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Cladodes of *Opuntia ficus indica* as a functional ingredient in the production of cookies: physical, antioxidant and sensory properties

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Opuntia ficus indica cladodes are a rich source of bioactive compounds and dietary fiber; therefore, they could be a functional ingredient in the production of health-promoting foods. This study aimed to develop cookies by substituting wheat flour with powdered cladodes in different proportions, 15, 20 and 25%, and analyze them in terms of physical and bioactive compounds and sensory characteristics. Proximal analysis showed that powdered cladodes were high in total dietary fiber (54.89%). The analysis of total phenolic compounds and antioxidant capacity revealed values of 2050.20 mg GAE per 100 g db and 312.14 mg Trolox per 100 g db. Powdered cladode supplementation increased hardness and decreased color parameters (L^*, a^* and b^*) compared to control (without cladode powder). Moreover, rising levels of powdered cladodes contribute to the increased total phenolic compound content and antioxidant capacity compared to control. Sensory analysis showed that 15% cladode supplementation was adequate for preparing an acceptable functional cookie. The sorption isotherm showed that it was possible to fit the GAB model to the experimental data and the cookies were stable at 25 °C. Cladodes of *Opuntia ficus indica* could be considered as a functional ingredient and a source of dietary fiber and antioxidants for the manufacture of foods with benefits to human health and nutrition.

Sustainability spotlight

Our research group harnessed *Opuntia ficus indica* cladodes for sustainable and health-conscious food choices, introducing a new dimension to functional ingredients. In this study, powdered cladodes replaced traditional wheat flour in cookie formulations, revealing substantial total dietary fiber content and serving as a potent source of bioactive compounds. The cookies exhibited promising stability at 25 °C, crucial for sustainability. *Opuntia ficus indica* cladodes offer a sustainable solution, enabling the food industry to contribute to health and environmental well-being. This research signifies a step towards integrating sustainable practices in food manufacturing for a healthier, environmentally conscious future.

1 Introduction

Products made with wheat flour, such as bread, cakes and cookies, are globally consumed in significant quantities; in fact, they are one of the main components of the human diet, owing to their low cost and easily made formulations, and the world total wheat output in 2023 was nearly 777 million tonnes.[1] The production and consumption of cookies have increased worldwide, and the market reached USD 121 billion by 2021 and is expected to reach USD 164 billion by 2024 (ref. 2). However, the main ingredients in wheat flour-based products may lack dietary fiber, minerals, and antioxidants, which are relevant to human nutrition and health; besides, the excessive consumption of ultra-processed foods contributes to unhealthy diets.[3] Conditions such as cardiovascular diseases and obesity limit fat intake in particular fats rich in saturated fatty acids, such as butter, and the use of unsaturated fatty acids should be recommended to reduce the risk of heart diseases.[4] According to the Institute of Food Technologists, healthy foods will continue to be a trend in the food market for future years.[5] In addition, a healthy diet rich in natural ingredients could act as functional foods and/or nutraceuticals. These types of ingredients have the potential to improve health and could help in the primary prevention of many chronic diseases, such as dyslipidemia and cardiovascular diseases. Therefore, consumers have become interested in food processing, food ingredients and how they interact with health;[5] also, food markets are interested in local, organic foods or products which incorporate these ingredients.

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On the other hand, epidemiological studies[6,7] have shown that regular consumption of plant-based foods is associated with reduced risk of chronic diseases related to oxidative stress. Therefore, functional ingredients rich in antioxidants, dietary fibers, minerals, and vitamins, low in calories and fat, and free of synthetic additives used in the formulation of conventional foods can enhance their nutraceutical potential. In this context, the composition of *Opuntia ficus indica* cladodes makes them a potential ingredient to produce functional foods that promote health. In Mexico, in addition to being consumed as a vegetable, *Opuntia ficus indica* cladodes are used in traditional medicine to treat diabetes, hyperlipidemia, obesity, and gastrointestinal disorders.[8,9] Cladodes contain high amounts of fibers, including mucilage, pectin, lignin, cellulose, and hemicellulose, which influence the metabolism of lipids and carbohydrates.[10–12] However, fresh cladodes have limited acceptance among many consumers including kids, due to their mucilaginous texture, acidity, astringency, and herbal taste.[13] Consequently, an alternative to increase their use and consumption is to transform them into powder and use them as a functional ingredient that maintains the nutritional, medicinal, and nutraceutical properties. Therefore, the incorporation of these powders in the formulation of flour-based products could represent a promising strategy, acting as a good carrier for functional ingredients and offering the possibility of introducing substances with health benefits through the diet to increase their nutritional profile and confer additional functional properties.[7,14–16]

In this context, the aim of the present study was to evaluate the effect of replacing wheat flour with powdered cladodes of *Opuntia ficus indica* var. Atlixco and replacing butter with vegetable oil in the preparation of cookies and how this substitution influences their physical properties, polyphenol content, antioxidant capacity, and sensory properties. Likewise, for the best evaluated sensorial cookie, the sorption isotherm was used as an instrument to predict the shelf life and acceptability of the products.

