RESEARCH ARTICLE

Synthesis and Characterization of Orotic Acid Loaded Chitosan Inclusion Complex

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ABSTRACT

The current study aims to improve drug release properties of orotic acid loaded with chitosan inclusion complex (OA/CS). The OA/CS inclusion complex was synthesized using the freeze-drying technique. The characterization of inclusion OA/CS was carried out using fourier transform infrared spectroscopy (FTIR), X-ray diffractometry (XRD), differential scanning calorimetry (DSC), zeta sizer, and transmission electron microscopy (TEM). Furthermore, the size of OA/CS ranged between 58 nm and 200 nm, and the zeta potential was 30 mV. Thus, this study indicates that OA/CS has a promising future to develop a carrier for drug delivery systems further.

Keywords*:* Chitosan, Drug delivery, Inclusion complex, Orotic acid, Synthesis.

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INTRODUCTION

The approach of drug delivery systems emphasizes the use of inclusion complex as a vehicle, offering the optional property of delivering drugs as a single dose rather than in multiple doses. This allows the application for the inclusion of complex use by implementing natural polymers as coated materials. Polymeric inclusion complex has been used frequently as a drug delivery system due to various advantages like a controlled release, non-toxic properties, and high bioavailability.[1-2] Chitosan is defined as a natural biodegradable and non-toxic polysaccharide used as an encapsulating agent in the pharmaceutical industry.[3] The current study focused on the synthesis and characterization of orotic acid loaded chitosan (OA/CS) inclusion complex using FTIR, XRD, DSC, TEM, and size distribution and zeta potential.

EXPERIMENTAL

Synthesize of Orotic Acid Loaded Chitosan inclusion complex

This requires preparing OA solution in CS at 1: 1 molar ratio; 0.5 g OA was dissolved in 50 mL under stirring, then mixed with 0.85g of CH dissolved in 100 mL of deionized water. They were then mixed using a high-pressure homogenizer for seven cycles and freeze-dried at -80°C.

Characterization of Orotic Acid Loaded Chitosan Inclusion Complex

The FTIR analysis was recorded over the spectral range of 4000-400 cm^{-1}. Powder X-ray diffraction patterns of OA, CS, and OA/CS inclusion complex were recorded using CuK_{α} incident beam, $\lambda = 1.5406$ A°, and voltage of 30Kv. Analysis of samples was performed at $2\theta = 20°–60°$ and scan speed of 2° per minute. The measurement of Differential scanning calorimetry was performed at a scanning rate of 10°C/min over the range of 25-1000°C and nitrogen flow rates of 50 mL/min. The Zeta potential and size of OA/CS were characterized using a zeta sizer with dynamic size. Transmission electron microscopy was used to determine the homogeneity and morphology of OA/CS.

In vitro drug release studies

The release was performed at room temperature by adding about 8 mg of OA/CS into 25 mL of PBS at pH 4.8 and 7.4. In order to compare the release rate of OA from OA/CS with that

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from a physical mixture, 0.6 g of a physical mixture containing OA (0.3g) and CS (0.3) was used to determine the release as described above.

RESULTS AND DISCUSSION

Figure1 shows the FTIR spectrum of the OA/CS inclusion complex; pure compounds as well as the physical mixture. The peaks obtained in orotic acid at 3470 cm^{-1} was due to the stretching modes of N-H bonds, and O-H stretched at 3123cm^{-1} (Figure 1). The band recorded at 2379cm^{-1} was due to C=C stretching vibration, while the band at 1343cm^{-1} was due C-H bending. A band due to C-N stretching was recorded at 1429 cm^{-1}. Band at 1667 cm^{-1} is attributed to C = O stretching. The spectrum of chitosan (CS) shows a strong band at 3441.12 cm^{-1} corresponded to –N-H– of amine and C = O stretching at 2920.38 cm^{-1} due to amide group. C-O-H and O-H bands were represented by stretching at 1094 cm^{-1} and 3726.89 cm^{-1}. The peaks of orotic acid have almost vanished in the OA/CS spectrum due to the restriction of OA vibration upon loading in CS cavity.

The XRD technique was carried out to investigate the complexation occurred between of orotic acid and gum arabic (Figure 2). Most of the peaks of OA were revealed in the physical mixture pattern by reduction of intensity. However, the diffraction peaks of the OA/CS are completely diffused due to the amorphous state and the lack of crystallinity of inclusion complex,[4-5] thereby explaining the formation of a new state.

