biology and life sciences forum

MDPI

Proceeding Paper

Effect of the Type of Thermal Treatment on the Nutritional and Nutraceutical Characteristics of Pacaya Inflorescences (*Chamaedorea tepejilote* Liebm) †

Pedro Mancera-Castro ‡, Aurea Bernardino-Nicanor, José Mayolo Simitrio Juárez-Goiz, Gerardo Teniente-Martínez and Leopoldo González-Cruz *

Departamento de Ingeniería Bioquímica, Tecnológico Nacional de México – Instituto Tecnológico de Celaya. Antonio García Cubas Pte. 600, Celaya 38010, Guanajuato, México; aurea.bernardino@itcelaya.edu.mx (A.B.-N.); mayolo.juarezg@itcelaya.edu.mx (J.M.S.J.-G.); gera_tm@hotmail.com (G.T.-M.)
‡ Presenting author: pedromancera.ibq@gmail.com
* Correspondence: leopoldo.gonzalez@itcelaya.edu.mx
† Presented at the 3rd International Electronic Conference on Foods: Food, Microbiome, and Health; online, 1-15 October 2022.

Abstract: *Chamaedorea tepejilote* Liebm is a palm native to the south of Mexico and Central America. In Mexico, the male inflorescences are roasted, fried, boiled, or accompanied with other ingredients to decrease its bitter aftertaste and can be consumed by the inhabitants. However has been observed that the raw inflorescences have hypoglycemic, antitussive, and antimicrobial potentials, but the thermal treatment effect in these activities has not been studied; for this reason, this study evaluated the impact of three thermal treatments (hydrothermal (HP), steaming at elevated pressure (SEP), and microwave (MW)) on the nutritional and nutraceutical characteristics of Pacaya inflorescences, inflorescences without thermal treatment (WTT) were considered as control. In nutritional characterization, only the protein content was the fraction that increased significantly ($p < 0.05$) when thermal treatment was applied. On the other hand, all thermal treatments modified significantly ($p < 0.05$) the chlorophyll "a" content (HP reduced 0.59-fold; SEP and MW increased 0.07-0.25-fold), and chlorophyll "b" decreased. A significant ($p < 0.05$) carotenoid content increase in all thermally treated samples (between 0.80-fold and 8.73-fold) and total phenolic compounds (between 7.75-fold and 8.16-fold) than in WTT samples were observed. Microwave cooking was the only thermal treatment that significantly ($p < 0.05$) increased 0.97-fold the antioxidant activity in the DPPH radical. HP (14.11%) and SEP (18.20%) significantly ($p < 0.05$) reduced the dipeptidyl peptidase-IV enzyme inhibition concerning WTT (24.42%). These changes have been associated with partial loss, destruction, or denaturalization of cell walls' proteins, lipids, or cellulose, causing the liberating or creation of compounds with nutritional and nutraceutical activity.

Keywords: Pacaya; thermal treatment; antioxidant activity; nutraceutical characteristics

Citation: Lastname, F.; Lastname, F.; Lastname, F. Title. *Biol. Life Sci. Forum* **2022**, *2*, x. https://doi.org/10.3390/xxxxx

Academic Editor: Firstname Lastname

Published: date

Publisher's Note: MDPI stays neutral about jurisdictional claims in published maps and institutional affiliations.

