

# BEET ROOT AS POTENTIAL INGREDIENT FOR FUNCTIONAL FOODS DEVELOPMENT

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## 1. ABSTRACT

The objective of this work was to develop cereal bars with beetroot (*Beta vulgaris* L var. *conditiva*) as a functional ingredient. Beet bars were obtained mixing dried ingredients (whole oat, corn crips and grated beet) and sugar matrix (glucose and honey), then the mix was spread in an aluminum plate, moulded and baked. Control bars (without beet) were obtained with the same procedure. Water activity, moisture content, color and texture of the formulated snack bars were analyzed. Active compounds of beetroot and bars samples, betalains and polyphenols, were also determined. Beet bars showed lower values of water activity (0.35) than those obtained for the control bars (0.41), indicating good microbiological stability of the formulated product. Moisture content values of 9.2 % for beet bars were obtained, similar to those found in control samples (7.7 %). Beet bars showed lower L and higher a\* values compared to control bars, due to the redness of beetroot. Similar b\* values were obtained for both, control and beet bars, denoting yellowness in the color of samples. Texture profile analysis and a three-point bending test were applied to the snack bars. Fracture stress, elastic modulus and fracture energy values were higher for the beet bars compared to the control, showing that beet incorporation modified the texture of the bars. With regard to the active compounds, high contents of betalains (> 95 %) were retained in the beet bars after baking. Beetroot addition increased the polyphenol content of the final product compared to the control, improving its nutritional profile. Beet bars had a good acceptance level

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(68 %) when were evaluated by an unentrained sensorial panel, showing that this product could be consumed as a healthy snack.

## 2. INTRODUCTION

Consumer's trend for natural and healthy foods demands for new products or existing ones with improved nutritional characteristics. Snack products, like cereal bars, are popular foods that could be modified in their composition to enhance their nutritional profile. An alternative could be the incorporation of vegetables in the formulation due to their health-promoting properties. The presence of vitamins, minerals, dietary fiber and polyphenols in the vegetables composition contributes to a healthy diet and may reduce the risk of chronic diseases.

Bar formulations often includes fruits in several formats (pieces, puree or powder) and their color and taste are preferred by consumers, especially children (Orrego *et al.*, 2014). However, consumption of vegetables is not as popular as fruits especially in snack products. In the present work, beetroot (*Beta vulgaris L var. conditiva*) is proposed as a functional ingredient for cereal bars due to its nutritional properties and attractive color.

Beetroot is a rich source of polyphenolic compounds and contains pigments called betalains, which are composed of red-violet betacyanins and yellow betaxanthins (Kujala *et al.*, 2002). The antioxidant properties of these compounds could be altered during beetroot processing (Ravichandran *et al.*, 2013), leading to loses in color and nutritional components. The objective of this work was to develop cereal bars with beetroot, to characterize their physico-chemical and texture properties and to evaluate their sensorial characteristics.

## 3. MATERIALS AND METHODS

Fresh beetroots were purchased in a local market and then were washed, peeled and grated. Samples were then vacuum dried in an oven (Arcano, China) connected to a vacuum pump PC 511 NT (Vacuubrand, Germany). Operation conditions for drying were 70 °C and 50 mBar (AOAC 934.01 method, AOAC, 2000).

Beet bars were obtained mixing cereals (whole oat, corn crips, 50:50), dried beet (4 % w/w) and sugar matrix (glucose:honey 70:30 w/w, respectively, previously

heated at 85-90 °C). The mix was homogenized during 5-10 min, then it was spread in an aluminum plate and oven at 130 °C for 30 min in a convection oven (Hitachi HTO-23, Argentina). The batch was allowed to cool at room temperature and cut with a stainless matrix to obtain the cereal bars (3cm ×10cm × 1cm). Control bars (without beet) were obtained with the same procedure. All bars were stored at room temperature in hermetic plastic bags until analysis.

#### **WATER ACTIVITY AND MOISTURE CONTENT**

The water activity ( $a_w$ ) of the bars was measured in triplicate in an Aqualab equipment (Series 3TE, USA) according to AOAC 978.18 method (AOAC, 2000). Bars samples ( $2 \pm 0,1$  g) were put in aluminum trays and dried in a convection oven (H701P, San Jor, Argentina) at 105 °C ( $\pm 1$  °C) until constant weight (AOAC 984.25 method, AOAC, 2000). Measurements were made by triplicate.

