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### OBTAINING A FUNCTIONAL TORTILLA MADE FROM NOPAL FLOUR FORTIFIED WITH SPIRULINA

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All content in this magazine is licensed under a Creative Commons Attribution License. Attribution-Non-Commercial-Non-Derivatives 4.0 International (CC BY-NC-ND 4.0). Abstract: Since ancient times, the procedure for making tortillas has not undergone significant changes in terms of the way it is made and its main ingredients. Being a highly consumed product in Mexico, it can be used to provide the population with some benefit at the time of ingestion. The new trends in food consumption and changing lifestyles have focused on finding healthier products that, in addition to their nutritional contribution, have a beneficial effect on health. These needs have prompted the search for alternatives for the production of functional foods. Spirulina has a very high macro and micronutrient content. It is considered a complete food supplement to fight malnutrition deficiencies. Finally, they have the competitive advantage of not requiring complex and expensive technological infrastructures for their largescale production.

Keywords: Fortified tortillas, Nopal flour, Spirulina

### INTRODUCTION

Corn is the main food of the Mexican population and an important ingredient in the diet of many countries in the Americas. Pozole and pinole are prepared from the grain, and tortillas, tamales and tostadas are obtained from the processed and ground grain (masa). The official Mexican standard (NOM-187-SSA1/SCFI-2002) defines Tortilla as a product made with dough that can be mixed with optional ingredients, subjected to cooking. It can also be defined as a flat, flattened, skinny, round bread made from corn. In Mexican cuisine, white corn, blue or purple corn tortillas are popular, with yellow corn being the most popular. Mexicans consume an average of 68 kg of tortillas per year, which is equivalent to an energy content of 410 Kcal (around 20% of daily caloric intake). The economic and social relevance of nixtamalized corn products has made them

an excellent alternative, to be used as carriers of macronutrients and micronutrients to improve the nutrition of large groups of people (Chuck Hernández and Serna Saldivar, 2019).

In developing countries, fortification and fortification of staple foods are the most effective ways to improve the nutritional status of the population. When widely practiced, enrichment and fortification improve nutrient intake, individual job performance, and public health (Serna-Saldivar, 2015).

The growing demand for so-called functional foods has highlighted the need to find other natural options for this type of food. The corn tortilla is considered an excellent source of calories due to its high starch content and a great option for people who are gluten intolerant. Unfortunately, it lacks good protein quality and adequate levels of micronutrients and dietary fiber (Serna-Saldivar, 2015). Tortillas have been fortified with soy proteins and vitamins, obtaining positive effects on their nutritional properties. Other authors have studied the sensory characteristics of fortified tortillas, managing to increase their protein value by adding Phaseolus lunatus concentrate, without affecting their acceptability.

Spirulina sp. (Arthrospira sp.) is the oldest living plant on earth (Soni, et al., 2017), it is an undifferentiated filamentous multicellular cyanobacterium, inhabiting alkaline lakes, which is cultivated for human consumption due to its nutritional content and has achieved considerable popularity in the health sector, the food industry and aquaculture (Soni, et al., 2017 and Ramírez-Moreno et al., 2006). It is divided into two every 7 hours, under ideal conditions it can generate about 15,000 kg/ha of dry material per year. It grows in adverse environments, which is why it has been able to absorb the necessary elements to survive, being one of the first algae to carry out photosynthesis, it has an enormous capacity to

store nutrients (Ponce López, 2013). Its value lies in the large amount of macronutrients and micronutrients it contains, some of which cannot be synthesized by the human organism (Ramírez-Moreno, et al., 2006). It is the most nutritious concentrated food known to mankind and is a rich source of antioxidants, phytonutrients, probiotics and nutraceuticals (Soni et al., 2017). It is legally authorized as a food supplement in Europe, Japan and the Pacific Rim. In the United States, the FDA authorized it from 1981, indicating that it is a protein source that also contains vitamins and minerals. It is rich in proteins (30-70%), amino acids (55%; essential), carbohydrates (20%), vitamins (vitamin A beta-carotene 4.60 times, vitamin B1 0.21, vitamin B2 riboflavin 0.21, vitamin B3 niacin 0.07, vitamin B6 pyridoxine 0.04, vitamin B12 5.33 times and vitamin E alpha-tocopherol 0.03) and others. nutrients (3%), so one of its main uses is as a food supplement, either powdered, encapsulated, in tablets or as a flour substitute, in pastes for soups, sauces, granola bars, candies, or instant fruit or vegetable drinks (Ramírez-Moreno and Olvera-Ramírez, 2006). Spirulina has 30-70% protein, which is higher if a comparison is made with different foods, such as soybean has 34%, corn 9%, and grain-fed beef 20% protein (Ponce López, 2013). The nopal for its part contributes a high proportion of water to the diet and is highly valued for its fiber content; They are part of the common diet of Mexico. It is rich in dietary fiber, its content comparable to that of various fruits and vegetables, including spinach, artichoke, chard, eggplant, broccoli, radish and others. The objective of the research was to formulate and characterize a tortilla based on nopal flour and Spirulina, which improves its nutritional quality by offering the consumer an alternative for healthy consumption, taking into account that current estimates indicate that 23.5% of the population lives in food poverty (CONEVAL, 2022). This tortilla could help to comply with the food sovereignty of several communities since the nopal is endemic to a wide geographical area of the country and there are several easy methodologies for the production of Spirulina where you can have a growth of 25% per day.

