Synthesis of nano-α mangostin based on chitosan and Eudragit S 100

Abstract

Alpha-mangostin is a xanthone compound isolated from the mangosteen plant (*Garcinia mangostana* L.), which has various pharmacological activities. However, in its utilization alpha-mangostin is unstable and shows low solubility in the oral delivery system. Nanoparticles can deliver specific drugs to their workplace and increase the solubility. The objectives of this study were to create and characterize the alpha-mangostin nanoparticles based on chitosan and Eudragit® S 100. The nanoparticles were made by the ionic gelation method with comparisons core: Coating FI (1:2), FII (1:1), and FIII (2:1). Nanoparticles powder obtained using the spray pyrolysis method. Characterization using Fourier transform infrared indicates that the nanoparticles have been coated properly, and no damage occurred in the formula. The particle sizes for FI, FII, and FII are 373.381 ± 138.023 nm, 398.333 ± 184.977 nm, and 326.567 ± 130.366 nm, respectively, with a smooth surface. The entrapment efficiency value of FI, FII, and FIII are, respectively, 99.7692%, 99.6535%, and 99.476%. Alpha-mangostin was successfully encapsulated in chitosan-tripolyphosphate polymer by ionic gelation method and then coated with Eudragit S 100. Alpha-mangostin chitosan-eudragit nanoparticles (core: Polymer ratio of 1:2) yielded more entrapment efficiency.

Key words: Alpha-mangostin, chitosan, eudragit, nanoparticles

INTRODUCTION

Mangosteen (*Garcinia mangostana* L.) is a tropical plant from the *Clusiaceae* family. Alpha-mangostin is a secondary metabolite that belongs to the main xanthone compound isolated from mangosteen (*G. mangostana* L.).[1] Alpha-mangostin has several pharmacological activities, including anticancer, antioxidant, anti-inflammatory, and antibacterial. In general, lipophilic nutrasecals such as alpha-mangostin show low solubility in the oral delivery system, this is one of the obstacles in utilizing alpha-mangostin activity.[2]

Nanoparticles can be defined as particles with diameters in the range of 1–100 nm, which have typical physicochemical properties. The main purpose of making nanoparticles is to modify particle size, surface properties, release profile, improve the delivery of drug compounds, improve therapeutic index, improve solubility, etc.[3,4] In the formation of a particle into nanosize, the physicochemical properties of the particle will undergo a slight change. Therefore, it is necessary to characterize nanoparticles to see changes in the properties of particles produced both in size, morphology and physically.[5]

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How to cite this article: Herdiana Y, Handaresta DF, Joni IM, Wathoni N, Muchtaridi M. Synthesis of nano-α mangostin based on chitosan and Eudragit S 100. J Adv Pharm Technol Res 2020;11:95-100.

Submitted: 22-Dec-2019
Accepted: 27-Apr-2020
Published: 14-Jul-2020

Access this article online

Quick Response Code:
Website: www.japtr.org
DOI: 10.4103/japtr.JAPTR_182_19
Chitosan has advantages, especially in the development of nanoparticles, because it has unique characteristics and beneficial biological properties such as nontoxic, biocompatibility, biodegradability, cationic properties, and has a mucoadhesive character.\textsuperscript{[6]} Its mucoadhesive properties provide direct contact between the nanoparticles and the gastrointestinal mucosa, which can increase the possibility of cellular uptake by endocytosis and increase the transit time of the nanoparticles in the gastrointestinal tract, leading to better therapeutic efficacy. Uptake of endocytosis from nanoparticles has been widely reported as a new approach to overcome multidrug resistance, a major cause of failure in cancer chemotherapy.\textsuperscript{[7]}

Chitosan is stabilized with polyanion such as sodium tripolyphosphate (TPP) so that the nanoparticles produced are more stable and stronger.\textsuperscript{[7]} Eudragit® S 100 is an anionic copolymer which is soluble in ethanol and acetone but insoluble in acid, pure water, and ethyl acetate. Eudragit® S 100 has been used as a polymer in the application of enteric coatings and colon-specific drug delivery systems.\textsuperscript{[8]}

**MATERIALS AND METHODS**

**Materials**

Alpha-mangostin was obtained from pharmaceutical, natural product laboratories, Padjadjaran University, Indonesia. Chitosan was obtained from PRINT-G labs, Padjadjaran University, Indonesia. Eudragit® S 100 was obtained as a gift sample from Evonik Indonesia Ltd., Jakarta, Indonesia. All other chemicals and reagents used in the study were of analytical grade.

**Methods**

**Preparation of chitosan solution**

The chitosan solution (0.1% [m/v]) was prepared by mixing chitosan powder as much as 0.2 g in 200 mL acetic acid 1%. The solution was stirred using a magnetic stirrer for 30 min.

**Preparation of sodium tripolyphosphate solution**

Crosslinker solution (0.07% [m/v]) was prepared by mixing 0.028 g sodium TPP into 40 mL distilled water and stirred with a magnetic stirrer for 30 min.