1. Introduction

Modifications in the functional characteristics of raw materials affect their application in food processing, quality, acceptance, and how they are used as ingredients in formulations of other foods. Thermal treatments on foods could significantly impact nutritional, nutraceutical, and functional properties, such as solubility, water, and oil absorption, gelation, ability to create emulsions, and others; the main food components that modify these properties are the polymers such the proteins or carbohydrates because thermal treatment may transform the composition, structure, conformation, and interaction with other food components such as lipids, and polyphenols. In particular, the thermal treatments may change the carbohydrates' composition, structure, and functionality, mainly on polysaccharides such as pectins, mucilages, cellulose, hemicelluloses, and starch [1]; these modifications can be observed primarily in foods such as flowers that are becoming increasingly popular because they represent a new source of nutraceutical food, which have proteins, carbohydrates, lipids, and carotenoids, among others. Recent research has reported that edible flowers may have antioxidant, anti-inflammatory, antibacterial, antifungal, and antiviral activity [2-4]. In the south of Mexico, Central America, and northwest Colombia [5] grow the *Chamaedorea tepejilote* Liebm palm. In some Mexico regions (Veracruz, Oaxaca, and Chiapas), their male inflorescence is consumed by indigenous communities; however, due to their bitter aftertaste, inflorescences are treated with different thermal treatments, such as roasted, fried, boiled, or mixed with other ingredients such as egg and tomato sauce [6,] in addition have been reported the incorporating of Pacaya powder in frequently consumed foods such as Mexican tostadas, Mexican chorizo, and breakfast cereal [5,7]. Some authors have evaluated the *in vivo* hypoglycemic potential in normoglycemic rats [8] and antitussive and antimicrobial activities concerning nutraceutical characteristics [9] in raw inflorescences. However, the Pacaya is an underutilized plant that has not been thoroughly studied; for this reason, this work aimed to identify some nutritional and nutraceutical modifications on the inflorescences of Pacaya by the effect of thermal treatment.

2. Material and methods

2.1. Vegetal material

Male inflorescences from Tapachula, Chiapas, Mexico, were collected in February 2022. The opaque yellow inflorescences without mechanical or microbiological damage were selected; later, they were cut into cubes of approximately 1 cm and stored under vacuum in PVC bags in batches of 300 g. Batches were refrigerated for no more than 24 h before processing.

2.2. Thermal treatments

Batches (300 g) in vacuum bags were thermally treated with established conditions previously reported by Hernández-Castillo et al. [10]: hydrothermal processing (90°C in a water bath for 15 min), steaming at elevated pressure (121°C and ~124 Pa for 15 min in a pressure cooker) and microwave cooking (1500 W and operating frequency of 2450 MHz for 15 min in a microwave oven). At the end of each thermal treatment, the samples were cooled, frozen, lyophilized, ground, sieved in mesh No. 40, and stored in airtight glass jars. Inflorescences without thermal treatment were considered a control.

2.3. Nutritional characterization

The proximal analysis was carried out with methods proposed by the AOAC [11]: moisture (925.09), protein (N x 6.25, 955.04), lipids (920.39), and ash (923.03). Nitrogen-free compound's content was determined by difference. The results are expressed as a percentage for each component on a wet basis of lyophilized samples.

2.4. Nutraceutical characterization

2.4.1. Total Phenolic Compounds (TPC)

The phenolic compounds were extracted using methanol 80% v/v for 30 min under stirring; extracts were later centrifugated at 5000 rpm for 5 min. The supernatants were stored at -20°C and protected from light until used. The Folin-Ciocalteu method quantified total phenolic compounds [15], using gallic acid as the standard. Total phenolic compounds are expressed as micrograms equivalents of gallic acid per gram of sample (μg EGA/g).

2.4.2. Chlorophyll a and b content and total carotenoids

To extract these components, 400 mg of Pacaya powder were dispersed in 20 mL of acetone 80% v/v and kept under constant stirring (150 rpm) for two hours, protected from light at 4°C. Subsequently, the dispersion was centrifuged at 3000 rpm for 5 min. The content of carotenoids and chlorophyll was estimated spectrophotometrically according to equation (1) to equation (3) proposed by Lichtethaler & Wellburn [16]:

$$C_a\ (\mu g/g) = 12.21[\text{Absorbance}_{663\ nm}] - 2.81[\text{Absorbance}_{646\ nm}] \quad (1)$$

$$C_b\ (\mu g/g) = 20.13[\text{Absorbance}_{646\ nm}] - 5.03[\text{Absorbance}_{663\ nm}] \quad (2)$$

$$C_{carotenoids}\ (\mu g/g) = \frac{1000[\text{Absorbance}_{470\ nm}] - 3.27[C_a] - 104[C_b]}{229} \quad (3)$$

2.4.3. Antioxidant activity assays

The DPPH* assay [17] was used with methanol 80% v/v as a dissolvent. Results were expressed as % radical inhibition.