#### **SUPERFICIAL COLOR MEASUREMENT**

Superficial color of bar samples was determined in a tristimuli colorimeter (Chroma meter CR-300, Minolta, Japan). Hunter  $L^*a^*b^*$  (L: luminosity;  $a^*$ : red-green and  $b^*$ : yellow-blue), Chroma and Hue from CIE scale were acquired by triplicate.

#### **ACTIVE COMPOUNDS DETERMINATION**

The pigments of beetroot and bars samples were extracted with water and water:ethanol (50:50), at 25 °C during 30 min with agitation. *Betalains* (*betacyanins* and *betaxanthins*) content was determined in the aqueous extract using a spectrophotometer (Shimadzu, UV-mini 1240, Japan) at 538 nm and 480 nm, according to Ravichandran *et al.* (2013). The absorbance reading obtained was used to calculate the betalain concentration for each sample. The betalain content (BC) was calculated as  $BC (mg/L) = [(A \times DF \times MW \times 1000) / (e \times l)]$ , where  $A$  is the absorption,  $DF$  the dilution factor and  $l$  the pathlength (1 cm) of the cuvette. For quantification of *betacyanins* and *betaxanthins*, the molecular weights ( $MW$ ) and molar extinction coefficients ( $e$ ) ( $MW=550$  g/mol;  $e=60,000$  L/mol cm in  $H_2O$ ) and ( $MW=308$  g/mol;  $e=48,000$  L/mol cm in  $H_2O$ ) were applied.

Total polyphenols content (TPC) in the ethanolic extract was determined by the Folin–Ciocalteu method (Singleton *et al.*, 1999). Briefly, 50 mL of the sample were mixed with 2.3 mL of distilled water and 50 mL of Folin–Ciocalteu reagent

(Anedra, Argentina, 1:1 diluted). Then 100 mL of Na<sub>2</sub>CO<sub>3</sub> (20 g/100 mL) (Ane-dra, Argentina) was added. After 30 min, sample absorbance was measured at 725 nm in the spectrophotometer. Gallic acid (Sigma-Aldrich, USA) was used as stan-dard. Results were expressed as mg gallic acid equivalents (GAE)/g bar.

#### **TEXTURE ANALYSIS**

Texture profile analysis (TPA) and a three-point bending test were applied to the snack bars. Both determinations, in at least five replications, were performed in a Texture Analyser T2i TA.XT2i-Stable Micro Systems (United Kingdom). Data were registered through Texture Expert Exceed software.

TPA was determined using a HDP/VB probe to penetrate the sample to a depth of 5 mm in the first bite simulation and 8 mm in the second one, at a constant speed of 5.0 mm/s.

Three-point bending test was performed using a HDP/3PB probe. The sample was held on two stationary bending supports 40 mm apart, while being displaced at a central axis by the bending probe attached to the moving crosshead. Force was applied to the center of the bar until fracture occurred. Texture parameters were de-termined from the force-deformation curve and calculated according ASTM Stan-dard D790-90 (Kim *et al.*, 2009).

#### **SENSORY ANALYSIS**

A consumer acceptability test of the beet bars was performed at Facultad de Inge-niería (Universidad Nacional de La Plata, Argentina). Fifty students and teachers (29 female and 21 male), aged between 18 and 49, were recruited and participated. Samples were presented in a plastic tray and labeled with a 3-digit random number. Purified water was available to rinse the mouth between samples. Sensory attribu-tes (appearance, color, taste, texture, overall liking) were evaluated in a 9-point he-donic scale, where 9=like extremely and 1=dislike extremely. At the end of testing, panelists were also asked about purchase interest of the bars.

#### **STATISTICAL ANALYSIS**

Data analysis was performed using the software SYSTAT INC. (Evanston, USA). Analysis of variance (ANOVA) and mean comparisons were carried out. Unless indicated, a level of 95 % of confidence ( $\alpha=0.05$ ) was used.

#### 4. RESULTS AND DISCUSSION

Figure 1 shows the aspect of the formulated cereal bars with and without beetroot. It can be seen that the pieces of beet are homogeneously distributed in the whole bar. The amount of dried beet used in the formulation, 4 %, was enough to show the color of this ingredient in the final product. The beet percentage could be increased to make the bar more attractive in color; however, sensorial properties should be taken into account due to the characteristic taste of beetroot.