2 Materials and methods

2.1. Preparation of cladode powder

Opuntia ficus indica variety Atlixco cladodes with more than 60 days of maturation and a length greater than 30 cm were collected in May 2023 in Atlixco, Puebla, Mexico (18° 54′ 4.935″ N 98° 27′ 11.346″ W), and were free of pesticides and chemical fertilizers. Manual depinning was performed with a stainless-steel knife, and they were washed with distilled water and disinfected with a solution of 10% sodium hypochlorite. Then, cladodes were sliced into strips of 2 cm lengthwise, dried in a dehydrator at 45 °C/30 h (Excalibur® EXC10EL, Sacramento, California, USA), and ground at 22 rpm/4 min (WARING, blender 7011 HS, USA). The powder obtained was stored at 25 °C hermetically in sealed bags until further use.

2.2. Proximal chemical analysis

Cladode powder analysis was carried out according to the methods of A. O. A. C. (2000).[17] The moisture content (950.46) and ash content (31.012) were determined from the weight difference after dehydration and calcination, respectively. The crude protein content was determined using the micro-Kjeldahl method (2001.11), and the total nitrogen content was used for protein content calculation using a factor of 6.25. The crude fat content (920.3) was determined by solvent extraction using a Fat Extractor E-500 (BUCHI, Fat Extractor E-500, Flawil, Switzerland). Total dietary fiber, insoluble dietary fiber, and soluble dietary fiber were determined using an enzymatic-gravimetric method 991.43 (Merck enzymes, 112979, Germany) and the procedure of Prosky *et al.*[18] The carbohydrate content was determined from the difference in the content of the other components.

2.3. Physical properties

2.3.1. Bulk density measurement. The assay was carried out according to the technique described by Chandra *et al.*[19] with slight modifications. Briefly, 10 g of sample was placed in a 25 mL graduated test tube, it was hit on a wooden table several times until a visible decrease in the level of the graduated test tube was no longer noticed, and depending on the weight and volume, the bulk density (g mL^{-1}) was calculated.

2.3.2. Water and oil holding capacity (WHC and OHC). Water and oil holding capabilities were determined using the procedure described by Chandra *et al.*[19] with minor modifications. 1 g of sample was mixed with 50 mL of distilled water or 50 mL of soybean oil, stirred for 1 min at high speed (Vortex-Gene 2 G560, Scientific Industries, USA), kept at rest at room temperature (25 °C) for 30 min, then centrifuged at 2000 × g for 30 min, and the volume of water or oil in the supernatant was measured. Water and oil holding capabilities were expressed as a percentage of bound water or oil per gram of powdered cladodes.

2.4. Bioactive properties

2.4.1. Extract obtention. The extract was obtained with the Santiago *et al.*[20] methodology with slight modifications. A mixture of 2 g of sample and 25 mL of methanol/water was added (50 : 50) and placed in an ultrasound bath (Branson, M1800H-E, USA) for 30 min. Then, the mixture was centrifuged at 2200 × g for 10 min (Centrifuge Z 366 K, HERMLE Labortechnik, Germany). A second extraction was performed with 25 mL of acetone/water (70 : 30). Both supernatants were mixed and filtered through a Whatman No. 1 (Sigma-Aldrich) filter paper and concentrated in a rotary evaporator (R-300, Buchi, Flawil, Switzerland) at 51 °C and 235 mm Hg until half of the initial volume was evaporated.

2.4.2. Determination of total phenolic compounds. Total phenolics quantification was made through the Folin–Ciocalteu colorimetric method.[21,22] A solution of the 0.1 M Folin–Ciocalteu reagent (Sigma-Aldrich) was mixed with 100 μL of extract. The mixture was incubated for 6 min, then 1 mL of Na_2CO_3 at 5% was added and incubated in the dark at room temperature (25 °C) for 30 min. Solutions were measured with a spectrophotometer at 765 nm (UV-Vis Spectrophotometer 1900i, Shimadzu, Japan). A calibration curve was prepared with gallic acid (Sigma-Aldrich, Mexico) at different concentrations: $y = 0.0042x$(0–200 ppm) + 0.0592, ($R^2 = 0.991$). The results were expressed as mg gallic acid equivalents (GAEs) per 100 g dry weight and calculated using eqn (1).

$$\text{GAE}\ (\text{mg mL}^{-1}) = \left(\frac{\text{Abs} - b}{m}\right) \times \text{DF} \tag{1}$$

where Abs is the absorbance of each solution (sample/standard) and DF is the dilution factor.

