Development of Nanoemulsions with Tucumã (Astrocaryum vulgare) Fruits Oil

Abstract

Astrocaryum vulgare is a palm species which belongs to the family Arecaeae, being commonly known as tucumã and widely distributed through Amazon. Tucumã fruit is a typically Amazonian raw material, which can be used for extraction of oil with great economic importance. However, to our knowledge, tucumã remain unexplored concerning development of a nanobiotechnology product. The aim of the present study was to develop nanoemulsions containing tucumã fruits oil. Formulations prepared with different blends of sorbitan monooleate and polysorbate 80 at HLB 11.0, 12.0, 12.25, 12.5, 12.75, 13.0, 13.25, 13.5, 13.75 and 14 were considered nanoemulsions. Smallest mean droplet (156.6±0.6557 nm) was observed for nanoemulsion at HLB 13, which was considered required HLB value of this oil. On this context, this study provides valuable information concerning nanobiotechnology of Amazonian oils with great interest for pharmaceutical, cosmetics and food industries.

Keywords: HLB; Nanoemulsion; Tucumã

Introduction

Astrocaryum genus represents palm species which belongs to the family Arecaeae, being Astrocaryum vulgare commonly known as tucumã and distributed through North (Amapá, Pará, Tocantins), Northeast (Maranhão) and Central-West (Mato-Grosso) regions of Brazil [1-3]. It is also found through other countries, such as Bolivia, Colombia, French Guiana, Suriname, Venezuela and Trinidad and Tobago [4]. Several biological activities, including anti-inflammatory [5,6], antibacterial and antifungal [7] properties have been reported for Astrocaryum species. Their fruits are important sources of minerals, including potassium, calcium, selenium and present great amounts of fatty acids [8], being indicated for the treatment of skin and eye diseases [9]. Tocopherols, phytosterols, quercetin, rutin, gallic and caffeic acids have also been previously described as chemical constituents of tucumã [5,7,10, 11]. Due to its high nutritional value and pleasant taste, tucumã fruits are widely consumed by local population as food, being used in natura and to produce ice cream, dessert, cakes and sandwiches [12,13]. Tucumã fruit is a typically Amazonian raw material, which can be used for extraction of an oil with great economic importance [1,14]. Oleic acid is a fatty acid constituent of this oil, however, carotenoids develop a main role as bioactive substances, being b-carotene the major substance. This substance is a pro-vitamin A and found in tucumã in higher concentrations than in carrot [10,13].

Nanotechnology involves manipulation of materials and structures in a nanometric scale, being considered a promising tool to develop innovative pharmaceutical, cosmetics and food products [15]. Concerning food nanotechnology, studies have focused mainly to food packaging [16] or development of a wide range of nano formulations [17-19]. On this context, nanoemulsions appear as promising food products [20]. They have small droplets ranging from 30-300 nm [21], which are associated to kinetic stability of these systems, being also associated to increased absorption and potential bioavailability of substances [22]. Despite great potential of tucumã, to our knowledge, its fruits remain unexplored concerning development of a nanobiotechnology product. The aim of the present study was to develop nanoemulsions containing tucumã fruits oil.

Materials and Methods

Chemicals

Sorbitan olate (HLB: 4.3) and Polysorbate 80 (HLB: 15) were purchased from Praid Produtos Químicos Ltda (São Paulo, Brazil). Astrocaryum vulgare ripe fruits were harvested in Cayenne (French Guyane) and identified by Dr. Didier Bereau.

Tucumã oil extraction

30 g of fresh pulp of tucumã were extracted with 300 mL of cyclohexane using a Soxhlet apparatus for 2 hours. Further removal of organic solvent was performed using a rotary evaporator under 35 °C. Tucumã oil was stored in amber glass flask under 4°C until it was used for nano emulsion preparation.

Emulsification method

Emulsions were prepared by phase inversion technique with some modifications [24]. Required amounts of both emulsifiers were dissolved in tucumã oil and heated at 65±5°C. Aqueous phase was separately heated at 65±5°C. When both phases reached the same temperature, aqueous phase was continuously added through the oil phase. The mixture was submitted to magnetic agitation for 10 min, furnishing a primarily emulsion. Final homogenization was achieved using a T25 Ultra-Turrax homogenizer (Ika-Werke, Staufen, Germany) equipped with a 25 N-18 G disperser for 5 min (8000 rpm).
Characterization of emulsions

Formulations were characterized after 1 and 7 days of manipulation. Macrophotocanal aspects (appearance, translucence, opacity, bluish reflect, phase separation, creaming and sedimentation) were examined. Polidispersity and mean droplet size were determined by photon correlation spectroscopy using a Zetasizer 5000 (Malvern Instruments, Malvern, UK). Each emulsion was diluted using ultra-pure Milli-Q water (1:25). Measures were performed in triplicate and average droplet size was expressed as the mean diameter.