2.4.4. Dipeptidyl peptidase IV (DPP-IV) inhibition assay

According to Lin et al. [18], the DPP-IV inhibitory activity was tested using porcine DPP-IV enzyme and Gly-Pro-p-nitroanilide as substrate. The reaction was incubated at 37 °C for one hour, and its absorbance was measured at 405 nm. Commercial sitagliptin (0.1 mM as positive control) and samples (1 mg/mL) were diluted in distilled water and centrifugated at 5000 rpm for 5 min. Results were expressed as % DPP-IV inhibition.

2.5. Experimental design and statistical analysis

The experiments were set up on a single-factor, completely randomized design with three replicates per level of treatment. The "thermal treatment" factor was evaluated at four levels: hydrothermal, steam pressure, microwave, and without thermal treatment (as control). All data are presented as the mean ± standard deviation. A one-way analysis of variance (ANOVA) was performed, followed by a *post-hoc* Tukey-Kramer analysis to identify differences between treatments at a p-value < *0.05*. The statistical package Origin Pro, Version 2021 (OriginLab Corporation, Northampton, MA, USA) was used.

3. Results and discussion

3.1. Nutritional characterization

The Pacaya powder obtained after the microwave cooking showed a significant (p < 0.05) decrease in the moisture content than in the other treatments. The protein content in all thermal treatments increased significantly (p < 0.05), while no significant differences (p > *0.05*) in ash content were detected between all thermal treatments. Similar findings were already reported by Sun et al. [19], who associated increased protein with partial loss or destruction of other components, for example, lipids or fiber. Microwave cooking may cause the partial loss or destruction of the volatile and water-soluble fatty acids, which decrease lipids content [19].

Table 1. Proximal analysis of Pacaya inflorescences with different thermal treatments

Treatment	Moisture (%)	Protein (%)	Lipids (%)	Ash (%)	NFC* (%)
Without thermal treatment	8.67 ± 1.15 ^{a}	18.40 ± 0.79 ^{c}	9.00 ± 0.16 ^{a}	12.35 ± 0.39 ^{a}	51.58 ± 0.47 ^{a}
Hydrothermal processing	7.58 ± 0.38 ^{a}	26.82 ± 0.98 ^{ab}	9.83 ± 0.29 ^{a}	10.59 ± 0.13 ^{a}	45.18 ± 0.43 ^{b}
Steaming at elevated pressure	7.33 ± 0.58 ^{a}	27.31 ± 0.83 ^{a}	9.29 ± 0.21 ^{a}	11.15 ± 1.57 ^{a}	44.92 ± 2.74 ^{b}
Microwave cooking	3.33 ± 0.58 ^{b}	24.81 ± 0.81 ^{b}	6.33 ± 1.05 ^{b}	12.12 ± 0.86 ^{a}	53.41 ± 1.51 ^{a}

Different letters in each column indicate significant differences at p < 0.05. *Nitrogen's free compounds (NFC = 100 – moisture – protein – lipids – ash).

3.2. Nutraceutical characterization

The chlorophyll "a" and "b", total carotenoids, total phenolic compounds, and other nutraceutical characteristics after different thermal treatments are shown in Table 2. The hydrothermal thermal treatment showed a significant ($p < 0.05$) decrease in the chlorophyll "a" content than the other treatments; nonetheless, exists an increase in steaming at elevated pressure treatment and microwave cooking treatment concerning without thermal treatment. Regarding chlorophyll "b" content, all thermal treatments significantly ($p < 0.05$) reduced it. Mazzeo et al. [20] observed similar comportment of these phytochemicals by the type of thermal treatment in green beans, asparagus, and zucchini. They ascribe these results due to their conversion to pheophytins or possibly thermal degradation of chlorophylls.

