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A novel method of cinnamon oil nanocapsulation in core-shell chitosan-alginate by freeze dryer

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ABSTRACT: The objective of this work was to characterize the cinnamon oil nanocapsule that contained alginate-chitosan as coating agents. In this work, cinamonn oil loaded chitosan nanoparticles (CS-alginate NP-cinamonn) are prepared by a two-step process including oil/water emulsion and ionic gelation. In this study, cinamon as a core material was nanoencapsulated with chitosan alginate at a ratio of 1:4 (core: wall). Fourier transform–infrared analysis revealed potential interactions among the constituents in the composite NPs. Scanning electron microscopic analysis showed that the particles were nearly spherical in shape with an average size of 100±20 nm. Encapsulation efficiency (%) of cinamonn in composite NPs showed considerable increase over ALG-CS NPs with tween 80. Results showed that emulation drop let size by this method were 40.2 ± 2.4 nm. TEM, it was indicated that particles size varies from the range of 50nm.

Keywords: *Alginate-Chitosan, Cinamon, Freeze dryer, Nanocapsulation, Ultrasound*

INTRODUCTION

Nanoencapsulation has been proposed as one such technology that has great potential to solving this problem. It is a branch of nanotechnology that has received a lot of attention by researchers recently especially in the pharmaceutical and biotechnology field. There are already approved products from such studies for clinical use in diagnostics, drug delivery, medical devices and imaging (Jafari, *et al.*, 2008). Nano capsulation within a biocompatible material has the potential to increase aqueous solubility of the oils, thereby improving their applicability as antimicrobials onto fresh produce (Sumita, *et al.*, 2003). They protect these molecules from environmental factors such as pH, oxygen, light etc., serving as barrier between the molecule and the environment. It also stabilizes volatile molecules, shielding them from oxidative degradation, evaporation and photo-degradation (Jain, 2003; Keawchaoon & Yoksan, 2011). Essential oils are susceptible to high temperatures, oxidation, UV light, and humidity. Nanoencapsulation may change essential oils into powder, protect the sensitive core material and reduce the

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amount of flavor which lost during storage. The coating material can be customized to determine the rate of delivery, embedded fragrances for branded perfumed clothing, Food additions, Increasing shelf life and stability of products. As the core material for different Nanocapsules may vary greatly in size, shape and composition, the encapsulated particle can be have an appearance that ranges from having regular, uniform shape through to being jagged and irregular (Risch, 1995; Becher, 2001; McClements, 2005; Izquierdo, *et al.*, 2002).

In present study alginate-chitosan was used as wall material. Cinnamon bark oil (*Cinnamomumverum*) is derived from the plant of the species name *Laurus-cinnamomum* and belongs to the Lauraceae botanical family. Native to parts of South Asia, today cinnamon plants are grown across different nations throughout Asia and shipped around the world in the form of cinnamon essential oil. Its' believed that today over 100 varieties of cinnamon are grown worldwide, but two types are definitely the most popular., the oil itself especially has strong antiphrastic, anti-inflammatory, antiplatelet and antiviral properties, which makes it extremely useful for enhancing immunity (Powers, 2016; Ghasemi & Abbasi, 2014; Sarode, *et al.*, 2016; Garcia-Amezquita, *et al.*, 2016).

MATERIALS AND METHODS

Materials

In this study, cinnamon oil (Merk, Germany) was used as the core material. The wall material was chitosan-alginate (Merk, Germany). N-Hexane, Isopropanol and Tween 80 were purchased from Merck Company (Germany). Distilled water was used for the preparation of all solutions. All general chemicals used in this study were of analytical grade.

Preparation of emulsions

Preparation of chitosan-alginate

20% of wall materials were dissolved in distilled water by magnetic stirring at 60 °C. They were kept overnight in ambient temperature in order to warrant a full saturation of wall materials. Cinnamon oil the ratio of 1:4 (core: wall) and 1% of Tween80 were added to emulsions. After that, they were stirred by magnetic stirring for pre-emulsion preparation.

Ultrasonication

An Ultrasonic Liquid Processor (Model S-4000-010, USA) was used in this study for transforming pre-emulsion to Nano-emulsion that equipped with an ultrasound probe with 4.8 mm in diameter. It was operated at 24 KHz with 600 W high-intensity for 130 s.