Results and Discussion

Studies focusing on HLB determination of oils are very important if development of emulsions is desired and is considered a valuable tool during development stage [25], including for nanoemulsions. This can be obtained by calculation the HLB value of a single surfactant or a blend of surfactants which induce formation of the most stable emulsion, among a set of formulations with different surfactant ratios in a wide range of HLB values [26]. On this context, several emulsions were prepared using tucumã fruits oil using a pair of lipophilic (sorbitan monooleate) and hydrophilic (polysorbate 80) surfactants. Set of formulations was prepared with HLB values ranging from 4.3 (HLB of sorbitan monooleate) to 15 (HLB of polysorbate 80). Emulsions prepared with HLB of 4.3, 5, 6, 7, 8 and 9 presented milky aspects, besides monodispersity and mean droplet size below 300 nm after one day of preparation, however, this size increased to 312.6±5.609 nm after seven days of storage. Polidispersity also increased, revealing a polimodal distribution. Coalescence of the droplets may be attributed to these effects, being associated to the inability of surfactant to absorb to the interface [28]. It was observed a decrease in the mean droplet size of formulations prepared with surfactants at HLB values of 11, 12 and 13. They ranged from 305.6±5.782 (HLB 11), 233.4±3.032 nm (HLB 12) and 198.9±1.601 (HLB 13) after 1 day of preparation to 211.5±6.825 (HLB 11), 190.1±7.736 (HLB 12) and 156.6±6.6557 (HLB 13) after 7 days of preparation. Utilization of volatile organic solvent may be associated to this phenomenon, considering that evaporation of residual solvent during storage may decrease droplet diameter [20]. Mean droplet size for formulation with HLB of 14 increased (219.8±11.20 nm after 1 day and 236.5±7.317 nm after 7 days). Considering that polidispersity did not present significant variation, this increase may be associated to destruction and regeneration of the micelles [29].

Additional formulations were prepared with HLB values close to 13, which presented the smallest droplet size (156.6±6.6557 nm) and polidispersity (0.240±0.006) after seven days of storage. Mean droplet size analysis revealed that these formulations presented values between 300-200 nm after 7 days of preparation, being also characterized as nanoemulsions. Mean droplet size and polidispersity are presented in Table 1.

Table 1: Mean droplet size and polidispersity of emulsions prepared during required HLB determination of tucumã fruits oil.

| HLB  | Mean diameter (nm) ± SD 1 day | Polidispersity ± SD 1 day | Mean diameter (nm) ± SD 7 days | Polidispersity (nm) ± SD 7 days |
|------|-------------------------------|---------------------------|-------------------------------|--------------------------------|
| 10   | 282.9±8.266                   | 0.581±0.079               | 312.6±5.609                   | 0.728±0.026                    |
| 11   | 305.6±5.782                   | 0.685±0.047               | 211.5±6.825                   | 0.406±0.013                    |
| 12   | 233.4±3.032                   | 0.443±0.009               | 190.1±7.736                   | 0.388±0.010                    |
| 12.25| 197.8±4.1158                  | 0.364±0.016               | 235.4±6.525                   | 0.443±0.025                    |
| 12.50| 199.3±8.057                   | 0.586±0.046               | 220.6±3.479                   | 0.390±0.013                    |
| 12.75| 301.3±54.82                   | 0.726±0.104               | 219.8±2.401                   | 0.41±0.008                     |
| 13   | 198.9±1.601                   | 0.377±0.024               | 156.6±6.6557                  | 0.240±0.006                    |
| 13.25| 220.3±7.481                   | 0.522±0.090               | 220.3±7.481                   | 0.52±0.090                     |
| 13.5 | 176.0±8.107                   | 0.426±0.020               | 264.1±2.991                   | 0.67±0.008                     |
| 13.75| 183±3.620                     | 0.433±0.044               | 250.5±11.76                   | 0.74±0.036                     |
| 14   | 219.8±11.20                   | 0.662±0.033               | 236.5±7.317                   | 0.665±0.073                    |

HLB: Balance hydrophilic-lipophilic; nm: nanometer

Citation: Silva CNS, Hyacienth DC, Ferreira AM. Vilhena JCE. Florentino AC, et al. (2015) Development of Nanoemulsions with Tucumã (Astrocaryum vulgare) Fruits Oil. J Nanomed Res 2(2): 00024. DOI: 10.15406/jnmr.2015.02.00024
Development of Nanoemulsions with Tucumã (Astrocaryum vulgare) Fruits Oil

Figure 1: Mean droplet size and polydispersity of nanoemulsions prepared with 5% (w/w) of tucuma oil, 5% (w/w) of surfactants (HLB 13) and 90% (w/w) of water. Day 1: Mean droplet - 198.9±1.60 nm; polydispersity – 0.377±0.024. Day 7: Mean droplet – 156.6±0.6557 nm; polydispersity – 0.240±0.006.

