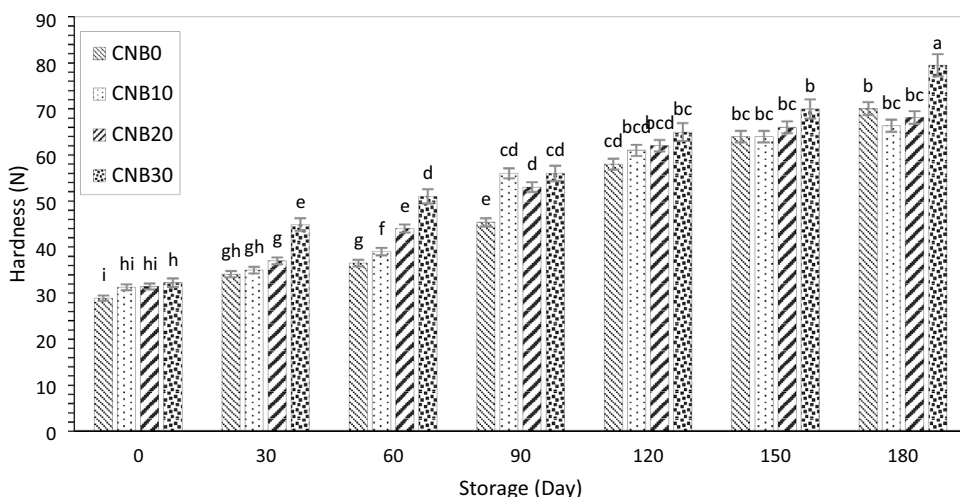
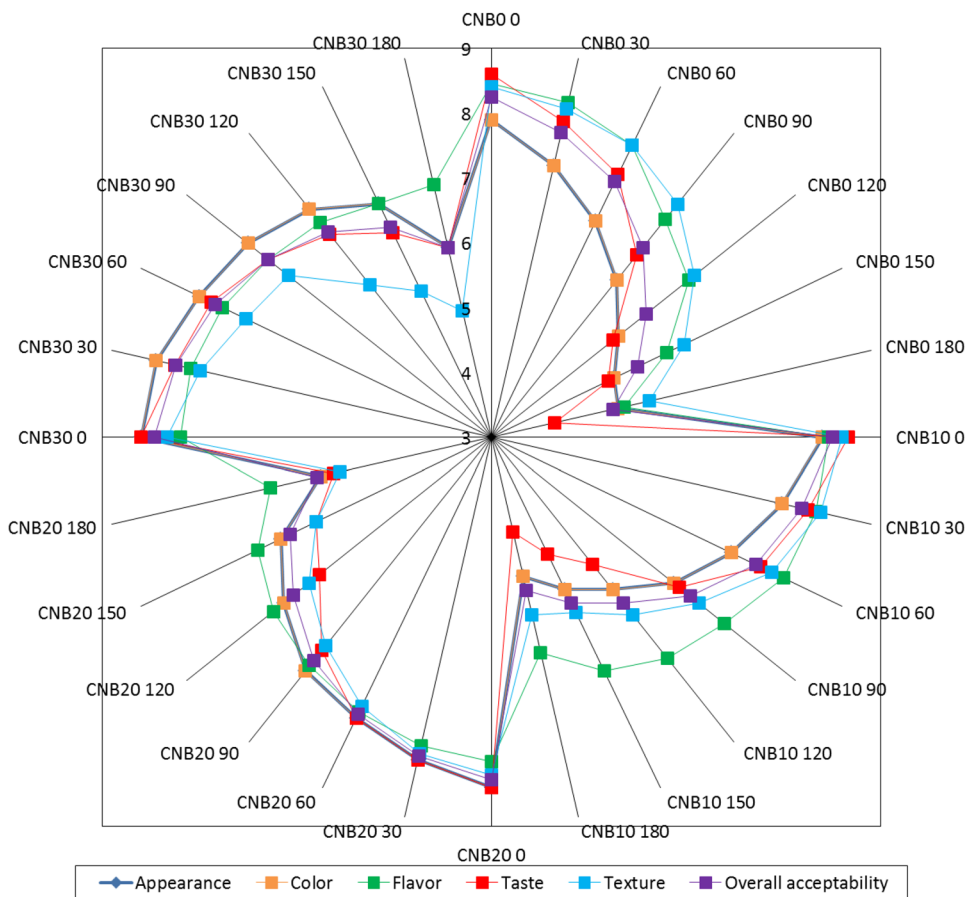


Fig. 2 Sensory analysis of nutrition bar formulations prepared from different percentages of cowpea



analysis of the samples to determine customer preference revealed that the cowpea nutrition bars were highly approved by the panelists for all sensory characteristics examined (Fig. 2). CNB0 and CNB30 flavor scores reduced from 8.45 and 7.81 on the first day to 5.10 and 7.03 at the end of 180 days of storage at room temperature. The changes in sample flavor scores were significant. This demonstrates that

a modification in the formulation has a substantial influence on the product’s storage stability in terms of flavor attributes. In the beginning, samples with a lower proportion of cowpea flour received higher flavor and taste scores (up to 90 days of storage). However, this trend was reversed and in the last days of storage, samples with more cowpea content had higher flavor and taste scores. This could be related to the

oil content of the nutrition bar. In this way, in the early days, a sample with higher oil content make consumers feel more palatable; but with increased storage time and the likelihood of lipid oxidation reactions, this trend changes and samples with lower fat content provide better flavor and taste. The protein in nutrition bars also tends to produce off-flavors and after-taste during prolonged storage [22].