Preliminary studies using fresh beet gave bars with high values of  $a_w$  and water content, thus, dried beet was selected for the formulation. Vacuum drying was applied to the beetroots since this method has shown to preserve natural color and nutrients (Dueik & Bouchon, 2013).

Lower values of water activity were obtained for the beet bars compared to those for the control ones (Table 1), indicating good microbiological stability of the formulated product. The incorporation of dried beetroot slightly increased the moisture content of the bars, respect to the control samples. The values of both parameters,  $a_w$  and moisture content, were agreed with those found in commercial cereal bars (Fuertes and Navarro, 2011).

An important attribute of snack bars to consumers is the surface color (Figure 1 and Table 1). The formulated beet bars can be described by their lightness (L), redness ( $a^*$ ) and yellowness ( $b^*$ ). Bars with beetroot in their formulation showed lower L and higher  $a^*$  values compared to control bars, due to the redness of beetroot. Similar  $b^*$  values were obtained for both, control and beet bars, denoting yellowness in the color of samples (Table 1). The use of the reducing sugars in the formulation and the elevated baking temperature could explain the light brown color detected in the bars, as a result of the non-enzymatic Maillard browning reaction and the caramelization process (Sun-Waterhouse *et al.* 2010).

#### ANTIOXIDANT COMPOUNDS IN BEET BARS

With regard to the total polyphenols content, beetroot addition improved the nutritional profile of the bars since higher amounts of polyphenols were found in beet bars compared to the control (Figure 2A). The antioxidant activity of beetroot is partly due to their phenolic acids content; however, different processing methods like vacuum or roasting modify the phenolic acid concentration (Ravichandran *et al.*, 2012).

Betalain stability is influenced by different internal factors such as pigment content, pH, moisture content and external factors such as temperature, light and oxygen, among others (Wootton-Beard and Ryan, 2011). Bars processing could have changed the content of betalains and, consequently, the food color as well as the antioxidant activity. However, in this work, high contents of betalains (> 95 %) were retained in the beet bars after baking. A relation approximately 1:1 was found for both pigments betacyanins and betaxanthins (Figure 2B). Therefore, the formulated beet bars constitute a rich source of antioxidants due to the contribution of polyphenols and betalains.

### **TEXTURE ANALYSIS**

The three-point bending test was used to determine the fracture properties of the cereal bars (Table 2). It is considered the most reliable method since it is the least dependent on particular equipment design, geometry and sample dimensions (Kim *et al.*, 2009). Table 2 shows that fracture stress, elastic modulus and fracture energy values were higher for the beet bars compared to the control, indicating that beet incorporation modified the texture of the bars.

The fracture stress is proportional to the maximum breaking force which can be related to the crispness, an attribute which denotes freshness and high quality of snack cereal products. Higher values of fracture stress were found for the formulated bars, showing that crispness increased after the incorporation of beetroot in the cereal bars.

### **SENSORIAL ANALYSIS**

Students of university level were selected as the test group for the sensorial analysis of the beet bars. All of them claimed to be snack consumers, especially cereal bars. An overall liking of 68 % was found in the test, showing a good acceptance level of the beet bars.

Panelists were also asked about the purchase interest and 58 % of the students answered that they would probably buy the beet bars if these products were in the market. Moreover, 15 % claimed that they definitely would buy the formulated product (Figure 3).

Both process and ingredient variables influence the physical properties and sensory attributes of final food products. In this case, beetroot demonstrated to be a

suitable ingredient which did not alter the overall appearance and taste searched for the bars consumers.

## 5. CONCLUSIONS

Beet bars could be a promising healthy snack since, as shown in this work, they had high antioxidant properties and good sensory attributes. The incorporation of vacuum dried beet allowed obtaining microbiologically stable products, due to their low values of  $a_w$  and moisture content.

Beetroot addition improved the texture and nutritional values of cereal bars. Increased phenolic content and significantly high amounts of betalains were found in the formulated products. Beet bars had a good acceptance level among students showing that this product could be consumed as a healthy snack.

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## 7. TABLES AND FIGURES

### FIGURES

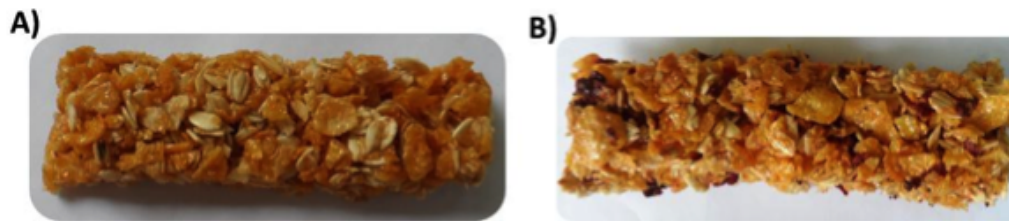


FIGURE 1. Images of formulated snack bars: (A) Control, (B) Beet bar.