2.4.3. *In vitro* antioxidant capacity by DPPH (2,2-diphenyl-1-picrylhydrazyl) assay. Antioxidant capacity was determined by DPPH assay developed by Brand-Williams *et al.*[23] with some modifications. A mixture of 200 μL of extract was added to 2 mL of DPPH ethanolic solution (0.1 mM). The resulting mixture was incubated for 30 minutes at room temperature (25 °C). Absorption was read at 517 nm on a UV-Vis spectrophotometer (UV-Vis Spectrophotometer 1900i, Shimadzu, Japan), and used to calculate the inhibition percentage (I%) of the antioxidant capacity (eqn (2)). A calibration curve was prepared using Trolox (6-hydroxy-2,5,7,8-tetramethylchroman-2-carboxylic acid; Sigma-Aldrich, Mexico) as the antioxidant at different concentrations. The concentration of Trolox was plotted against the percentage of inhibition to obtain the calibration curve ($I(\%) = 0.872 \times [0\text{–}300\ \text{ppm}] - 0.418$, $R^2 = 0.962$). The results were expressed as mg Trolox equivalents (TEs) per 100 g dry weight (eqn (3)).

$$I(\%) = \frac{A_{\text{DPPH}} - A_{\text{S}}}{A_{\text{DPPH}}} \times 100 \tag{2}$$

$$\text{Antioxidant capacity (mg TE per 100 g)} = \left(\frac{I(\%) - 0.419}{0.872}\right) \times \text{DF} \tag{3}$$

where A_{DPPH} is the absorbance of the blank without sample or standard, A_{S} is the absorbance of the sample after 30 min of reaction, and DF is the dilution factor.

2.5. Preparation of functional cookies

2.5.1. Cookie formulation. For the preparation of cookies, a full factorial design was used, and different proportions of canola oil/butter and cladode powder/wheat flour were used for cookie elaboration following the values shown in Table 1. The basic formulation consisted of canola oil/butter creamed with 33 g of brown sugar in a Kitchen aid blender (Whirlpool Corporation, USA). Subsequently, 18 g of eggs and 2 mL of vanilla were added. Finally, 100 g of a mixture of cladode powder and wheat flour was added until a homogeneous and smooth dough was obtained. The resulting dough was molded with a roller and the cookies were cut and cooked at 180 °C for 15 min in a convection oven (Zucchelli Forni, Italy). The cookies were allowed to cool (20 min) and were stored in airtight bags for further analysis.

2.5.2. Characteristics of cookies. After elaboration, cookies were evaluated in terms of physical properties, water activity, color (L^*, a^*, b^*), and hardness. In addition, biochemical compounds (total polyphenol content and antioxidant capacity by DPPH assay) were also determined. Finally, sensory evaluation (color, smell, texture, taste, and general acceptance) was performed.

Table 1 Experimental design

Formulation	% Cladode/wheat flour powder/wheat flour	% Vegetable oil/butter
Control	0/100	0/100
F1	15/85	70/30
F2	25/75	70/30
F3	25/75	50/50
F4	25/75	30/70
F5	15/85	30/70
F6	20/80	70/30
F7	20/80	30/70
F8	20/80	50/50
F9	15/85	50/50

2.5.2.1. Water activity. The determination of water activity (a_w) was performed with Aqualab 4TEV equipment, and a proper amount of cookie sample powder was placed on the tray for the determination.

2.5.2.2. Color. Determination was made on the surface of the cookies with the help of a colorimeter (Konica Minolta CR-410, Tokyo, Japan) in the transmittance mode with a D65 standard illuminant and a 2° standard observer. The parameters L^* (luminosity), a^* (trend towards green − or red +), and b^* (trend towards blue − or yellow +) were recorded.

2.5.2.3. Hardness. The texture of the cookies was measured with an EZ-SX texturometer (Shimadzu Corporation, Kyoto, Japan). The maximum force (hardness) for cookie break was measured with a cutting device in the middle of the cookie, and the upper blade was moved down at a speed of 1 mm s^{-1} with a penetration of 5 mm. The hardness value is expressed in newton (N).

2.5.2.4. Bioactive compounds of cookies. Total phenolic compounds and antioxidant capacity were determined following the methodology previously mentioned.

2.5.2.5. Sensory evaluation. The preferences or acceptance of cookies were evaluated with potential consumers. At the beginning of the test, each judge was informed about the test they were going to perform. Only judges who agreed with the evaluation were allowed to participate. Judges were also informed that "all products were safe for consumption" and that they could leave the test at any time without a reason. The evaluation panel consisted of 30 untrained men and women with ages between 22 and 59 years from the Universidad de las Americas Puebla (UDLAP). Evaluators tasted the cookies and rated the color, smell, texture, taste, and general acceptance, using a hedonic scale of 9-point where 1 = I dislike it very much and 9 = I like it very much. The samples were placed on white plates identified by random three-digit numbers. One sample was given at a time.

2.6. Moisture sorption isotherms

Moisture sorption isotherms were obtained for the control cookie and the cookie best ranged in sensory attribute of general acceptance. The evaluation of moisture sorption isotherms was carried out, as a quality parameter during storage at different humidity levels. The methodology of Jiménez, *et al.*[24] with slight modifications was used. Saturated solutions of salts used to control relative humidity were LiCl (11.3%), CH_3COOK (22.5%), K_2CO_3 (44.3%), $Mg(NO_3)_2$ (53.6%), NaBr (56.6%), NaCl (75.3%), $BaCl_2$ (90.3%), and K_2SO_4 (97%). Then, 0.5 g of cookie in triplicate was placed inside the hermetically sealed glass jars. The complete set of solutions was placed in an environment with a controlled temperature of 25 °C. The weight of the samples was recorded every three days for four weeks. The experimental data of moisture sorption were adjusted to the GAB (Guggenheim, Anderson, and De Boer) model. The moisture content was obtained by the AOAC 950.46 method.