### METHODOLOGY

Obtaining nopal flour, the nopal was cut into slices approximately one centimeter thick, and they were dried in a convection oven at  $80\pm0.2$ °C for 48 hours. Once the nopal was dehydrated, the nopal was ground and sieved using a # 80 sieve, with a particle size of 177µm.

Tortilla preparation, two tortilla formulations and one control (TC) were made. The treatments were coded as follows: T1X, Y; where the subscript X corresponds to the percentage of nopal flour and the subscript Y to the percentage of Spirulina used (Table 1). The tortillas were made with the partial substitution of wheat flour for nopal flour and Spirulina. For the molding of the tortilla, a manual tortilla machine with a plastic cover was used, standardizing the weight, size, thickness and cooking time of each tortilla (Table 3). The dough was gently pressed to obtain the tortilla, cooking at a temperature of  $150 \pm 5^{\circ}$ C, with a cooking time of 1:40 min for each tortilla, distributed as follows: 30 seconds on the first side and then flipping the second side with a time of 50 seconds and returning to the first side with a time of 25 seconds.

Ingredient	T <sub>c</sub>	T1	T2
Nixtamalized corn flour (%)	100	80	75
Nopal flour (%)	_	19	23.5
Spirulina (%)	_	1	1.5
Water (mL)	150	150	150

Table 1. Formulations for the preparation of tortillas fortified with Spirulina and nopal flour.

### Bromatological analysis

The determination of humidity, ashes, protein, fat and fiber was carried out on the Spirulina sample (Mi granero, San Pedro Cholula, Puebla).

### Moisture determination

The samples were dried at a temperature of 100-103°C for a period of 24 hours. The weight of the crucible with dry sample was taken, the data is recorded and the corresponding calculations were made.

### Determination of total ash

This analysis was carried out using a dry method and the sample obtained from humidity was used, which were pre-incinerated until they stopped emitting smoke, then they were placed in the muffle for a period of 2-3 hours at a temperature of 600 °C. The data is recorded and the corresponding calculations were made.

## Determination of ether extract or crude fat Soxhlet method

4 g of dry sample were used on filter paper on an analytical balance and later deposited in cellulose cartridges. Hexane was added to the flasks, coupled to the refrigerant of the Soxhlet device, extracting for a period of 8 hours, counting the time from when it began to boil.

### Determination of crude fiber

The previously defatted sample of the determination obtained in ethereal extract was used, 2 g was weighed and placed in Berzelius glasses, 100 mL of 0.225 N sulfuric acid was added to each glass. The fiber was passed back to the Berzelius glass, now adding 100 mL of 0.313 N sodium hydroxide, left for 30 minutes, counted from the start of boiling, after the After some time, the glasses were removed and again the contents

were filtered. It was washed with 3 portions of 100 mL of hot distilled water. The sample was dried at a temperature of 100-103 °C for a period of 12 hours. Finally, the samples in the crucibles were pre-incinerated on electric grills and placed in the muffle for 3 hours at a temperature of 600 °C.

#### Crude protein Kjeldahl method

Digestion: 1 g of sample was weighed, the catalyst and 30 mL of concentrated sulfuric acid were placed, finally the flask was placed in the kjeldahl apparatus in the digestion section, connecting the fume extractor. This is done for each of the sample replicates.

Distillation: The result of digestion was diluted with 300 mL of distilled water and cooled. Separately, in an Erlenmeyer flask, 50 mL of 4% boric acid and 5 drops of mixed indicator (methyl red and bromocresol green) were added. Subsequently, 110 mL of 45% sodium hydroxide and 3 zinc shots were added to the Kjeldhal flask without stirring.