Emulsion droplet size analysis

The nanoemulsions were investigated for their particle size and particle size distribution by dynamic light scattering (DLS) using a Zetasizer Nano ZS ((Model PMX200C, Germany). at 25 °C. Nano-emulsions were diluted by distilled water at a ratio of 1:40.

Freeze draying

The emulsions were immediately dried by pilot-plant freeze drier (Model Alpha-12LD plus, Germany). The Freezer drier was operated at -120 °C for 36h. The nozzle air pressure was 6 bar. Ultimately, dried powders were collected and stored at 4 °C for further analysis.

Encapsulation efficiency analysis

One of the important parameters for encapsulated powders is the encapsulation efficiency (EE) during the process. By definition, it is the amount of core material encapsulated inside the powder particles [1]. Encapsulation efficiency was calculated according to the following formula:

Fourier Transform Infrared Spectroscopy (FTIR)

FTIR analysis showed the existence of reactive groups on the surfaces of nanocapsulation cinamonn oil.

$$EE = \frac{\text{Total oil content} - \text{surface oil content} \times 100}{\text{Total oil content}}$$

Scanning electron microscopy (SEM) of encapsulated powder

Microstructural properties of the encapsulated powder were evaluated by scanning electron microscopy (Model HHS-2R, Japan). Powders were placed on SEM stubs using a two-sided adhesive tape. The sam-

Table 1. The mean particle sizes as determined by DLSs.

Encapsulation efficiency (%)	Emulsion size(nm)	pH After adding cinnamon oil	pH Before adding cinnamon oil
98.33 ± 0.03	40± 0.2	6.82 ± 0.03	6.43 ± 0.06

ples were coated with gold using Auto sputter coater (ModelHHS-2R, Japan) and samples analyzed at voltage of 15 kV. The images were obtained with instrument's software installed on a PC connected to the system.

Transmission electron microscopy (TEM) of encapsulated powder

In order to study of Nano-capsules morphology, the samples must be examined in a vacuum, it is necessary to separate the samples water with acetone or alcohol; the samples were prepared by a sharp object in the form of very fine powder. The samples were studied by TEM, according to the results Nano-capsules range was between 40-60 nm.

RESULTS AND DISCUSSION

Dynamic light scattering (DLS)

In case of the nanoemulsions, the mean particle sizes as determined by DLS, the results are shown in the Table 1 below. Based on the results emulsion droplet size was from 30-40 nm.

FT-IR analysis

FT-IR spectra were analyzed to characterize the potential interactions in the nanostructure. FT-IR spectra of Alg-CS and nanocapsulation of Cinamonn are shown in Fig. 2. This Figure indicates the spectrum of ALG the broad band at 3426.3 cm^{-1} corresponds to amine groups; the peaks near 1632 cm^{-1} and 1465 cm^{-1} were caused by symmetric and asymmetric stretching vibrations of COO– groups, respectively. The bands around 1109 cm^{-1} (C-O-C stretching) are attributed to its saccharide structure. In Fig. 2b, nanocapsulation of Cinamonn, the bands at 1623 cm^{-1}, 1465 cm^{-1}, and 1086 cm^{-1} correspondingly shifted to 1637 cm^{-1}, 1423 cm^{-1}, 1034 cm^{-1}.

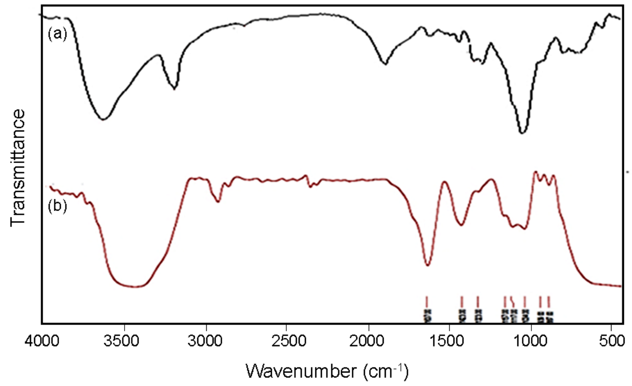

Fig. 2a,b. FTIR spectra of ALG-CS and nanocapsulation of ALG-CS cinnamon oil.

