Use of coconut mesocarp and extracts of cocoa cuticle to increase antıoxıdant capacity of bakıng products.

Ortiz-Moreno A.[1*], Cervantes-Serrato Lf.[1], Plazola-Jacinto Cp.[1], Garduño-Siciliano L.[2], Sánchez-Pardo Me.[1]

[1]1Instituto Politécnico Nacional, Escuela Nacional de Ciencias Biológicas, Departamento de Ingeniería Bioquímica. Avenida Wilfrido Massieu esquina Cerrada Miguel Stampa S/N. c. p. 07738 Delegación Gustavo A. Madero. Ciudad de México.

[2] Instituto Politécnico Nacional, Escuela Nacional de Ciencias Biológicas, Departamento de Farmacia. Avenida Wilfrido Massieu esquina Cerrada Miguel Stampa S/N. c. p. 07738 Delegación Gustavo A. Madero. Ciudad de México.

*corresponding author:

e-mail: ortizalicia@hotmail.com

Abstract

Due to their phytochemical content in the last decade the interest for using food by-products has been increasing. These compounds could be used as raw material to generate improved food products. The Mexican chocolate and cocoa industries generate annually approximately 171 thousand tons of byproducts even than coconut industry. Nowadays the disposal of these byproducts is inadequate, generating pollution problems.

The aim of this work was to evaluate the increase of phytochemical content and antioxidant activity of “chapata” (a Mexican bakery product), added with coconut mesocarp and cocoa cuticle extract. Chapata presented 10 fold higher polyphenol content than control; the antioxidant capacity was 5 fold higher measured by DPPH and 3 fold higher by ABTS method. Confocal Laser Scanning Microscopy micrograps, showed formation of alveoli uniformly distributed with smaller size in relation to the control chapata, due to the higher fiber content. The improved chapata, showed greater acceptance in terms of flavor and color, obtaining an "I like" rating with 100 untrained panelist.

Keywords: Byproducts, coconut, cocoa, polyphenols, antioxidant activity.

1. Introduction

Nowadays, the reuse of forest, agricultural or livestock byproducts is a priority to reduce environmental pollution (Saval, 2012; Yepes *et al*., 2008). An advantage of these byproducts is their low cost affordable for using them as a raw material. The cocoa and coconut industries produce around 80% of byproducts, which represents for Mexico approximately 170 000 tons (SAGARPA, 2013). Among the compounds that these byproducts contain, the phenolic compounds have nowadays economic relevance. Phenols are biomolecules capable of neutralizing free radicals (Zia-ui-hag *et al*. 2014). The identification, quantification and extraction of phenols have aroused great interest because they have been found to possess beneficial properties for human health such as: anticancer activities, anti-inflammatory, among others. The positive health effects have attracted the attention of researchers for incorporating these compounds into foods (Charles, Martínez-Flórez, González-Gallego, & Tuñón, 2002). Besides of the phenol content, the agro industrial residues are rich in fiber, recommended as part of a healthy diet and is related with a decrease in the glycemic index (Ramírez and Pérez, 2010; Jerkins *et al*., 2005). The incorporation of other components to foods has the objective of improve the nutrient intake for maintain a balanced diet (Ramírez and Pérez, 2010). Some foods made from wheat flour have been incorporated into products containing fiber and bioactive compounds, in order to provide health benefits, in addition to developing a functional product (Irakli *et al*., 2015; Peng *et al*., 2010, Trinidad *et al*., 2006).

The coconut is native to Southeast Asia, whose scientific name is *Cocos nucifera* L., is a monocotyledon of the order of the Palmae, of the family Arecaceae and is the only species that has the genus Cocos. Approximately 60% of the coconut fruit are discarded waste, without any utility. They have a high content of fiber and phenolic compounds (Abad *et al*., 2002; Raghavendra *et al*., 2006).