All thermal treatments significantly ($p < 0.05$) increased the total carotenoid content, which could be attributed to the carotenoids on some occasions being associated with proteins and cellulose or immersed in lipid droplets, and the thermal treatment released a higher carotenoid content than in the samples without thermal treatment. Therefore, an appropriate thermal treatment (with adequate cooking conditions) could denature proteins and break down cellulose structure, releasing carotenoids by softening cell walls [21,22]. The other point is that some pigment complexes with protein may be created during thermal treatments and liberated during extraction, altering the carotenoids' quantification. Therefore, it is essential to determine the carotenoid profile to know crucial changes in these phytochemicals' content.

Also, thermal treatments significantly ($p < 0.05$) increased approximately 8-fold to 11-fold the total phenolic compounds. These changes are associated with a substantial weakening of cell walls by heat that facilities polyphenols release; another point is the effect of concentration in the food matrix after partial or total moisture evaporation increases the polyphenols concentration. Similarly, *de novo* compounds production has been reported, such as Maillard reaction products reacting with Folin-Ciocalteu reagent [23].

Microwave cooking was the only thermal treatment that significantly ($p < 0.05$) increased 0.97-fold the antioxidant activity by the DPPH method in the Pacaya after thermal treatments. Hydrothermal processing and steaming at elevated pressure were not caused substantial changes. Jiménez-Monreal et al. [24] suggested four possibilities for the increase in antioxidant activity in some cooking methods: 1) the release of high amounts of antioxidants due to thermal destruction of cell walls and subcellular compartments; 2) production of more robust radical-scavenging antioxidants by thermal-chemical reaction; 3) elimination of the oxidation capacity of antioxidant by thermal inactivation of oxidative enzymes; and 4) the formation of new compounds with antioxidant activity as a result of the Maillard reaction.

It was observed that hydrothermal processing and steaming at elevated pressure significantly ($p < 0.05$) reduced the DPP-IV enzyme inhibition. Sitagliptin (0.1 mM) as positive control showed 95% inhibition. The hypoglycemic activity of *Chamaedorea tepejilote* inflorescences has been studied. However, the mechanism of action and thermal treatment's effect is not reported. Riquett Robles et al. found that 300 mg/kg of aqueous extract administration to normoglycemic mice reduced blood glucose by 29.77% compared to the control group [8]. Plant dipeptidyl peptidase-IV inhibitors characterized and studied are phenolic compounds and protein hydrolysates (bioactive peptides) [25].

Table 2. Effect of thermal treatment on chlorophyll a and b, total carotenoids, total phenolic compounds, and other nutraceutical characteristics of Pacaya inflorescences

Treatment	**Chlorophyll a**	**Chlorophyll b**	**Carotenoids**	**TPC**	**DPPH**	**DPP-IV**
		μg/g		**μg GAE/g**	**% Inhibition**	
Without thermal treatment	27.71 ± 2.62 [a]	35.07 ± 3.95 [a]	4.28 ± 0.80 [d]	0.36 ± 0.23 [c]	5.60 ± 0.14 [b]	21.42 ± 1.04 [a]
Hydrothermal processing	16.26 ± 2.52 [b]	15.44 ± 3.42 [b]	8.11 ± 0.68 [c]	3.15 ± 0.06 [b]	5.03 ± 0.29 [bc]	14.11 ± 0.33 [c]
Steaming at elevated pressure	34.64 ± 4.77 [a]	21.15 ± 2.97 [b]	28.11 ± 1.96 [b]	3.96 ± 0.06 [a]	4.55 ± 0.05 [c]	18.20 ± 0.22 [b]
Microwave cooking	29.83 ± 0.81 [a]	17.12 ± 1.31 [b]	41.66 ± 0.44 [a]	3.30 ± 0.32 [b]	11.06 ± 0.56 [a]	20.76 ± 0.13 [a]

Different letters in each column indicate significant differences at $p < 0.05$. Abbreviations: total phenolic compounds (TPC), gallic acid equivalents (GAE), and dipeptidyl peptidase-IV (DPP-IV).

4. Conclusions

Tepejilote inflorescences are an excellent source of nutrients, mainly proteins and lipids; as well as nutraceutical compounds, such as carotenoids and chlorophyll, to mention a few, which have shown antioxidant activity and inhibit the enzyme DPP-IV (important in glucose metabolism).

