

Article

Microwave technology applied to natural ingredient extraction from Amazonian fruits

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Abstract: Extraction is one of the crucial steps for research and development of plant secondary metabolites, above all, in emergent and promissory plants. To date, many techniques have been developed but natural products demand more eco-commitment from the value productive chain. Microwave-assisted extraction is one of the principal techniques for extracting valuable compounds from vegetal materials, due to the versatility of microwaves. Milpesillos (*Oenocarpus mapora*), asai (*Euterpe precatoria*) and seje (*Oenocarpus bataua*), American palms from the Colombian Amazon region exhibit a high polyunsaturated fatty acid content and attractive profile for natural ingredients. MAE can be improved in order to increase extraction yield rate compared with other extraction methods such as solid liquid extraction (SLE) (-59%) or mechanical extraction with expeller press (EPE) (-50%). The MAE lead to oils with more unsaturated fatty acids combined with sustainable extraction conditions that make it friendly with the environment and competitive for production purposes. MAE has also been used in pigment extraction such as for carayuru (*Arrabidaea chica*) and achiote (*Bixa orellana*) plants and the pigment has shown higher stability than those obtained by chemical methods. Efficiency is also higher in MAE, as compared with other methods due to the short period required to obtain final products (+85%).

Keywords: Emergent species; extractive industries; fatty acids; food process technology; vegetal oils; palms; pigment extraction; plant metabolites

1. Introduction

Microwave heating is the result of the interaction between an alternating electromagnetic field and a dielectric material. The principal mechanisms involved in microwave heating food are orientation polarization and interfacial distribution. Some dielectric materials contain permanent dipoles (i.e. water, oil) that tend to be reoriented under the influence of alternating fields, causing a polarized orientation [1-2]. Heat and molecule vibration is generated by the friction of the molecules with other molecules when they are rotating in order to be oriented and maintain alignment with the alternating field of microwaves. In the interfacial distribution, any charged particle in the food experiences an alternating force at the speed of the frequency of the microwave. The net force accelerates a particle in one direction and in the opposite one (the electric field to which it is subjected is alternate). Particle collisions with adjacent particles generate heat [3]. Microwave heating has several advantages over conventional heating like the increase in the heating rate; produces desirable physicochemical effects; achieves better control and faster process control; provide purity of the final product; and improved reproducibility [4-5]. On the contrary, microwaves lacks of heat homogeneity and therefore trials to adjust the technology to the food product is desirable.

A relatively new application is their use as a method of extracting soluble products as a fluid from a wide range of materials using microwave energy. Microwave-assisted extraction (MAE) provides a technique where the compounds can be selectively removed in a short amount of time as compared with conventional methods of extraction, and consumes less power than conventional extraction processes while reducing waste substances. The absorption of electromagnetic energy during MAE varies with the food matrix and the chemical nature of the compounds that are being exposed to microwave radiation. Under the conditions of MAE, solvents are chosen for their ability to dissolve the target analyte compound and its relative transparency to microwaves (stays cooler than the irradiated matrix) [6].

The main factors affecting MAE as an open system are: nature of the solvent, contact time and radiation power. The choice of solvent for MAE is dictated by the solubility of the target analyte by the interaction between the solvent and the matrix and finally by the microwave absorbing properties of the solvent [6].

Furthermore, Amazonian biodiversity provides interesting extractable materials because of their remarkable content of bioactive compounds [7]. In order to recommend MAE technologies for extractive industries of some Amazonic fruit, a comparison is performed in the species mentioned above among MAE and other extraction techniques such as liquid-solid (LSE) or Soxhelt and mechanical extraction with an Expeller (EPE). In addition, studies were carried out on the yield and fatty acid profile of the oils obtained by MAE.

2. Experimental Section

2.1. Plant material

The species of palms of the Colombian Amazon region selected for this study were milpesillos (*Oenocarpus mapora*), asai (*Euterpe precatoria*) and seje (*Oenocarpus bataua*). Other the plant species prized for the chemical dyes with potential use in the food and cosmetic industries that were tested here were carayuru (*Arrabidaea chica*) and achiote (*Bixa orellana*). All plant material proceeded from northeast colombian Amazonia. Achiote seeds and carayuru leaves were collected in greenhouse at the department of Vaupés, and palm fruits were collected from forest and small tree nurseries.