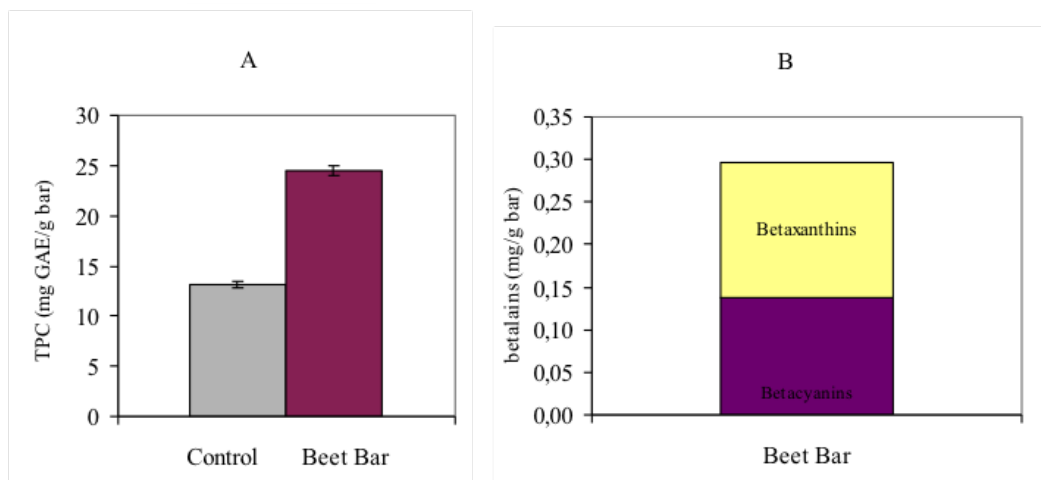


FIGURE 2. A) Total polyphenols content for control and beet bars, B) Total betalains content of beet bars, showing the contribution of pigments.

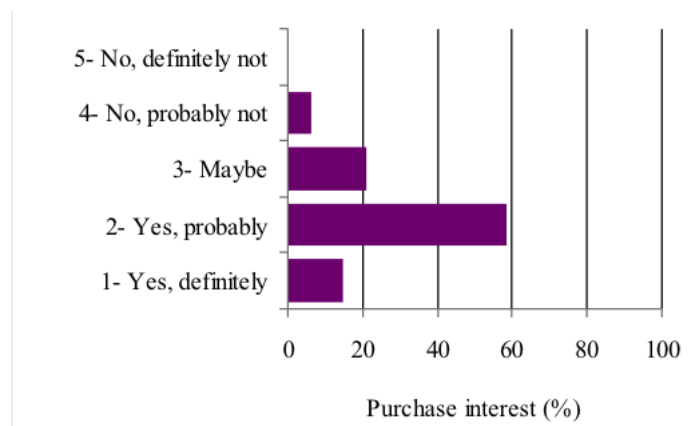


FIGURE 3. Purchase interest of beet bars as a result of the sensorial evaluation by an untrained panel.

## TABLES

**TABLE 1.** Physicochemical parameters of snack bars

Snack bar	a <sub>w</sub>	Moisture content (%)	Hunter Lab values		
			L	a*	b*
Control	0.41	7.7 ± 0.3	47.35 ± 4.37	3.29 ± 1.35	34.61 ± 2.87
Beet bar	0.35	9.2 ± 0.2	42.89 ± 5.87	7.23 ± 0.84	28.26 ± 4.78

*Note:* values are expressed as mean ± standard deviation.

**TABLE 2.** Mean values of instrumental parameters determined by the three-point bending test

Snack bar	Fracture Stress (KPa)	Fracture Strain (-)	Elastic Modulus (KPa)	Fracture Energy (N.mm)
Control	273.25 ± 8.23	0.18 ± 0.03	1.36 ± 0.25	47.74 ± 15.64
Beet bar	541.46 ± 0.37	0.23 ± 0.01	2.34 ± 0.08	90.21 ± 9.74

*Note:* values are expressed as mean ± standard deviation.