The endothermic peak of OA vanished completely in the DSC pattern of OA/CS. These marked changes are indicative of the amorphous phase of solid dispersion, as well OA encapsulation into the nanocavity of CS. Furthermore, the endothermic peak of OA has appeared in the pattern of physical mixture associated with broadening and marked reduction of less intensity (Figure 3). The appearance of such a broadening peak may demonstrate the fusion between OA and CS or the masking of OA melting endotherm.[6-8]

The OA/CS inclusion complex prepared exhibited particle sizes ranged from 58 to 200 nm (Figure 4). In addition, TEM showed that OA/CS sizes were ranged from 20 to 45 nm as (Figure 5).

After 40 hours at pH 4.8, OA/CS exhibited 65% of release, while 45% of orotic acid was released at pH 7.4. The fast release of orotic acid from the chitosan inclusion complex is suggestive of the lack of interactions between the different elements of the physical mixture (Figure 6).

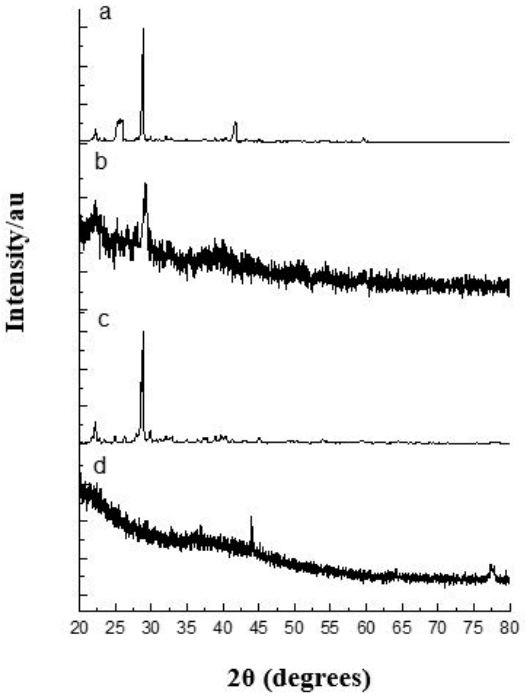

Figure 2: X-ray diffractograms of (a) OA, (b) CS, (c) Physical mixture, (d) OA/CS.

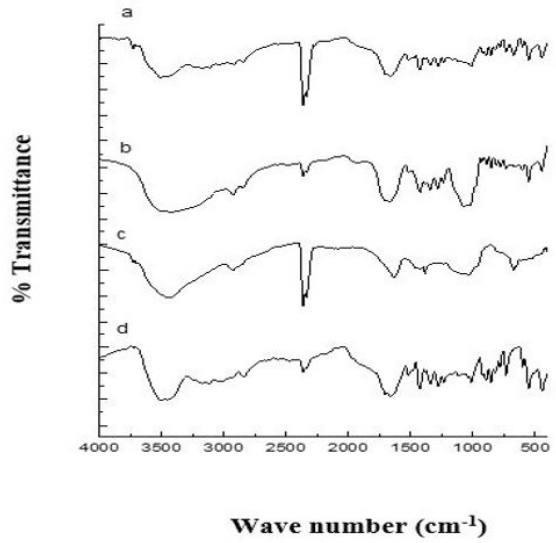

Figure 1: Fourier transform infrared spectra of (a) OA, (b) CS, (c) Physical mixture, (d) OA/CS.

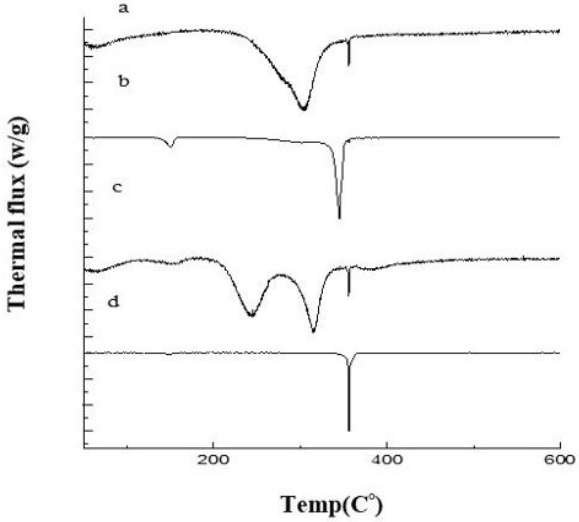

Figure 3: DSC patterns: (a) OA, (b) CS, (c) physical mixture, (d) OA/CS.