Assessment: After 250 mL of distillate, they were titrated with 0.1 N sulfuric acid until a blue to pale pink color was obtained and with the reading obtained from the spent mL of sulfuric acid, the corresponding calculations were made using the following formula:

 $%N = \frac{(mL \text{ spent from acid} - mL \text{ of blank}) (N \text{ from acid}) (0.014)}{\text{g of sample used}} X100$ %PC = (%N) (Conversion factor)

### Statistic analysis

All the determinations were made in triplicate, reporting the average value of the repetitions and their standard deviation, additionally a one-way analysis was applied followed by a comparison of means by Tukey with a reliability of 95%. The program used was Sigmaplot 12.0. The independent variable was established as the type of formulation used to make the tortilla and the amount of protein, fat, ash, and moisture as the dependent variable.

### RESULTS

### OBTAINING AND CHARACTERIZATION OF NOPAL FLOUR

The nopal flour presented the appearance of a fine powder with small agglomerated particles, not uniformly distributed that break apart under pressure; light green, opaque; intense smell, slightly spicy on the nose, vegetal, herbaceous, presenting a soft and adherent texture to the manual touch (Figure1).



Figure 1. Obtaining nopal flour, a) fresh nopal and b) nopal flour.

Table 2 shows the chemical characteristics of nopal and Spirulina flour. Regarding the nopal flour obtained, the fiber content ( $51.21 \pm 1.16\%$ ) is similar to that reported by Castillo et

al., (2013), with a content of 47.65%. Rodiles-López et al., (2017) mention that nopal flour has 37.45% fiber, which is lower than what was reported in this research. The contribution in dietary fiber of the nopal is important, by virtue of the fact that the consumption of soluble type fibers represents a significant improvement in digestive processes, constipation and dietary foods demanded by consumers (Berigüete et al., 2012). Diego-Sarate et al., (2021), indicate that the cladodes of the nopal, stems or pencas of the nopal are an important source of fiber, mucilage and calcium, three components that are necessary to integrate a healthy diet. In addition, they possess antidiabetic and antioxidant activities, which have been attributed to significant amounts of polysaccharides, lignans, flavonoids, and phenolic acids. They also contain palmitelaidic acid (C16:1 trans-9), a monounsaturated fatty acid that regulates fatty acid metabolism for use in body weight control.

As for Spirulina, it has drawn the attention of researchers and experts in human nutrition due to its high content of macro and micronutrients beneficial to health. The chemical composition of Spirulina was quantified in order to verify its nutritional quality (Table 2), observing that it has a high protein content (35.87 ± 0.32%). Gutiérrez (2015), in his study, carried out a laboratory analysis of Spirulina, in which he shows the results generated, reporting the following components, fat 4.3g, carbohydrates 17.8g, and vitamin A. Regarding its protein content, it is reported that it has a protein content of 40-70%, finding the results in the reported range (35.87  $\pm$  0.32%). The nutritional composition of Spirulina corresponds to an energy value of 378 ± 1 kcal/100g (da Silva et al., 2019). In general, these results corroborate previous investigations of the nutritional composition of the Spirulina.

Treatment	Protein	Lipids	Ashes	Humidity	Fiber
			(%)		
Prickly pear flour	$0.80 \pm 0.02$	0.50 ± 0.26	7.37 ± 0.25	11.36 ± 1.32	51.21 ± 1.16
Spirulina	$35.87\pm0.32$	$2.57\pm0.20$	$6.26\pm0.29$	$11.50 \pm 1.28$	$0.54\pm0.07$

Table 2. Chemical composition of nopal flour and Spirulina

Treatment	Mass weight (g)	Diameter (cm)	Tortilla Weight (g)	Thickness (mm)
TC	$34.93 \pm 0.70$	$12.09\pm0.24$	$23.20\pm0.30$	$1.83 \pm 0.07$
T1 <sub>19:1</sub>	$34.60\pm0.61$	$12.12\pm0.20$	$22.96 \pm 0.43$	$1.56\pm0.33$
T2 <sub>23.5:1.5</sub>	$35.31\pm0.66$	$12.30\pm0.68$	$23.64\pm0.54$	$1.78\pm0.28$

Table 3. Physical characteristics of the tortilla fortified with nopal flour and Spirulina.

Treatment	Protein	Lipids	Ashes	Humidity	Fiber
			(%)		
T <sub>c</sub>	$11.23\pm0.26^{\rm a}$	$5.68\pm0.72^{\text{a}}$	$1.69\pm0.30^{\rm a}$	$10.36\pm0.70^{\rm a}$	$7.43\pm0.37^{\text{a}}$
T1 <sub>19:1</sub>	$22.82\pm0.78^{\rm b}$	$10.33\pm0.90^{\rm b}$	$2.64\pm0.58^{ab}$	$15.20\pm0.82^{\rm b}$	$8.27\pm0.17^{\rm b}$
T2 <sub>23.5:1.5</sub>	$26.25\pm0.94^{\circ}$	$11.34\pm0.48^{\rm b}$	$3.39\pm0.31^{\rm b}$	$13.45\pm0.51^{\circ}$	$8.92\pm0.13^{\circ}$

Table 4. Chemical composition of tortillas fortified with nopal flour and Spirulina.