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Cacao (*Theobroma cacao* L.) is a tropical plant and expands from the Mexican jungles to the Amazon. Their domestication was induced by the pre-Hispanic Mesoamerican cultures that called it "food of the gods"; The Mayans and Aztecs, besides using it as a drink, also used their grains as a coin. The fruit commonly called "cacao ear" is an elliptical berry of yellow, red, purple or coffee that can reach a length of 15 to 25 centimeters. This cocoa ear is composed of 20% of grains, 22% of cuticle, 2% of mucilage and 56% of pod, with white, purple or reddish-brown cotyledons depending on the genotype and the seeds are wrapped in a pulp or mucilage very moist, white and sweet; which is why fresh or recently extracted cocoa from the "cob" is called "cocoa in drool". It is usually a small tree, between 4 and 8 meters high (Baena *et al*, 2012; Cacao Mexico, 2016).

In this work the evaluation of the antioxidant capacity of a bakery product added with agroindustrial residues of cocoa cuticle and coconut mesocarp was carried out, with the purpose of developing new products that contribute to attenuate health problems in the human body.

2. Materials and methods

Chapata bread was made by replacing wheat flour with coconut mesocarp powder and cocoa cuticle extract, keeping other baking ingredients constant. The baking process consisted of the following stages: mixing, fermentation, rounding and baking, using lab traditional instruments and equipment of the baking process.

The analyzes performed in the products included:

Total Polyphenols. Extraction and Quantification. The extraction and quantification of total polyphenols were performed according to the methodology of Ribeiro *et al*. (2008).

Antioxidant capacity was performed by ABTS and DPPH. Baking Quality, Specific volume (EV) 10-05.01 (AACC, 2001).

Confocal Laser Scanning Microscopy (MCBL). Two stains were made using Calcofluor and Rodamina B. The calcofluor dyes to the cellulose emitting a wavelength of 500 to 520. A laser light was used at 405 nm.

Sensory evaluation. It was determined based on a hedonic scale, carried out by 100 non-trained evaluators.

Statistical analysis. The different measured parameters were analyzed using a multiple ANOVA ($P < 0.05$), followed by a Tukey test for the comparison of means (95% confidence level). Minitab 17 software was used for statistical analysis.

3. Results and discussion

The content of total polyphenols and their corresponding antioxidant capacity of coconut mesocarp powder and aqueous extract of cocoa cuticle are shown in Table 1.

Table 1. Total polyphenols content and antioxidant activity of coconut mesocarp powder and cocoa cuticle.

Sample	Coconut mesocarp powder	Cocoa cuticle
Total polyphenols (mg GAE/g f.w.)	33.77 ± 7	2.21 ± 0.16
Antioxidant activity (µmol TE/g f.w.)	473.71 ± 1.45	255.94 ± 3.87

Average values ± triplicate standard deviation.

f.w. fresh weight basis

The content of phenols in coconut mesocarp (33.77 mg EAG / g sample) was higher than values that reported by Rodríguez *et al*., (2008) for the same mesocarp (22.44 ± 0.15 mg GAE / g sample). The variation in the content of phenolic compounds, is influenced by the solubility of these compounds in the solvent used during the extraction and also is affected by the geographical origin of the crop, the harvest season, the variety, the degree of maturity and the storage time (Charles, 2012; Martínez-Flórez *et al*., 2002; Shahidi and Ambigaipalan, 2015).

The coconut mesocarp powder showed an antioxidant capacity of 473.71 ± 1.45 µmol TE / g sample. This value is higher than fruits that are considered high antioxidant capacity, such as blueberry (149.8 µmol TE / g sample), blackberry (114.8 µmol TE / g sample) and strawberry (44.4 µmol TE / g sample), as reported by Huang *et al*. (2012). These results suggest that coconut mesocarp powder could be considered as a good source of polyphenols (Shahidi and Ambigaipalan, 2015).

The content of total polyphenols in the aqueous cocoa cuticle extract resulted of 2.21 ± 0.16 mg GAE / 100 g sample. Although polyphenols are low soluble in water, our aqueous extract presented a value similar to that reported by Sangronis *et al*., 2014, for Venezuelan coconut cuticle powder (2.49 ± 0.06 mg GAE / 100 g sample) but lower than other agricultural residues as mango seed (48.58 mg GAE / g sample) (Paz *et al*., 2015). This variation depends on the solubility of these compounds in the solvent used during the extraction process. (Martinez-Flórez *et al*. 2002; Shahidi and Ambigaipalan, 2015). Since our extract was done with water in order to use this solution as raw material for the chapata production, the extraction was low but enough for our objective.