2.7. Statistical analysis

All experiments were performed in triplicate except for texture analysis where five samples were used for the test. The results were expressed as mean ± standard deviation. The comparison of the means was analyzed by analysis of variance (ANOVA) followed by the Tukey test to identify significant differences ($P \leq 0.05$) between the samples. Data were analyzed using Minitab software (version 19, Minitab Inc., USA).

3 Results and discussion

3.1. Chemical composition of cladode powder

The results obtained for cladode powder composition are presented in Table 2. The protein content (11.5%) obtained was similar to those reported from other studies.[25,26] Differences in protein content values depend on the cultivar variety. Specifically, for Atlixco variety, the values range from 9.6%[25] to 12.6%.[26] On the other hand, the main component of cladode powder was total dietary fiber (TDF) (54.89%). Specifically, insoluble dietary fiber (IDF) (cellulose, hemicellulose, and lignin) was higher than soluble dietary fiber (SDF) (pectin, gums, and mucilage) which is typical of this type of product. The values correspond to those previously reported by other authors; TDF values in the range of 45 to 66%, IDF in the range of 32 to 56%, and SDF in the range of 5.6 to 25.22%.[27–29] However, the proportion of IDF to SDF changes with the maturation stage, IDF increases with maturation.[29,30] High values of TDF indicate that cladode powder could be considered as a high source of fiber and a key ingredient in food fortification. Different studies have demonstrated the beneficial effect of cladodes; Uebelhack *et al.*[31] showed that cladode fiber helps reduce body weight by binding to dietary lipids and increasing their excretion. Moran-Ramos *et al.*[9] tested the effect of powdered cladodes in diet of obese rats, and a decrease of total cholesterol serum levels was observed. Also, the authors suggest an effect on liver function by decreasing triglyceride accumulation and fatty acid peroxidation, thereby reducing hepatic oxidative stress. Padilla *et al.*[32] studied changes in cholesterol levels in mice and prevented hyperlipidemia. Finally, the ash content agrees with previous studies.[26,33] In addition, the authors also mention that the main minerals found in cladodes are potassium, calcium and magnesium; however, mineral content varied with the maturation stage.[34]

Table 2 Characterization of cladode powder from *Opuntia ficus indica* var. Atlixco

Parameters	Value[a]
Moisture (g per 100 g db)	8.52 ± 0.88
Proteins (g per 100 g db)	11.50 ± 0.56
Lipids (g per 100 g db)	1.83 ± 0.17
Carbohydrates[b]	62.77
Total dietary fiber (g per 100 g db)	54.89 ± 3.76
Insoluble dietary fiber (g per 100 g db)	35.25 ± 2.74
Soluble dietary fiber (g per 100 g db)	19.64 ± 1.49
Ash (g per 100 g db)	15.38 ± 0.10
Bulk density (g mL^{-1})	0.77 ± 0.01
Water holding capacity%	34.67 ± 1.15
Oil holding capacity%	8.0 ± 1.5
Total polyphenols (mg GAE per 100 g db)	2050.20 ± 71.5
Antioxidant capacity (DPPH; mg Trolox per 100 g db)	312.14 ± 55.64

[a] Values are the mean ± standard deviation. [b] Calculated from the equation: carbohydrates = 100 − (moisture + protein + lipids + ash), db: dry basis.

3.2. Physical properties

The physical characteristics of cladode powder are presented in Table 2. The bulk density was 0.77 g mL^{-1} (Table 2), which is consistent with the value reported by Ayadi *et al.*[16] of 0.647 g mL^{-1} for powdered cladodes. Water and oil retention capacities are important parameters that affect the texture, mouthfeel and consistency of food products.[6] Regarding the value of WHC (water holding capacity), powdered cladode showed around 34.67% which was higher than the OHC (oil holding capacity) by 8%; this is related to the high fiber content (54.89 ± 3.76 g per 100 g db), as well as the protein content (11.50 ± 2.74 g per 100 db), since its hydrophilic and hydrophobic nature allows it to interact with water and/or oil in food.[15] Cladode powder, due to its high water retention capacity and swelling properties, may be appropriate to improve the texture and stability of a variety of foods such as bakery products; likewise, hydrophobic components, such as insoluble fiber (35.25 ± 2.74 g per 100 g db, Table 2), may improve fat retention and flavor in some foods, as in the case of cookies.[12]

3.3. Bioactive properties

Phenolic compounds are an important component contributing to the antioxidant capacity (AC) of foods and there is a synergy with vitamins, carotenoids and other components.[35] The total content of polyphenols (Table 2) in powdered cladodes was 2050.20 mg GAE per 100 g db, and is similar to those obtained by Msaddak *et al.*,[12] 2485 mg GAE per 100 g db, and Bensadón *et al.*,[35] 2690 mg GAE per 100 g db. The small difference in total phenolic compounds could be related to the variety, maturity stage, and the extraction method/solvent. Regarding the AC, a value of 312.14 mg Trolox per 100 g db was obtained. The combination of the high percentage of dietary fiber and the content of phenolic compounds present in powdered cladodes can result in an antioxidant dietary fiber with beneficial properties for use as a functional ingredient in food processing.[36]