However, the thermal treatments modified all the characteristics evaluated, possibly due to denaturation, partial or total loss of cell wall components, such as proteins, cellulose, and hemicellulose, which causes the release or creation of compounds with nutraceutical activity.

Author Contributions: Conceptualization, P.M.-C., A.B.-N., and L.G.-C.; methodology, P.M.-C.; software, P.M.-C.; validation, J.M.S.J.-G., and G.T.-M.; formal analysis, P.M.-C.; investigation, P.M.-C.; resources, A.B.-N., J.M.S.J.-G., and L.G.-C.; writing—original draft preparation, P.M.-C.; writing—review and editing, A.B.-N., and L.G.-C.; supervision, G.T.-M., J.M.S.J.-G., A.B.-N., and L.G.-C.; project administration, A.B.-N., and L.G.-C.; funding acquisition, A.B.-N., and L.G.-C. All authors have read and agreed to the published version of the manuscript.

Funding: This research received no external funding.

Acknowledgments: Pedro Mancera-Castro thanks CONACyT-Mexico for a doctoral fellowship (No. 734408).

Conflicts of Interest: The authors declare no conflict of interest.

References

1. Ma, Z.; Boye, J.I.; Simpson, B.K.; Prasher, S.O.; Monpetit, D.; Malcolmson, L. Thermal processing effects on the functional properties and microstructure of lentil, chickpea, and pea flours. *Food Res. Int.* **2011**, *44*, 2534-2544.
2. Mlcek, J.; Rop, O. Fresh edible flowers of ornamental plants – A new source of nutraceutical foods. *Trends Food Sci. Technol.* **2011**, *22*, 561-569.
3. Fernandes, L.; Casal, S.; Pereira, J.A.; Saraiva, J.A.; Ramalhosa, E. An overview on the market of edible flowers. *Food Rev. Int.* **2019**, *36*, 258-275.
4. Janarny, G.; Gunathilake, K.D.P.P.; Ranaweera, K.K.D.S. Nutraceutical potential of dietary phytochemicals in edible flowers – A review. *J. Food Biochem.* **2021**, *45*, 13642.
5. Palacios Pola, G.; Rodríguez García, T.; Ayvar Ramos, P.; Pantoja Enríquez, J.; Bartolón Roblero, Y.I.; Abud Archila, M. Incorporación de chaya *Cnidoscolus aconitifolius* (Mill.) I.M. Johnst. (Euphorbiaceae) y pacaya *Chamaedorea tepejilote* Liebm. (Arecaceae), en alimentos de consumo frecuente con medición de parámetros de calidad sanitaria, sensorial y nutricional. *Lacandonia* **2014**, *8*, 27-32.
6. Castillo-Mont, J.J.; Gallardo, N.R.; Johnson, D.V. The Pacaya Palm (*Chamaedorea Tepejilote*; *Arecaceae*). *Econ. Bot.* **1994**, *48*, 68-75.
7. Montejos Ramos, O.; Márquez Montes, R. Incorporación de la inflorescencia comestible de palma (Arecaceae: *Chamaedorea tepejilote* Liebm.) en un cereal para desayuno. *Lacandonia* **2012**, *6*, 111-121.
8. Riquett Robles, D.; Solórzano Carranza, E.R. Actividad hipoglucemiante de *Chamaedorea tepejilote* Liebm. (pacaya). *Rev. Cubana Plant. Med.* **2013**, *18*, 27-33.
9. Pérez, G.C.; Zavala, S.M.A.; Ventura, R.E.; Pérez, G.S.; Ponce, M.H. Evaluation of anti-tussive activity of *Chamaedorea tepejilote*. *J. Ethnopharmacol.* **2008**, *120*, 138-140.
10. Hernández-Castillo, J.B.E.; Bernardino-Nicanor, A.; Vivar-Vera, M.A.; Montañez-Soto, J.L.; Teniente-Martínez, G.; Juárez-Goiz, J.M.S.; González-Cruz, L. Modification of the protein characteristics of pacaya caused by thermal treatment: A spectroscopic, electrophoretic and morphological study. *Polymers* **2020**, *12*, 1016.
11. AOAC. Association of Official Analytical Chemists. In *Official Methods of Analysis*, 15th ed.; AOAC International: Arlington, VA, USA, 1990; pp. 993–994.
12. Bashir, K.; Swer, T.L.; Prakash, K.S.; Aggarwal, M. Physico-chemical and functional properties of gamma irradiated whole wheat flour and starch. *LWT - Food Sci. Technol.* **2017**, *76*, 131-139.
13. Chen, Y.; Zhang, B.C.; Sun, Y.H.; Zhang, J.G.; Sun, H.J.; Wei, Z.J. Physicochemical properties and adsorption of cholesterol by okra (*Abelmoschus esculentus*) powder. *Food Funct.* **2015**, *6*, 3728-3736.
14. Dong, W.; Wang, D.; Hu, R.; Long, Y.; Lv, L. Chemical composition, structural and functional properties of soluble dietary fiber obtained from coffee peel using different extraction methods. *Food Res. Int.* **2020**, *136*, 109497.
15. Singleton, V.L.; Orthofer, R.; Lamuela-Raventos, R.M. Analysis of total phenols and other oxidation substrates and antioxidants by means of Folin-Ciocalteau reagent. *Methods Enzymol.* **1999**, *299*, 152-178.