On the present study, formulations at HLB 11.0, 12.0, 12.25, 12.5, 12.75, 13.25, 13.5, 13.75 and 14 were considered nanoemulsions. These are a special type of nano formulation with mean droplet ranging from 30-300 nm, being characterized by the dispersion of two immiscible liquids and presence of one or more surfactants [21]. They are also called mini emulsions [31] and small droplets are associated to kinetic stability of these systems, being also associated to increased absorption and potential bioavailability of substances [22]. Development of nanoemulsions as enhancers of chemical stability of natural products is considered promising, allowing, protection of substances from degradation, including oxidation [32]. Moreover, nanoemulsions prepared with carotenoids have been considered potential food products and are in the spotlight of nanobiotechnology research [20].

Conclusion

The present study allowed achievement of nanoemulsions using different blends of non-ionic surfactants. Moreover, considering the lowest mean droplet (156.6±0.6557 nm) observed after seven days of storage, it was possible to determine required HLB value of this oil (HLB 13). On this context, this study provides valuable information concerning nanobiotechnology of Amazonian oils with great interest for pharmaceutical, cosmetics and food industries.

Acknowledgement

Authors would like to thank CNPQ, Guyamazon I and FAPEAP for their financial support.

References

1. Schroth G, Mota MSS, Lopes R, Freitas AF (2004) Extractive use, management and in situ domestication of a weedy palm, Astrocaryum tucuma, in the central Amazon. Forest Ecology and Management 202(1-3): 161-179.
2. Filho OCS, Sagrillo MR, Garcia LFM, Machado AK, Cadona F, et al. (2013) The In Vitro Genotoxic Effect of Tucuma (Astrocaryum aculeatum), an Amazonian Fruit Rich in Carotenoids. J Med Food 16(11): 1013-1021.
3. Leitman P, Soares K, Henderson A, Noblick L, Martins RC (2015) Aracaceae in Lista de Espécies da Flora do Brasil. Jardim Botânico do Rio de Janeiro.
4. Meyer, Wilhelm GF (2014) Astrocaryum Aculeatum Tropicos® Missouri Botanical Garden.
5. Bony E, Boudard F, Brat P, Dussossoy E, Portet K, et al. (2012) Awara (Astrocaryum vulgare M.) pulp oil: Chemical characterization, and anti-inflammatory properties in a mice model of endotoxic shock and a rat model of pulmonary inflammation. Fitoerapia 83(1): 33-43.
6. Bony E, Boudard F, Brat P, Dussossoy E, Portet K, et al. (2012) Chemical Composition and Anti-inflammatory Properties of the Unsaponifiable Fraction from Awara (Astrocaryum vulgare M.) Pulp Oil in Activated J774 Macrophages and in a Mice Model of Endotoxic Shock. Plant Foods Hum Nutr 67(4): 384-392.
7. Jobim ML, Santos RCV, Alves CFS, Oliveira RM, Mostardeiro CP, et al. (2014) Antimicrobial activity of Amazon Astrocaryum aculeatum extracts and its association to oxidative metabolism. Microbiol Res 169(4): 314-323.
8. Yuyama LKO, Aguiar JPL, Teixeira AP, Lopes TM, Yuyama K, et al. (2005) Polpa e casca de Tucumã (Astrocaryum aculeatum) quais os constituintes nutricionais? Nutr 332.
9. Kahn F (2008) The genus Astrocaryum (Arecaceae). El género Astrocaryum (Arecaceae). Rev Peru Biol 15(1): 31-48.
10. Rosso VW, Mercadante AZ (2007) Identification and Quantification of Carotenoids, By HPLC-PDA-MS/MS, from Amazonian Fruits. J Agric Food Chem 55(13): 5062-5072.
11. Gonçalves AESS, Lajolo FM, Genovese MI (2010) Chemical composition and antioxidant/antidiabetic potential of Brazilian native fruits and
commercial frozen pulps. J Agric Food Chem 58(8): 666-4674.
12. Lim CS, Berruti FM, Palmisano R, Berruti F, Briers C, et al. (2013) Fast pyrolysis of Amazon tucumã (Astrocaryum aculeatum) seeds in a bubbling fluidized bed reactor. Journal of Analytical and Applied Pyrolysis 99: 23-31.