Table 3 Physical characteristics of cookies made with cladode powder/wheat flour and vegetable oil/butter[a]

Formulation	CP/WF%	O/B%	a_w	L^*	a^*	b^*	Hardness (N)
Control	0/100	0/100	0.14 ± 0.03^{e}	67.52 ± 2.24^{d}	5.83 ± 0.95^{d}	29.36 ± 0.94^{c}	5.56 ± 1.62^{d}
F1	15/85	70/30	0.20 ± 0.004^{d}	$49.76 \pm 1.81^{a,b}$	$1.18 \pm 0.43^{a,b}$	24.60 ± 0.89^{a}	$12.32 \pm 4.93^{a,b}$
F2	25/75	70/30	0.20 ± 0.005^{d}	43.55 ± 1.43^{c}	2.38 ± 0.84^{a}	19.42 ± 2.90^{b}	$8.92 \pm 1.01^{a,b,c,d}$
F3	25/75	50/50	0.20 ± 0.001^{d}	47.41 ± 1.08^{b}	1.03 ± 0.48^{b}	$23.09 \pm 0.56^{a,b}$	$7.88 \pm 1.10^{b,c,d}$
F4	25/75	30/70	0.29 ± 0.003^{a}	47.79 ± 0.37^{b}	-0.43 ± 0.32^{c}	23.49 ± 1.20^{a}	12.50 ± 1.64^{a}
F5	15/85	30/70	$0.28 \pm 0.014^{a,b}$	52.71 ± 0.25^{a}	-0.62 ± 0.41^{c}	26.79 ± 1.21^{a}	$9.13 \pm 1.65^{a,b,c,d}$
F6	20/80	70/30	0.23 ± 0.001^{c}	48.06 ± 1.08^{b}	-0.26 ± 0.34^{c}	23.96 ± 0.71^{a}	$8.36 \pm 1.82^{a,b,c,d}$
F7	20/80	30/70	0.26 ± 0.008^{b}	$50.03 \pm 1.30^{a,b}$	-0.69 ± 0.28^{c}	24.91 ± 1.94^{a}	$10.03 \pm 2.47^{a,b,c}$
F8	20/80	50/50	0.26 ± 0.003^{b}	48.63 ± 0.52^{b}	-0.72 ± 0.40^{c}	23.12 ± 0.77^{a}	$6.37 \pm 1.65^{c,d}$
F9	15/85	50/50	$0.22 \pm 0.001^{c,d}$	51.93 ± 0.13^{a}	-0.50 ± 0.14^{c}	25.33 ± 0.30^{a}	5.63 ± 2.14^{d}

[a] CP/WF: cladode powder/wheat flour, O/B: vegetable oil/butter, a_w: water activity, color parameter: (L^*, a^*, and b^*). Values are the mean ± standard deviation. Different letters in the same column mean significant differences ($p < 0.05$).

3.4. Physical characteristics of formulated cookies with powdered cladode

3.4.1. Water activity. Water activity values (a_w) were in the range of 0.20 to 0.29; the control showed a value of 0.14 (Table 3). Values lower than 0.3 indicate that the cookies are chemically and microbiologically stable.[37] The difference in the a_w of the control in relation to cookies formulated with powdered cladodes may be due to the content of dietary fiber present since it has a greater water holding capacity. The a_w values showed significant differences ($p < 0.05$) according to the formulation. The highest a_w (0.290 ± 0.003) belongs to the formula with the highest content of powdered cladodes (25%). Similarly, high values of a_w were observed for formulae with 15 and 20% of cladode powder but had the same percentage of vegetable oil/butter (30/70); therefore, a higher butter content also influenced the a_w of the cookies due to the presence of milk solids in butter such as proteins and lactose. The lowest values were for cookies with a higher percentage of vegetable oil/butter (70/30). The different ingredients, such as powdered cladodes, wheat flour, vegetable oil and butter, given their chemical structure and water retention capacity, will influence the final a_w. Similar a_w values (0.25 to 0.41) were reported by Uriarte *et al.*[37] for cookies made with amaranth, mushrooms and powdered cladodes.

3.4.2. Color. The addition of cladode powder changes the color characteristics (L^*, a^*, and b^*) of cookies with yellow/brown color to different shades of green. The three-color coordinates present significant differences ($p < 0.05$) according to the cookie composition (Table 3). Specifically, the color parameter L^* decreases in relation to the control (67.52), and the lowest value was for cookies with 25% powdered cladodes. The values of a^* and b^* also decreased in relation to the control (5.83 and 29.36, respectively). Values for a^* were near zero or negative, and b^* values were greater than 19 indicating the predominance of green shade. This may be due to the content of chlorophyll and other phenolic compounds present in the powdered cladodes and the nonenzymatic reaction (Maillard reaction) that takes place between reducing sugars and amino acids causing a browning effect during cooking.[6]