2.2. Preliminary unit operations before extraction

The pulp for this study was obtained according to the procedure standardized by the laboratory of post-harvest and post-production of the Sinchi Institute, in the pilot plant of the ASOPROCEGUA association in San Jose del Guaviare. Milpesillos and seje pulp were dried at 130°C (Jouas[®] brand oven) for 12h and asai pulp was dried at 60 ° C for 48h, until constant weight.

The samples of achiote and carayuru were first ground and sieved before being submitted to MAE.

2.3. Palm oil extraction

The dry pulp was subjected to three extraction processes:

1. LSE Soxhlet apparatus (JSGW Brand, Ambala, Haryana, India) for 6 h, using 10 g of sample and petroleum ether as solvent. The extracted sample was filtered, the solvent recovered by rotoevaporation and finally heated to 70°C to remove all traces of the solvent.

2. Mechanical extraction with expeller press (EPE) was conducted at the Sinchi Institute using 70 g of material at different speeds of rotation of the screw (60 rpm) and jacket temperatures (60°C). The extracted material was heated for 4 h at 70°C to decrease its viscosity and allow settling of particulates, finally the supernatant oil was vacuum filtered.

3. Microwave-assisted extraction prototype equipment was built from a domestic microwave (Whirlpool Brand, WM111D00 model). An Allihn reflux unit was installed besides the 1100 W maximum potential oven (Figure 1) coupled to an extraction balloon. A 100 g sample was weighed and solvent (acetone for seje and hexane for milpesillos) was added at a ratio of 2:3 (sample/solvent). The contact time was 15 min.

Upon completion of the extraction process, vacuum filtering was used to remove debris generated during the exposure to the electromagnetic field. Subsequently a rotoevaporator (Heidolph brand) was employed for solvent's recovery. Once the oil was obtained, the oil yield was quantified on a fresh weight basis as the ratio between the mass of oil obtained and the mass of the vegetable matrix employed.

2.4. Fatty acids profile of palm oil

For the oils obtained, the profile of fatty acids was determined by gas chromatography by conversion of fatty acids into their respective fatty acid methyl esters (FAME's). These volatiles were

later quantified by gas chromatography with a flame ionization detector (GC-FID) following to standardized methodologies ISO 5509 and ISO 5508 [8-9]. As the certified reference material, this study used the mixture Supelco™ 37 Component FAME Mix, Supelco, Bellefonte, PA, Cat N. <47 885-U. The chromatographic analysis of the sample was done in a gas chromatograph (GC) 7890A (Agilent Technologies, Palo Alto, California, USA), with a flame ionization detector (FID). The column used in the analysis was HP-88 (J & W Scientific, Folsom, CA, USA) [(88%-cyanopropyl)-aryl-polysiloxane), 60 mx 0.25 mm x 0.20 µm]. The injection was performed in split mode (10:1) and injection volume of 2 µL. Detector temperature (FID) was 285°C, gas flow was flow 30 mL·min⁻¹ for Helium (carrier gas), 300 mL·min⁻¹ of air and 25 mL·min⁻¹ H₂. Oven temperature was 100°C and raised up to 240 °C in 4 min at 30C·min⁻¹. Pressure 13.45 psi and constant flow

2.4. Pigments extraction

For MAE extraction of the pigments of achiote, ethanol was used as the solvent, and for carayuru, water. A 50% maximum power of the microwave was used for 15 minutes for both extractions. Skin colour was recorded in L*C*H* (lightness, chroma, hue angle) trichromatic coordinates (D65 illuminant; 2° observer) [10].