16. Lichtethaler, H.K.; Wellburn, A.R. Determinations of total carotenoids and chlorophylls a and b of leaf extracts in different solvents. *Biochem. Soc. Trans.* **1983**, *11*, 591-592.
17. Brand-Williams, W.; Cuvelier, M.E.; Berset, C. Use of a free radical method to evaluate antioxidant activity. *LWT - Food Sci. Technol.* **1995**, *28*, 25-30.
18. Lin, Y.S.; Chen, C.R.; Wu, W.H.; Wen, C.L.; Chang, C.I.; Hou, W.C. Anti-α-glucosidase and anti-dipeptidyl peptidase-IV activities of extracts and purified compounds from *Vitis thunbergii* var. *taiwaniana*. *J. Agric. Food Chem.* **2015**, *63*, 6393-6401.
19. Sun, H.; Mu, T.; Xi, L.; Song, Z. Effects of domestic cooking methods on polyphenols and antioxidant activity of sweet potato leaves. *J. Agric. Food Chem.* **2014**, *62*, 8982-8989.
20. Mazzeo, T.; Paciulli, M.; Chiavaro, E.; Visconti, A.; Fogliano, V.; Ganino, T.; Pellegrini, N. Impact of the industrial freezing process on selected vegetables-Part II. Colour and bioactive compounds. *Food Res. Int.* **2015**, *75*, 89-97.
21. Danowska-Oziewicz, M.; Narwojsz, A.; Draszanowska, A; Marat, N. The effects of cooking method on selected quality traits of broccoli and green asparagus. *Int. J. Food Sci. Technol.* **2019**, *55*, 127-135.
22. Lee, S.W.; Kim, B.K.; Han, J.A. Physical and functional properties of carrots differently cooked within the same hardness-range. *LWT - Food Sci. Technol.* **2018**, *93*, 346-353.
23. Arkoub-Djermoune, L.; Boulekbache-Makhlouf, L.; Zeghichi-Hamri, S.; Boukhalfa, F.; Madani, K. Influence of the thermal processing on the physicochemical properties and the antioxidant activity of a solanaceae vegetable: eggplant. *J. Food Qual.* **2016**, *39*, 181-191.
24. Jiménez-Monreal, A.M.; García-Diz, L.; Martínez-Tomé, M.; Mariscal, M.; Murcia, M.A. Influence of cooking methods on antioxidant activity of vegetables. *J. Food Sci.* **2009**, *74*, 97-103.
25. Turdu, G.; Gao, H.; Jiang, Y.; Kabas, M. Plant dipeptidyl peptidase-IV inhibitors as antidiabetic agents: a brief review. *Future Med. Chem.* **2018**, *10*, 1229-1239.