3.4.3. Hardness. The hardness values (Table 3) showed a significant difference ($p < 0.05$) between all samples. The control cookie (5.56) and the formula with 25% powdered cladodes and an O/B percentage of 30/70 (F4) have the highest increase in hardness. Hardness increases with the substitution of oil at 70% and 30%, in contrast, a slight decrease at 50% of oil substitution is observed. Similar trends were observed at the three levels of cladode addition. This could be related to the amount of air entrapped into the cookie dough during its elaboration, but this cannot be retained in the cookie making them hard.[38] In general, all formulae with powdered cladodes caused an increase in the hardness of the cookies. Similar trends were reported by Msaddak *et al.*[15] in cookies made with

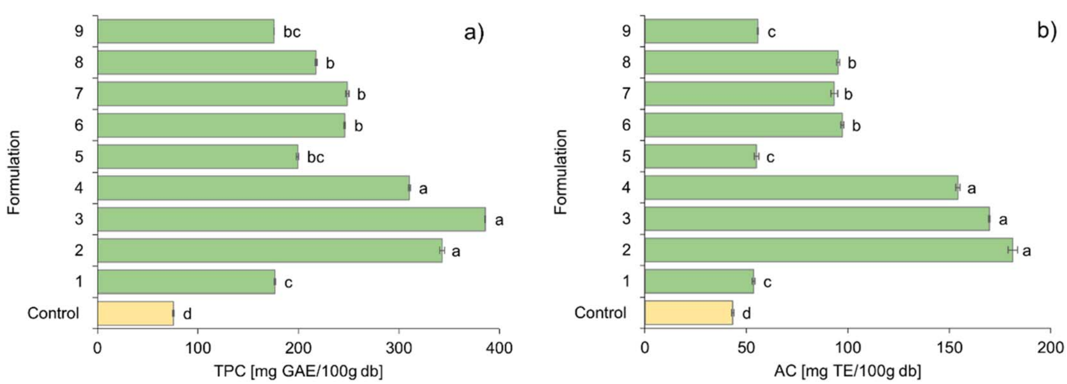

Fig. 1 (a) Total phenolic compounds (TPCs) and (b) antioxidant capacity (AC) of the different cookies' formulations. Different letters between formulations mean significant differences ($p < 0.05$).

powdered cladodes. As the percentage of cladode increased, the hardness of the cookies also increased. This could be attributed to the spreadability of dough during baking; the internal microstructure of cookies changes because of the particle size of cladode powder, which prevents the formation of air cells and a foamy texture.

3.4.4. Bioactive compounds of cookies. The results for total phenolic compounds (TPC) and antioxidant capacity (AC) of cookies are shown in Fig. 1. The TPC content for control was 75.74 mg GAE per 100 g db and was significantly lower ($p < 0.05$) than those of the rest of the formulations. According to Fig. 1a, the increase in the concentration of powdered cladodes increases the TPC. In contrast, the oil/butter (O/B) ratio did not influence the TPC of the cookies. No significant difference ($p < 0.05$) was observed between sample 5, 9 and 1 (values near 180 mg GAE per 100 g db); 4, 3 and 2 (values near 230 mg GAE per 100 g db); and 7, 6 and 8 (values near 340 mg GAE per 100 g db). Different studies suggest that TPC values varied in the range of 100 to 500 mg GAE per 100 g db,[6,7,13,15] based on foods such as cookies, bread, cake, and cladode flakes in different percentages with supplementation of powdered cladodes around 10 to 25%.

For the AC, similar behaviors to TPC were observed (a correlation between both values was $R^2 = 0.8741$); the control cookie had a value of 43.28 mg TE per 100 g db showing a significant difference ($p < 0.05$) with respect to the other cookies. In all cases, by increasing the content of powdered cladodes, the CA was higher, with a good capacity to eliminate free radicals by the DPPH method. The resulting values are in agreement with those found by other authors,[7,15,39] in similar foods with data ranging from 53.29 to 120 mg TE per 100 g db. The results obtained show that cookies added with powdered cladodes have a greater antioxidant potential than control cookies, which reinforces their nutritional value.

3.4.5. Sensory evaluation. The sensory analysis of the different formulations of cookies, as well as the control, was carried out for color, smell, texture, taste and general acceptance, and the results are presented in Table 4. Fig. 2 shows the cookies elaborated for sensory analysis. Control cookies were better rated compared to the cladode added cookies, most of the values were up to 8 score (Fig. 3). Specifically, lower values were obtained in color for formulations F2, F3, F4, F6, F7 and F8, with 20 and 25% (6.58 to 7.10), and as was expected the control (8.5 score) and the lower concentration cladode powder (7.13 score) cookies present the greatest evaluation ($p < 0.05$); this difference could be related to the green color of the cookies with a high amount of cladode powder (Fig. 2 and Table 3). With respect to smell, no significant difference was observed ($p > 0.05$) except for the F8 sample; neither the percentage of cladode nor the ratio of oil/butter influences acceptability. In terms of texture, the increase in hardness due to the addition of the cladode powder influences the preference of cookies. However, as the oil content increases, this could result in hard texture as was discussed previously, and this could affect the acceptability. However, F6 has most of the values near to nine (Fig. 3).