3. Results and Discussion

LSE generally achieved better palm oil yields than the other two methods, with the highest yield (62.2%) in the milpesillos oil extraction followed by seje oil extraction (47%) (Table 1). EPE did not provide quantifiable results for asaí extraction due to problems with the design of the equipment. MAE achieved lower yields (Table 1), but with superior organoleptic oil quality and less floating and suspended material (data not shown). MAE can be less effective because of the geometry of the resonance chamber which influences the distribution of electromagnetic radiation. Dead zones are located towards the center and the ends of the cavity which caused uneven heating of the matrix and always affect oil yield [11]. In addition, these results were preliminary and the MAE equipment used is capable of improvement.

As regards the lipid profile of the palm oils obtained using the MAE technique (Table 2), total saturated (SFA) and unsaturated fatty acids (UFA) of milpesillos and seje were similar, with a slightly higher concentration of UFA in milpesillos but with a richer fatty acid profile than seje. In fact seje oil was simplified (only myristoleic, palmitic and oleic acids). Milpesillos oil contains more chemical species of SFA, notably stearic and palmitic acids (Table 2). SFAs has opposing cardioprotective effects. Palmitic has a marked atherogenic effect and stearic is considered more neutral in how it alters serum lipid levels [12]. In this vein, acai oil was significantly different from the other two palm oils, having the highest content of SFAs (Table 2), particularly palmitic acid (16.7%), but also containing the highest concentration of stearic acid (6.8%) of the three oils. The three oils have high UFA and they would be more susceptible to oxidation by factors such as atmospheric oxygen and light.

The MAE parameter of the carotenoids extraction in achiote and carayuru respectively were 11.4% and 19% yield, with around 15 or 3 min contact times. These values show promise in the use of this technique, considerably shortening the contact time, thus saving energy and material besides reducing emissions into the environment. Furthermore, the pigments obtained by MAE had intense color than

those obtained by artisanal means, indeed carayuru pigment extracted by MAE technique had a intense red color (data not shown). Consequently, potential use of MAE for pigments extraction is very promising.

4. Conclusions

MAE can be improved in order to increase extraction yield rate compared with other extraction methods such as solid liquid extraction or mechanical extraction with expeller press. MAE leads to oil ingredients from three palm fruits (asai, seje and milpesillos) that show enough good composition characteristics such as more unsaturated fatty acids contents. MAE is also capable of improving the pigment extraction of achiote and carayuru with a better color and stability, shortening the pigment extraction time. Thus MAE extraction conditions make it friendly with the environment because of its saving energy and reducing emissions inside green chemistry.

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Conflict of Interest

The authors declare no conflict of interest.

References and Notes

1. Kwon, J.H.; Bélanger, J.M.R.; Paré, J.R.J. Optimization of microwaveassisted extraction (MAP) for ginseng components by response surface methodology. *J Agr Food Chem.* **2003**, *51*(7), 1807-1810.
2. Alfaro, M.J.; Bélanger, J.M.R.; Padilla, F.C.; Paré JR. Influence of solvent, matrix dielectric properties, and applied power on the liquid-phase microwave-assisted processes (MAP(TM)) extraction of ginger (*Zingiber officinale*). *Food Res Int.* **2003**, *36*(5), 499-504.
3. Sumnu, G. A review on microwave baking of foods. *Intl. J. Food Sci. Technol.* **2001**, *36*, 117-127.
4. Solanki, H.; Prajapati, V.; Jani, G. Microwave technology- a potential tool in pharmaceutical science. *Intl. J. Pharm. Tech. Res.* **2010**, *2*(3), 1754-1761.
5. Desai, M.; Parikh, J.; Parikh, P.A. Extraction of natural products using microwaves as a heat source. *Separ. Purif. Rev.* **2010**, *39*, 1–32
6. Kravic, S.; Suturovic, Z.; Svarc-Gajic, J.; Stojanovic, Z.; Pucarevic, M. Determination of trans fatty acids in foodstuffs by gas chromatography-mass spectrometry after simultaneous microwave-assisted extraction-esterification. *J. Serb. Chem. Soc.* **2010**, *75*(6), 803-812.
7. Schultes, R.E. Amazonia as a source of new economic plants. *Econ. Bot.* **1979**, *33*, 259–266.