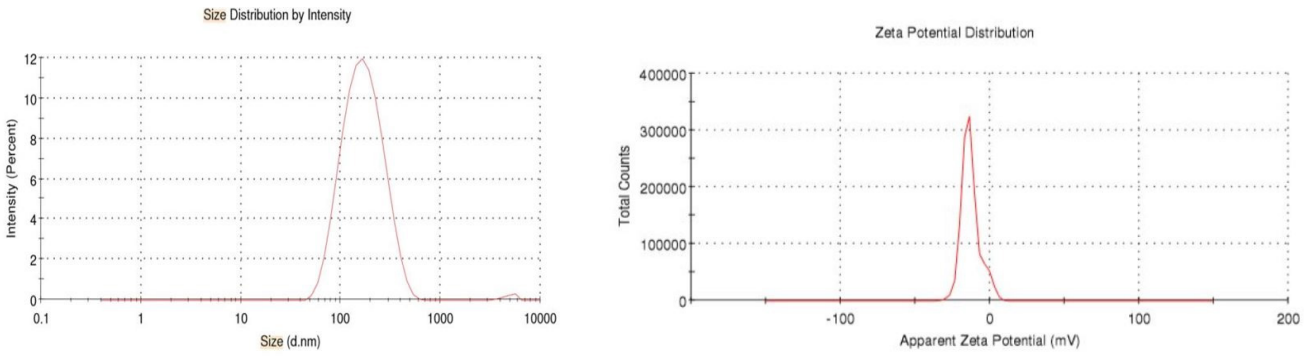

Figure 4: Size distribution and zeta potential of OA/CS inclusion complex

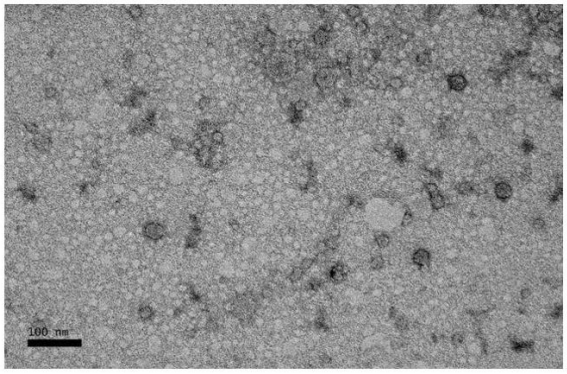

Figure 5: Transmission electron microscopy (TEM) micrograph of OA/CS inclusion complex.

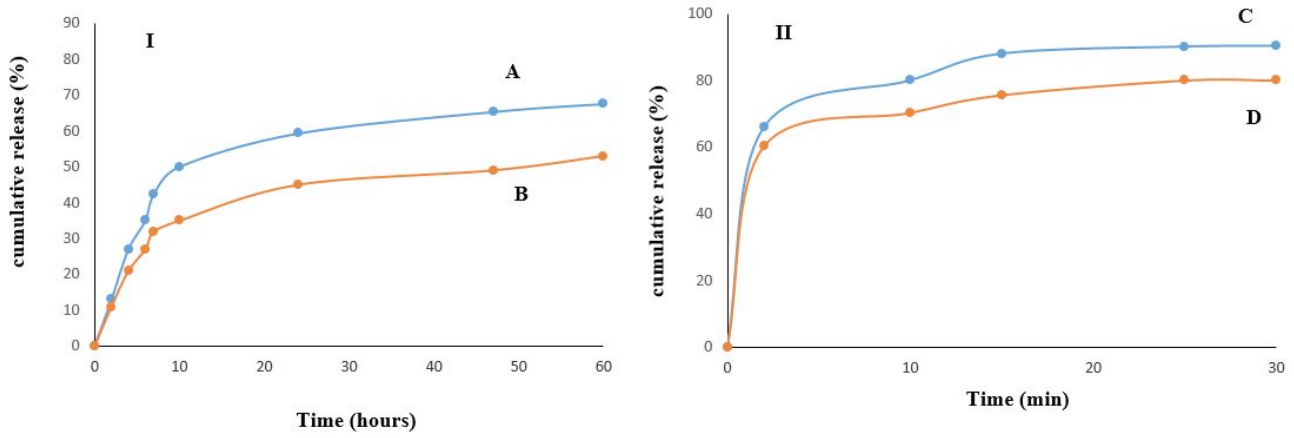

Figure 6 (I): Release profiles of orotic acid from the OA/CS inclusion complex at pH 4.8 (A) and pH 7.4 (B), **(II)** Inset exhibits the release profiles of orotic acid from its physical mixture of orotic acid with CS at pH 4.8 (C) and pH 7.4 (D).

CONCLUSION

The results suggested that the characterization of the OA/CS inclusion complex using FTIR, XRD, DSC, and TEM confirm the incorporation of OA into chitosan cavity. Taking into accounts, the results provide information about the encapsulation of OA with CS, which can lead to essential evaluations of the physicochemical properties of OA/CS, such as stability and morphology. Based on the outcomes retrieved from this study, it can be concluded that the OA/CS inclusion complex has a promising future in the evaluation of drug delivery systems.

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