The scores for the taste characteristic show a significant difference ($p < 0.05$) between the control (8.55) and formulae 1, 2, 4, and 8 with scores of 7.10 to 7.37, and these formulae have percentages of powdered cladodes of 15, 20, and 25%. Therefore, the acceptance of the flavor is also influenced by the content of vegetable oil/butter since higher scores generally have a higher butter content or a 50/50 ratio. Finally, for the general acceptance of the cookies, there was no significant difference between the control cookie, with a score of 8.5 and F5 and F9 (15% of powdered cladodes, O/B 30/70 and 50/50) with values of 7.97 and 7.77, respectively. The results obtained show that in general the best evaluated sensory attributes, after the control cookie scores, are for cookies with an addition of 15% powdered cladodes and a vegetable oil/butter ratio of 30/70.

3.5. Moisture sorption isotherms

Sorption isotherms (Fig. 4) were obtained from both the control cookie and the cookie with the highest score in overall acceptance, corresponding to 15% CP and 30% oil/70% butter (F5). An initial humidity of 1.87% was observed for control cookies and 3.24% for the cookies with 15% CP. The isotherms showed an increase in equilibrium moisture content with increasing a_w at the studied temperature of 25 °C. The water adsorbed for each sample corresponds to 0.11 to 0.99 g water per g dry solids for control and 0.17 to 1.15 g water per g dry

Table 4 Results of the sensory evaluation for different cookies made with cladode powder/wheat flour and vegetable oil/butter[a]

Formulation	CP/WF%	O/B%	Color	Smell	Texture	Taste	General acceptance
Control	0/100	0/100	8.50 ± 0.84^{a}	8.0 ± 1.52^{a}	8.30 ± 0.81^{a}	8.55 ± 0.61^{a}	8.50 ± 0.61^{a}
F1	15/85	70/30	6.58 ± 1.54^{b}	$7.19 \pm 1.42^{a,b}$	$6.71 \pm 1.53^{c,d}$	7.35 ± 1.45^{b}	$7.03 \pm 1.22^{c,d}$
F2	25/75	70/30	6.58 ± 1.61^{b}	$6.84 \pm 1.21^{a,b}$	6.29 ± 1.60^{d}	7.10 ± 1.35^{b}	6.61 ± 1.38^{d}
F3	25/75	50/50	7.10 ± 1.56^{b}	$7.39 \pm 1.15^{a,b}$	$6.68 \pm 1.70^{c,d}$	$7.71 \pm 1.13^{a,b}$	$7.06 \pm 1.15^{b,c,d}$
F4	25/75	30/70	6.61 ± 1.63^{b}	$7.03 \pm 1.40^{a,b}$	$6.58 \pm 1.86^{c,d}$	7.32 ± 1.30^{b}	$6.97 \pm 1.05^{c,d}$
F5	15/85	30/70	$7.45 \pm 1.31^{a,b}$	$7.39 \pm 1.31^{a,b}$	$8.00 \pm 0.77^{a,b}$	$8.00 \pm 1.00^{a,b}$	$7.97 \pm 0.84^{a,b}$
F6	20/80	70/30	6.67 ± 1.38^{b}	$7.53 \pm 1.11^{a,b}$	$6.70 \pm 1.62^{c,d}$	$7.47 \pm 1.28^{a,b}$	$7.07 \pm 1.41^{b,c,d}$
F7	20/80	30/70	6.80 ± 1.56^{b}	$7.07 \pm 1.36^{a,b}$	$7.23 \pm 1.38^{a,b,c,d}$	$7.70 \pm 1.15^{a,b}$	$7.40 \pm 1.33^{b,c,d}$
F8	20/85	50/50	6.70 ± 1.64^{b}	6.70 ± 1.76^{b}	$6.87 \pm 1.57^{b,c,d}$	7.37 ± 1.33^{b}	$7.07 \pm 1.08^{b,c,d}$
F9	15/85	50/50	$7.13 \pm 1.68^{a,b}$	$7.27 \pm 1.53^{a,b}$	$7.63 \pm 1.27^{a,b,c}$	$7.77 \pm 1.43^{a,b}$	$7.77 \pm 1.10^{a,b,c}$

[a] CP/WF: cladode powder/wheat flour, O/B: vegetable oil/butter. The values are the mean ± standard deviation ($n = 30$). Values that do not share letters (a, b, c, and d) in the same column are significantly different ($p < 0.05$).

Fig. 2 Images of the cookies obtained for the different formulations (Table 1).

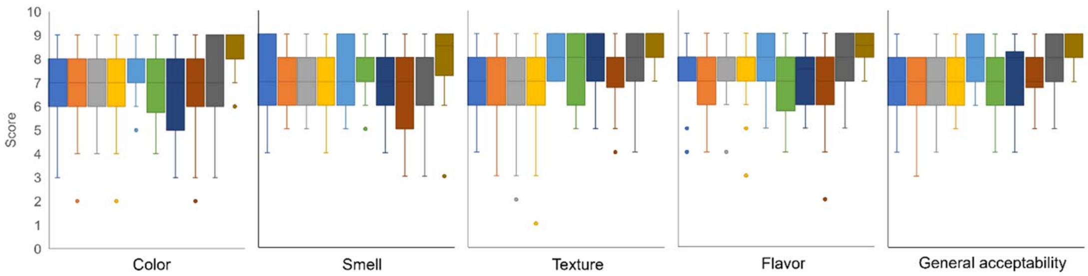

Fig. 3 Effect of the % cladode powder (CP) and % vegetable oil (oil) on the sensory attributes. ■ F1, ■ F2, ■ F3, ■ F4, ■ F5, ■ F6, ■ F7, ■ F8, ■ F9, ■ control.