13. Yuyama LKO, Maeda RN, Pantoja L, Aguiar JPL, Marinho HA, et al. (2008) Processamento e avaliação da vida-de-prateleira do tucumã (Astrocaryum aculeatum) desidratado e pulverizado. Gên. Tecnol Aliment 28(2): 408-412.
14. Barbosa BS, Koonen HJF, Barreto AC, Silva JD, Figlioulro R, et al. (2009) Aproveitamento do Òleo das Amêndoas de Tucumã do Amazonas na Produção de Biodiesel. Acta Amazônica 39(2): 371-376.
15. Brumfiel G (2006) Consumer products leap aboard the nano bandwagon. Nature 440(7082): 262.
16. Duncan TD (2011) Applications of nanotechnology in food packaging and food safety: Barrier materials, antimicrobials and sensors. Journal of Colloid and Interface Science 363(1): 1-24.
17. Gomes LMM, Petito N, Costa Vg, Falcão Dq, Amújo Kgl (2014) Inclusion complexes of red bell pepper pigments with b-cyclodextrin: Preparation, characterisation and application as natural colorant in yogurt. Food Chem 148: 428-436.
18. Surassmo S, Min S, Bejrapha P, Choi M (2010) Effects of surfactants on the physical properties of capisicum oleoresin-loaded nano-capsules formulated through the emulsion–diffusion method. Food Research International 43(1): 8-17.
19. Leong W, Lai O, Long K, Man YBC, Misran M, et al. (2011) Preparation and characterisation of water-soluble phytosterol nanodispersions. Food Chemistry 129(1): 77-83.
20. Silva HD, Cerqueira MA, Souza BWS, Ribeiro C, Ardes MC, et al. (2011) Nanoemulsions of β-carotene using a high-energy emulsification-evaporation technique. Journal of Food Engineering 102(2): 130-135.
21. Zhang Y, Gao J, Zheng H, Zhang R, Han Y (2011) The preparation of 3,5-dihydroxy-4-isopropylthiobene nanoemulsion and in vitro release International ln t] Nanomedicine 6: 649-657.
22. Solans C, Izquierdo P, Nolla L, Azemar N, García-Celma MJ (2005) Nano-emulsions. Current Opinion in Colloid & Interface Science 10(3-4): 102-110.
23. Fernandes CP, Mascarenhas MP, Zibetti FM, Lima RG, Oliveira RPPE, et al. (2013) HLB value, an important parameter for the development of essential oil phytopharmaceuticals. Revista Brasileira de Farmacognosia 23(1): 108-114.
24. Costa IC, Rodrigues RE, Almeida FR, Favacho HA, Falcão DQ, et al. (2014) Development of Jojoba Oil (Simmondsia chinesis (Link) CK Schneid.) Based Nanoemulsions. Latin American Journal of Pharmacy 33 (3): 459-63.
25. Schmidt T, Dobler D, Guilda AC, Paulus N, Runkel F (2010) Multiple W/O/W emulsions-Using the required HLB for emulsifier evaluation. Colloids and Surfaces A: Physicochem Eng Aspects 372(1-3): 48-54.
26. Rodríguez-Rojo S, Varona S, Núñez M, Cocero MJ (2012) Characterization of rosemary essential oil for biodegradable emulsions. Industrial Crops and Products 37(1): 137-140.
27. Tadros T, Izquierdo P, Esquena J, Solans C (2004) Formation and stability of nano-emulsions. Adv Colloid Interface Sci 108-109: 303-318.
28. Kentish S, Wooster TJ, Ashokkumar S, Balachandran S, Mawson R, et al. (2008) The use of ultrasounds for nanoemulsion preparation. Innovative Food Science and Emerging Technologies 9(2): 170-175.
29. Patist A, Kanicky JR, Shukla PK, Shah DO (2002) Importance of Micellar Kinetics in Relation to Technological Processes. J Colloid Interface Sci 245(1): 1-15.
30. Fernandes CP, Almeida FB, Silveira AN, Gonzalez MS, Mello CB (2014) Development of an insecticidal nano emulsion with Manilkara subsericea (Sapotaceae) extract. J Nanobiotechnology 12: 22.
31. Rodrigo-Carmona A, Esquena J, González C, Solans C (2009) Studies of the relation between phase behavior and emulsification methods with nanoemulsion formation. Progr Colloid Polym Sci 115: 36-39.
32. Dias DO, Colombo M, Kolmann RG, Souza TP, Bassani VL, et al. (2012) Optimization of headspace solid phase microextraction for analysis of β-caryophyllene in a nanoemulsion dosage form prepared with copaiba (Copaifera multijuga Hayne) oil. Anal Chim Acta 721: 79-84.