The aqueous extract of cocoa cuticle showed an antioxidant capacity of 255.94 µmol TE / g sample, which was lower than that reported by Sangronis *et al*., 2014, for a Venezuelan cocoa cuticle of 422 µmol TE / g sample, this difference could be due to the polyphenols extracted with water in this research, presented a lower antioxidant activity. However, the antioxidant effect of this product exceeds the values reported by Huang *et al* (2012) for different fruits considered to have high antioxidant capacity, such as blueberry (149.8 µmol TE / g sample), blackberry (114.8 µmol TE / g sample) and strawberry (44.4 µmol TE / g sample). These results suggest that cocoa cuticle aqueous extract as an ingredient could become a good source of polyphenols. (Shahidi and Ambigaipalan, 2015).

Table 2. Total polyphenol content and antioxidant activity of chapata

	CC	CCP	CCE	CCPCE
Total polyphenols (mg GAE / g sample d. w.)	0.45 ± 0.01[a]	5.86 ± 0.05[b]	1.17 ±0.06[c]	5.43 ± 0.07[b]
DPPH (µmol TE/ g sample d. w.)	102.02 ± 1.59[a]	493.31 ± 0.46[b]	184.59 ± 4.41[c]	503.57 ± 0.29[b]
ABTS (µmol TE/ sample d. w.)	1061.56 ±171[a]	3116.89 ± 341[b]	1545.35 ± 182[c]	2909.48 ± 120[b]

Average values ± triplicate standard deviation. Different letters in the same row indicate significant differences according to the Turkey test at a P <0.05. CC - Chapata control: 0% replacement;

CCP - Chapata with coconut mesocarp powder: 6% of substitution;

CCE - Chapata with cocoa cuticle extract: 50% substitution.

CCPE - Chapata with coconut mesocarp powder and cocoa cuticle extract: 6% substitution and 50% substitution, respectively.

d.w. dry weight basis

The antioxidant capacity of chapatas made with coconut mesocarp, cocoa cuticle aqueous extract and chapatas added with both ingredients are presented in Table 2. As expected, chapatas polyphenol content, antioxidant capacity measured by DPPH and ABTS assays were higher than control. Polyphenol content of chapata added with both ingredients presented significant (P <0.05) increase from 0.45 to 5.43 mg GAE/g sample d.w. while antioxidant capacity also presented significant increases (P <0.05) from 102 to 503 µmol TE/g sample d.w. using DPPH and from 1061 to 2909 µmol TE/g sample using ABTS method.

Peng *et al.*, 2010, have reported that the incorporation of grape seed extracts into the baking formulation are affected in the concentration of antioxidant capacity, due to several factors, such as heat induced reactions, Maillard reactions and oxidative stress (induced by the incorporation of oxygen during the kneading phase, during the cooking process). Therefore, the polyphenol antioxidant capacity in the chapata was increased in almost 5 fold measured by DPPH method and almost 3 fold with ABTS method.

Figure 1 shows the microphotographs of the chapata control and added with coconut mesocarp powder and cocoa cuticle aqueous extract. This image allows observing the topography of the products. In the Figure 1A is possible to observe control chapata starch granules with shape of disc and the alveoli formed by the gluten network. In Figure 1B is shown the microphotograph of chapata added with residues and is possible to observe that the alveolar distribution is smaller in comparison with the control chapata. This can be explained by the presence of fiber that decreases the formation of the three-dimensional network; however, it is possible pointed out that exist the formation of evenly distributed alveoli with a smaller size in relation to the control chapata.