The texture scores of CNB0 and CNB30 decreased from 8.41 and 8.03 on the first day to 5.50 and 4.99, respectively at the end of 180 days of storage. The changes in the body and texture scores were significant during storage. The body and texture scores of all samples decreased below 6.0 after 180 days of storage. Samuel and Peerkhan had also suggested that nutrition bars harden during storage resulting in lower consumer acceptance [24].

The color and appearance scores of the CNB0 and CNB30 increased from 7.90 to 8.42 at the first day. The CNB0 and CNB30 scores decreased to 5.02 and 6.03, respectively at the end of 150 days of storage. The changes in the scores of color and appearance among the samples were significant. This different trend of increased scores of the color and appearance in samples with higher cowpea content in all days of storage is probably due to the light color that increased with increasing the ratio of cowpeas.

The overall acceptability scores of all samples at the first day were similar to 8.25. The changes in the scores of overall acceptability among the samples were significant. The rate of decrease in overall acceptance scores of the samples declined with increasing cowpea content. Therefore, after 150 days of storage, the overall acceptance score of CNB0 and CNB30 reached 4.92 and 6, respectively. Based on sensory evaluation it was concluded that CNB20 and CNB30 samples were acceptable up to 180 days of storage. The results also showed that the scores of all studied traits for these sample up to day 90 were above 7. Researchers observed that all sensorial scores of omega-3 rich energy bars, energy bar utilizing potato extrudates, and Cereal Based Energy Bar were above 7 after 90 days at ambient storage [31–33]. Padmashree et al. also found that the initial score for sensory characteristics of a choco-quinoa Nutri-bar within metalized polyester packaging was over 8, but dropped to approximately 7 after 9 months of storage at 37 °C [30]. In contrast, Sobana reported that the overall acceptance score of the composite sport bar was decreased from 8.0 to 1.4 after 90 days at 37 °C [34].

Microbial quality

According to the previous results, the CNB30 bar was more acceptable. Therefore, it was initially tested for *coliforms* (total and fecal) and *Bacillus Cereus*, but neither were observed. CNB30 bar was screened for aerobic mesophilic count (CFU/g) and yeast and mold count (CFU/g)

Table 3 Microbial count detected for CNB30 bar stored for a period of 180 days

Day	Aerobic mesophilic count (CFU/g)	Yeast and mold count (CFU/g)
0	4500.00 ± 243.22 ^a	4.10 ± 0.36 ^c
60	780.00 ± 10.75 ^b	45.00 ± 3.25 ^a
120	145.00 ± 5.68 ^c	13.00 ± 2.12 ^b
180	98.00 ± 1.23 ^c	6.00 ± 0.45 ^c

Data are expressed as mean ± standard deviation. Values in the same columns with different superscripts are significant ($p \leq 0.05$)

every 2 months for a period of 180 days, and the results are reported in Table 3. The aerobic mesophilic count was initially somewhat greater, but dropped steadily with storage time ($p < 0.05$). This decline might be due to a decrease in accessible oxygen in the packing material. Yeast and mold count increased considerably ($p < 0.05$) over the first 60 days of storage but subsequently decreased significantly ($p < 0.05$) with additional storage duration. The changes in microbial count might be attributed to differences in water activity during storage; nonetheless, the product remained stable and safe for consumption until the end of the 180-day period since the overall bacterial count was within the standard acceptable range (1000,000 CFU) [35]. A rise in total bacterial count as well as yeast and mold count (log cfu/g) from 3.4 to 4.6 and 3.45 to 3.62 was detected, respectively, following 90 days of storage in an energy bar containing flax seeds [31].

Conclusions

Cooked and oven-dried cowpea in different four concentrations of 0%, 10%, 20%, and 30% was used to formulate the nutrition bars (CNB0, CNB10, CNB20, and CNB30 respectively) and measure change in nutrition value, moisture content, texture, organoleptic properties and microbial count. Bar CNB30 was generally more nutritious than other treatments with less energy value. Moisture content decreased in bars during storage which affected their hardness. Nevertheless, reaction between water and protein reduced hardness of the samples. Cowpea protein led to higher organoleptic scores and CNB30 got the highest score in overall acceptability. Microbial count of sample CNB30 was within the standard acceptable range showing the product remained stable and safe for consumption until the end of 180 day. Increased cowpea protein use in nutrition bar applications stands to benefit the confectionery industry through increased demand for complete protein bars. However, other researches need to be conducted on other protein sources and concentration to fortify nutrition bars with proteins.

Data availability The authors confirm that the data supporting the findings of this study are available within the article.

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