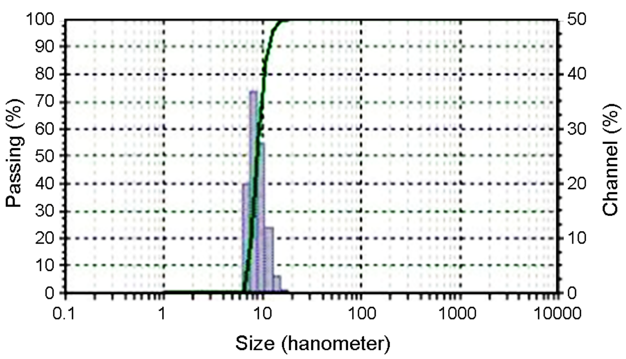

Fig. 1. distribution of emulsion droplet size of nanocapsulation of cinnamon oil.

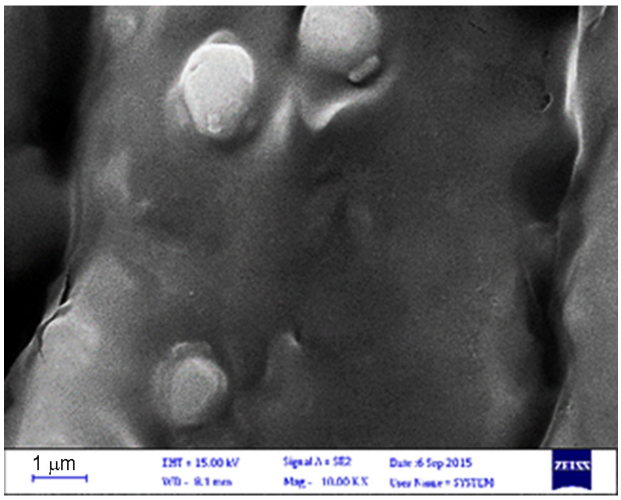

Fig. 3. Scanning electron microscopic photographs of Cinnamon oil encapsulated powder.

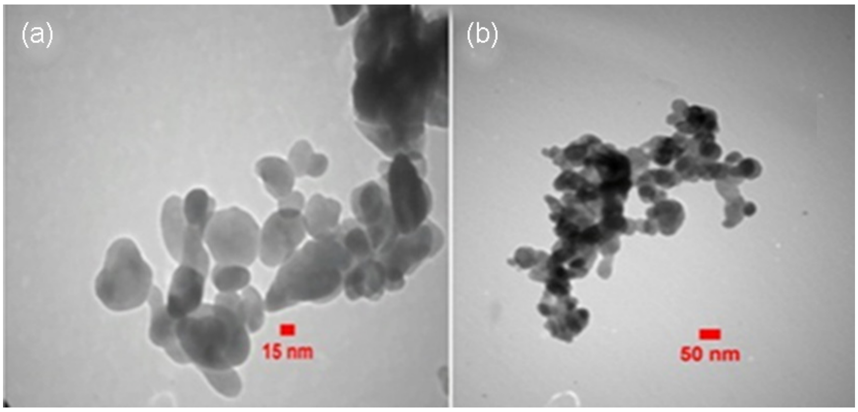

Fig. 4a, b. Transmission electron microscopic photographs of Cinnamon oil encapsulated powder.

Scanning electron microscopy (SEM)

Based on SEM photographs obtained capsules diameter were in the nanometer range, which indicates that the core (Cinnamon oil) covered completely by chitosan-alginate (cover). The shelf life of this oil increased.

CONCLUTIONS

In this work, Cinnamon oil was Nano-encapsulated by freeze dryer very well and Nano-emulsions droplet sizes were in the range of 40 nm. Figures of scanning electron microscopy and transmission have shown that Nano-size capsules were 50 Up to 60 Nanometers, shows that Nano-encapsulation. The technique used in this research was conducted successfully. The encapsulation efficiency was 98% above that shows Cinnamon oil well covered by chitosan-alginate. The purposes of the Nano-capsules productions have longer stability that it would prevent them from settling and also has resistance against chemical degradation can be seen from this product in the pharmaceutical and food that we the product stability is used. The encapsulation into nanoemulsion-based delivery systems of cinnamon oils was investigated as a method to improve the safety and quality of foods through the addition of natural preservatives.

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