8. International Organization for Standardization. “ISO 5509-2000 Animal and vegetable fats and oil. Preparation of fatty acid methyl esters of fatty acids”. July 2011. <http://down.40777.cn/stardard/11/ISO%205509-000.pdf>
9. International Organization for Standardization. “ISO 5508-1990 Animal and vegetable fats and oil chromatography of methyl ester of fatty acids” July 2011. <https://law.resource.org/pub/et/ibr/et.iso.5508.1990.pdf>
10. Hernandez, M. S., Martínez, O., & Fernández-Trujillo, J. P. (2007). Behavior of araza fruit quality traits during growth, development and ripening. *Scientia Horticulturae*, 111, 220–227.
11. Mosquera, D.; Díaz, R.; Cardona, J.; Gutiérrez-Bravo, R.; Hernández, M.S. Obtención de aceites vegetales provenientes de especies Amazónicas mediante la técnica de extracción asistida por microondas (EAM). *Vitae* **2012**, 19 (Suppl. 1), 57-59.
12. Castro-Bolaños, M.; Herrera-Ramírez, C.; Lutz-Cruz, G. Composición, caracterización y potencial aterogénico de aceites, grasas y otros derivados producidos o comercializados en Costa Rica. *AMC* **2005**, 47(1), 10-15.

TABLES AND FIGURES

Figure 1. Diagram of the components of the equipment used for microwave-assisted extraction (MAE).

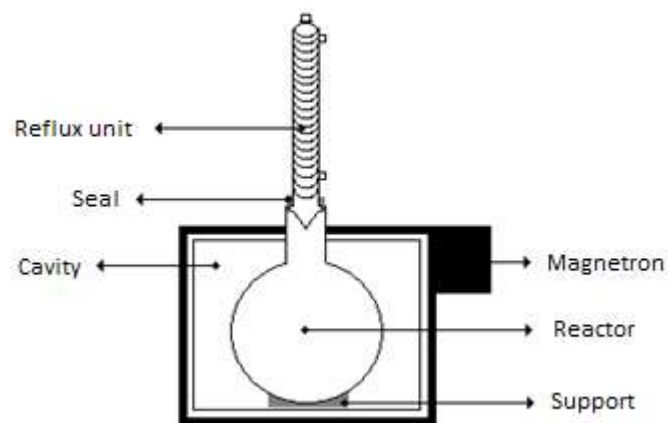


Table 1. Yield of oil extraction for milpesillos, seje and asai by three methods. LSE, liquid-solid or Soxhelt extraction. EPE, mechanical extraction with an expeller. MAE, microwave-assisted extraction with solvent.

Species	Extraction technique and yield (% w/w)		
	LSE	EPE	MAE
Asaí	2	NS	37.0
Seje	47	11	45.0 ^a
Milpesillos	62.2	48.3	37.0 ^b

NS: No significant extraction. ^a Acetone solvent. ^b Hexane solvent.

Table 2. Fatty acid profile of asaí extract, seje and milpesillos oil extracted by microwave assisted extraction (MAE).

Reported fatty acid		Relative concentration (%)		
		Asaí	Milpesos	Milpesillos
Myristic [*]	C14:0	0.1	-	0.1
Myristoleic ^{**}	C14:1	< 0.1	0.7	-
Pentadecanoic [*]	C15:0	-	-	0.3
Palmitic [*]	C16:0	16.7	12.8	12.7
Palmitoleic ^{**}	C16:1	0.1	-	0.4
Heptadecanoic [*]	C17:0	0.1	-	0.1
Stearic [*]	C18:0	6.8	-	3.6
Oleic ^{**}	C18:1n9c	70.0	86.5	78.6
Linoleic ^{**}	C18:2n6c	3.3	-	1.8
Arachidic [*]	C20:0	0.4	-	0.1
Linolenic ^{**}	C18:3n3	0.8	-	0.6
Eicosenoic ^{**}	C20:1	0.1	-	0.1
Behenic [*]	C22:0	0.1	-	<0.1
Total Saturated [*]		25.7	12.8	18.5
Total Unsaturated ^{**}		74.3	87.2	81.5