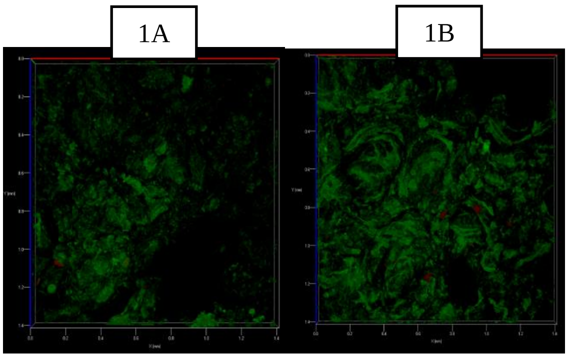

Figure 1. Topography of A) control chapata, B) Chapata added with residues

Sabanis *et al.*, 2009, have reported that the incorporation of fiber (hydroxymethylpropylcellulose) into the baking formulation alters the structure, because the fiber component within the highly developed protein-starch-fiber network may decrease starch binding -protein and thus explain the increase in bread firmness, which represents a dense structure.

The bread with coconut mesocarp and cocoa cuticle extract obtained a level of acceptance of "like" of 100 untrained evaluators (see table 3); 19% rated "I like it very much," and 40% gave a "like" rating. The comments were: the product has a good appearance, a strong smell and flavor, with a compact crumb, which improve the flavor.

Table 3. Sensoryal analysis of chapata

Parameter	CC	CCP	CCE	CCPCE
Color	7.0 ±1.79[a]	6.51 ±1.88[a]	7.74 ±1.41[b]	6.65 ±1.91[a]
Odor	7.82 ±1.62[a]	7.34 ±1.54[a]	7.88 ±1.55[a]	7.45 ±1.59[a]
Texture	7.4 ±1.81[a]	7.34 ±1.91[a]	7.6 ±1.85[a]	7.17 ±2.11[a]
Flavor	7.37 ±1.58[a]	7.48 ±1.43[a]	8.11 ±1.31[b]	7.68 ±1.72[a]

Average values; ± triplicate standard deviation. Different letters in the same row indicate significant differences according to the Turkey test at a P <0.05. * CC - Chapata control: 0% replacement;

CCP - Chapata with coconut mesocarp powder: 6% of substitution;

CCE - Chapata with cocoa cuticle extract: 50% substitution.

CCPE - Chapata with coconut mesocarp powder and cocoa cuticle extract: 6% substitution and 50% substitution, respectively.

There were not significant differences (P <0.05) between the level acceptance of the control chapata and chapata added with coconut mesocarp and cocoa aqueous extract; indicating that the preference of the consumer did not change when adding the residues to the baking formulation.

In previous research (Raghavendra *et al.*, 2006) reported that when fibers are added to cooking products, smell and taste are favored, because the components responsible for these attributes are retained by the fibers.

4. Conclusions

Coconut mesocarp powder and the cocoa cuticle aqueous extract increased the phenol content and the antioxidant capacity of chapata obtaining an acceptable product from the point of view of its organoleptic characteristics. Both byproducts could be considered a good source of phenolic compounds. Further research has to be conducted in order to determine phenols extraction process at industrial level and cost analysis.

References

AACC. (2001). The definition of dietary fibre, AACC Report. Cereal Food World, 46, 112–126.

Baena, L. M., García Cardona, N. A., Industrial, Q. (2012). Obtención y caracterización de fibra dietaría a partir de cascarilla de las semillas tostadas de *Theobroma cacao L*. de una industria chocolatera colombiana. *Química Industrial. Retrieved* from http://repositorio.utp.edu.co/dspace/hand6

Cacao México. (2016). Retrieved from http://www.cacaomexico.org/?page_id=1051

Charles, D. J. (2012). Antioxidant properties of spices, herbs and other sources. *Springer Science & Business Media*. 3-65. http://doi.org/10.1007/978-1-4614-4310-0_1

Huang, W., Zhang, H., Liu, W., LI, C. (2012) Survey of antioxidant capacity and phenolic composition of blueberry, blackberry, and strawberry in Nanjing. *Journal of Zheijang University SCIENCE B-Biomedicine & Biotechnology*. 13(2), 94-103.

Irakli, M., Katsantonis, D., Kleisiaris, F. (2015). Evaluation of quality attributes, nutraceutical components and antioxidant potential of wheat bread substituted with rice bran. *Journal of Cereal Science*. 65, 74–80. http://doi.org/10.1016/j.jcs.2015.06.010

Jerkins, D. (2005). Fibra y enfermedad cardiovascular, V Simposio Nestle Fibra Alimentaria en la Salud y en la enfermedad, Glosa, S. L., Barcelona, España.