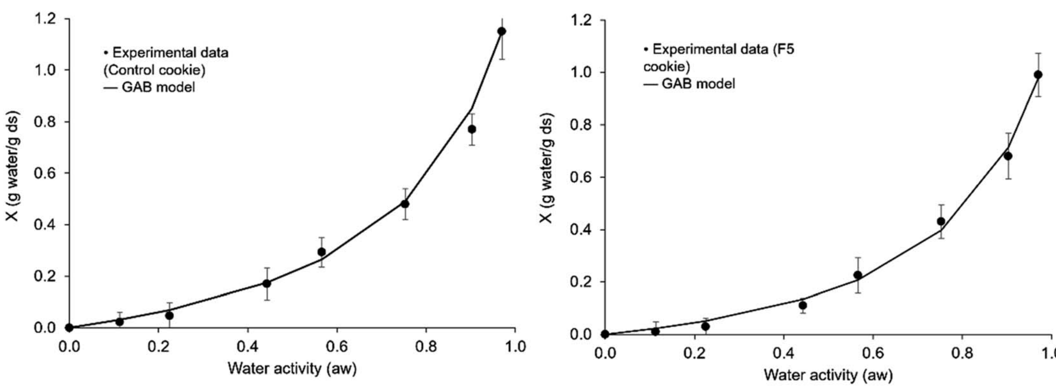

Fig. 4 Sorption isotherms for the control cookie and the cookie with 15% cladode powder (CP) at 25 °C, adjusted to the GAB model. The values are the mean ± standard deviation.

solids for the selected cookie (F5). The shape of the isotherms was sigmoidal type III, which corresponds to products rich in sugar.[40] The moisture sorption data were adjusted to the GAB mathematical model, and the results obtained for the model parameter are shown in Table 5. The value of the monolayer moisture content (X_M) indicates the amount of water that is strongly adsorbed at specific sites on the surface and is considered as the desirable value for food stability.[41,42] In this study, the X_M values were 0.27 and 0.31 g water per g dry solids for the control and selected cookie (15% CP), respectively, and the difference may be due to the soluble dietary fiber content (mucilage) present in the powdered cladodes, which causes greater water sorption.[42] The correlation coefficients R^2 of the isotherms were greater than 0.9, and the error less than 0.01, indicating that the GAB model can be used to predict the behavior of the experimental data. The water sorption properties of food are of great importance in selecting the right packaging materials and being able to predict changes in stability and humidity during storage. An increase in moisture content can directly affect the texture of cookies, which is a key factor in their acceptance by consumers.[43]

Table 5 GAB model parameter values and statistical coefficients for cookie isotherms evaluated at 25 °C[a]

Parameter	Control cookie	Cookie 15% CP
X_M	0.2723	0.3169
C	0.8148	0.9817
K	0.8155	0.810
R^2	0.9980	0.997
E	0.006	0.002
E_{RMS}	0.0313	0.0161

[a] X_M: moisture content of the monolayer. C and K are interaction energy constants for the monolayer and the other water molecules. R^2: linear regression coefficient, E: error, E_{RMS}: root mean square error.

4 Conclusion

In summary, the proximal chemical analysis of the powdered cladodes of *Opuntia ficus indica* var. Atlixco showed a high content of dietary fiber and phenolic compounds with antioxidant capacity. In the formulation of the cookies, the substitution of wheat flour for powdered cladodes significantly improved the content of total phenolic compounds and the antioxidant capacity in all cases compared to cookies made only with wheat flour, being higher for formulations with 25% powdered cladodes. For the sensory evaluation, cookies with 15% powdered cladodes and 30% vegetable oil content showed the best taste and overall acceptability score, relative to the control. The sorption isotherm showed that it was possible to fit the GAB model to the experimental data and the cookies are stable at 25 °C. Based on the results obtained, powdered cladodes can be considered as a functional ingredient and a source of dietary fiber and antioxidants for the manufacture of foods with benefits to human health and nutrition.

Author contributions

Aparicio-Ortuño Rocío: conceptualization, methodology, investigation, data curation, writing-original draft preparation, writing-review and editing, and visualization; Jiménez-González Oscar: data curation, formal analysis, and methodology; Lozada-Ramírez J. Daniel: conceptualization, methodology, formal analysis, draft preparation, writing-review and editing, and supervision; Ortega-Regules Ana E: conceptualization, methodology, validation, formal analysis, investigation, resources, writing-original draft preparation, writing-review and editing, supervision, project administration, and funding acquisition.

Conflicts of interest

The authors declare no conflict of interest.

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