### PREPARATION AND CHARACTERIZATION OF TORTILLAS

Consumers are increasingly interested in foods that can reduce the risk of disease, which benefits their health and well-being. Spirulina is known as a superfood, due to its high concentrations of nutrients and some other beneficial properties for health due to the presence of bioactive compounds, which include polyunsaturated fatty acids, phycocyanin and carotenoids (Franco Lucas et al., 2023). Currently, studies have been carried out to evaluate consumer perceptions about Spirulina, finding that the main reasons why participants consumed Spirulina as a food or dietary supplement was related to improving and maintaining health (Moons et al., 2018; Rzymski et al., 2017). Franco Lucas et al., (2023), conducted a study on

consumer attitudes towards Spirulina as a food ingredient, they report that consumers indicated that the products that were most likely to be enriched with Spirulina in their diet were pasta and cereal bars, and their work contributed to broaden the knowledge about consumer attitudes towards Spirulina. Therefore, hence the feasibility of being able to strengthen a food of first consumption in Mexico, such as the tortilla, with an ingredient such as Spirulina. Table 3 shows the physical characteristics of the tortillas made, such as the weight of the dough, diameter, weight of the tortilla, and thickness. It was tried that all the characteristics of the three treatments did not have significant differences so that they did not intervene in the results of the tests.

The fortified tortillas presented resistance to handling and color change, in figure 2 the two treatments can be seen (T1<sub>19,1</sub> and T2<sub>25,3:1.5</sub>) compared to control (TC). Table 4 shows the chemical composition obtained from each treatment. The results show that the protein and fat content increased significantly for the fortified treatments (T119.1: 22.82 and 10.33 %;T2<sub>235,15</sub>: 26.25 and 11.34 % respectively) with the addition of nopal flour and Spirulina compared to the control (11.23 and 5.68%), this increase being desirable considering that, in nopal, 80% of its fatty acids are mainly made up of linoleic acid (Tang and Suter, 2011), an unsaturated and essential fatty acid for humans; and the protein content for the nutritional value that Spirulina provides. Chuck Hernández and Serna Saldivar, (2019), report the nutritional value of regular tortillas (corn flour), containing minerals (1%), moisture (40%), protein (6%), and fat (3%), only the moisture value being higher compared to the tortillas obtained with nopal/ Spirulina. In the same way, they fortified and enriched the tortilla by adding soy flour, reporting a moisture content of 7.19%, ash 1.96%, protein 10.56%, fat 3.90%, and fiber 1.22%. The results they report are similar to those found with the control treatment (TC) of the tortillas, and the values for the treatments are higher.: T1<sub>191</sub> and T2<sub>25315</sub> Compared to other authors, the protein and fat values reported in this work are higher, Salinas-Moreno et al (2017), indicates values between 7 and 10% and 2.4 to 3.5% respectively, likewise for tortillas enriched with seaweed .: Ulva clathrata, Quintero-Gutiérrez et al 2014, report 10.66% protein, 1.03% fat and 72.85% carbohydrates, a figure that represents almost double what is contained in the tortilla of the present investigation. Some of the foods that have been enriched with spirulina include yogurt (da Silva et al., 2019).



Figure 2. Fortified tortillas, a) TC, b) T1<sub>19,1</sub> and c) T2<sub>25,3:1.5</sub>

Expressed in grams per 100 g of tortilla. (%). Where  $T_c$ : Control treatment; $T1_{x,y}$ ; where the subscript X corresponds to the percentage of nopal flour and the subscript Y to the percentage of spirulina used.

<sup>abc</sup> Different letters in the same column indicate a significant difference (P>0.05) between the values. One-way analysis of variance and comparison of means by Tukey.

### CONCLUSIONS

Fortification with Spirulina and nopal flour shows great potential to increase the nutritional capacity of tortillas. The implications of this study are that the addition of nopal flour and Spirulina allows the design of functional foods, since apart from being fortified, it could provide antioxidant content (which is under evaluation). The combination of a traditional staple food and a traditional medicine vector is an innovative approach to promote the systematic intake of healthy food products with potential improvement in human health in, for example, vulnerable rural and urban communities where profound change in the agri-food system is necessary if we are to feed more millions of hungry and nutritionally deficient people; sustainable food production are crucial to help alleviate the risks of hunger in Mexico.

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