Martínez-Flórez, S., González-Gallego, J., Culebras J. M., & Tuñón M. J., (2002). Los flavonoides : propiedades y acciones antioxidantes, *Nutrición Hospitalaria*. 271–278.

Paz, M., Gúllon, P., Barroso, M. F., Carvalho, A. P., Domingues, V. F., Gomes, A. M., Becker, H., Longhinotti, E., Delerue-Matos, C. (2015). Brazilian fruit pulps as functional foods and additives: Evaluation of bioactive compounds. *Food Chemistry*. 172, 462–468.

Peng, X., Ma, J., Cheng, K. W., Jiang, Y., Chen, F., Wang, M. (2010). The effects of grape seed extract fortification on the antioxidant activity and quality attributes of bread. *Food Chemistry*. 119(1), 49–53. http://doi.org/10.1016/j.foodchem.2009.05.083

Raghavendra, S. N., Ramachandra, Swamy, S. R., Rastogi, N. K., Raghavarao, K. S. M. S., Kumar, S., Tharanathan, R. N. (2006). Grinding characteristics and hydration properties of coconut residue: A source of dietary fiber. *Journal of Food Engineering*. 72(3), 281–286. http://doi.org/10.1016/j.jfoodeng.2004.12.008

Ramírez, Z. R. M., Pérez, B. J. A. (2010). Alimentos funcionales: principios y nuevos productos. (Trillas, Ed.) México. 15-159. ISBN 978-607-17-0542-6.

Ribeiro, S. M. R., Barbosa, L. C. A., Queiroz, J. H., Kno, M. (2008). Phenolic compounds and antioxidant capacity of Brazilian mango (Mangifera indica L.) varieties. *Food Chemistry*. 110, 620–626. http://doi.org/10.1016/j.foodchem.2008.02.067

Sabanis, D., Lebesi, D., Tzia, C. (2009). Effect of dietary fibre enrichment on selected properties of gluten-free bread. *LWT-Food Science and Technology*. 42, 1380–1389.

SAGARPA. (2013a). Producción nacional de cacao en México. Retrieved from http://www.siap.gob.mx/cierre-de-la-produccion-agricola-por-estado/

SAGARPA. (2013b). Producción nacional de coco de palma en México. Retrieved from http://www.siap.gob.mx/cierre-de-la-produccion-agricola-por-estado/

Sangronis, E., Soto, M. J., Valero, Y., Buscema, I. (2014). Cascarilla de cacao venezolano como materia prima de infusiones. *Archivos Latinoamericanos de Nutrición*. 64(2), 123–130.

Saval, S. (2012). Aprovechamiento de residuos agroindustriales : pasado, presente y futuro. *BioTecnología*. 16(2), 14–46.

Shahidi, F., Ambigaipalan, P. (2015). Phenolics and polyphenolics in foods, beverages and spices: antioxidant activity and health effects - A review. *Journal of Functional Foods*. 18, 820–897. http://doi.org/10.1016/j.jff.2015.06.018

Trinidad, T. P., Mallillin, A. C., Valdez, D. H., Loyola, A. S., Maglaya, A. S., Chua, M. T. (2006). Dietary fiber from coconut flour : A functional food, Innovative. *Food Science and Emerging Technologies*. 7, 309–317. http://doi.org/10.1016/j.ifset.2004.04.003

Yepes, M. S., Montoya, N. L. J., Orozco, S. F. (2008). Valorización de residuos agroindustriales frutas en Medellín y el Sur del Valle del Aburrá, Colombia. *Revista Facultad Nacional de Agronomía, Medellín*. 4422-4431.

Zia-UI-Hag, M., Ahmad, S., Bukhari, S.A., Amarowicz, R., Ercisil, S., Jaafar, H.Z.E. (2014). Compositional studies and biological activities of some mash bean (*Vigna mungo* (L.) Hepper) cultivars commonly consumed in Pakistan. *Biological Research*. 47, 23-35.