**Preparation of Eudragit® S 100 solution**

Eudragit® S 100 solution (0.05% [m/v]) was prepared by mixing Eudragit® S 100 powder as much as 0.1 g in 200 mL ethanol 96%. The solution was stirred using a magnetic stirrer for 30 min.

**Preparation of nanoparticles**

The method used in the formulation of alpha-mangostin nanoparticles with chitosan polymer and Eudragit® S 100 was carried out by the ionic gelation method. Alpha-mangostin solution is added dropwise to chitosan solution (0.1% in acetic acid), followed by the addition of sodium TPP solution (0.07% in distilled water) under constant magnetic stirring and a chitosan-alpha-mangostin-TPP suspension is obtained. Then, the solution of Eudragit® S 100 is added dropwise to chitosan-alpha-mangostin-TPP with a core ratio: 1:2 coating; 1:1; and 1:0.5. The nanoparticle suspension is then sprayed with pyrolysis and characterized. Pyrolysis spray process is carried out at a temperature of 80°C and a flow rate of 5 L/min.

**Characterization Fourier transform infrared**

Characteristics properties of functional groups were analyzed using Fourier transform infrared (FTIR) using KBr pellets. A total of 2 mg of sample powder was mixed with 200 mg KBr to produce a homogeneous grinding pellet and printed with a vacuum. Then, the pellets are subjected to infrared light at wavenumbers from 4000 to 400 cm\(^{-1}\). FTIR was performed on each raw material and nanoparticles powder FI, FII, and FIII.

**Surface morphology and particle size distribution**

The surface morphology characterization of nanoparticles was carried out using a scanning electron microscopy (SEM). The nanoparticle powder is placed on a stub using adhesive on both sides. Then, the powder is made to be electrically conductive with a beam of thin platinum (coating) for 30 s at a pressure of 10 mA. The photo is taken at 10 kV with the desired magnification. Characterization of particle size distribution was determined by processing SEM photos using Image J and Origin Software 8.5 Image J Software, a Java-based image processing program developed at the National Institutes of Health and the Laboratory for Optical and Computational Instrumentation (LOCI, University of Wisconsin).

**Determination of entrapment efficiency**

The determination of entrapment efficiency begins with the separation between adsorbed alpha-mangostin and free alpha-mangostin. The separation was carried out by centrifugation at 3000 rpm for 10 min. The supernatant is taken, then its absorption is measured using an ultraviolet-visible spectrophotometer at the wavelength that has been obtained previously. Determination of the percentage efficiency of alpha-mangostin absorbed from chitosan-alpha-mangostin nanoparticles was calculated using the equation:

\[
\text{\%Entrapment Efficiency} = \frac{W_f - W_t}{W_t} \times 100\%
\]

\[W_f = \text{Weight of total alpha-mangostin}\]

\[W_t = \text{Weight of free alpha-mangostin}\]

**RESULTS**

In this study, nanoparticle powder was obtained by a spray pyrolysis method where drying occurs due to the
evaporation of solvents from a sample transported by gas through a heated tube. The temperature and flow rate used in the drying process are 80°C and 5 L/min; this selection is based on optimization results. Based on the results, nanoparticles obtained yields for FL, FII, and FIII are 78.5 mg, 61.4 mg, and 75.6 mg, respectively. Organoleptic physical examination results of the suspension of FIII nanoparticles before drying have a more turbid color compared to FI and FII [Figure 1], whereas the nanoparticle FI powder after drying has a whiter color compared to FII and FIII.

**Characterization Fourier transform infrared**

As shown in Figure 2, it can be seen that each raw material and nanoparticles have a typical absorption band at a certain wavenumber.

**Surface morphology and particle size distribution**

As shown in Figure 3, the morphology of the three formulas based on SEM with the magnification of × 20,000 is round with a smooth surface, and this is in accordance with the research of Deladino et al., where the optimal morphological form of the encapsulated results is the spherical shape with a smooth surface, which shows the active compound is absorbed well.\(^{[9]}\)

Particle size distribution can be determined through SEM photo results which are then processed using Image J Software and Origin 8.5 to obtain particle size distribution graph. Based on Figure 4. It can be seen that the particle size of FL, FII, and FIII are 373.381 ± 138.023 nm, 398.333 ± 184.977 nm, and 326.567 ± 130.336 nm, respectively. The resulting particle size can be categorized into nanoparticles because it is still in the range of 10–1000 nm.\(^{[10]}\)

In Figure 5, the entrapment efficiency values of FL, FII, and FIII are 99.7692%, 99.6535%, 99.476%, respectively. The value of entrapment efficiency from FL to FIII has decreased.

**DISCUSSION**

Most of the cytotoxic drugs are administered by the intravenous route to attain maximum bioavailability, still,

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**Table 1: The result of chitosan wave number compared with the literature**

| Wavenumber (cm\(^{-1}\)) | Functional group |
|--------------------------|-----------------|
| Result                   | Literature\(^{[11]}\) |
| 350,954                  | 347,868         | O-H stretch dan N-H stretch |
| 289,424                  | 292,413         | C-H stretch |
| 165,302                  | 165,688         | C=O |
| 1597                     | 157,105         | N-H bend |
| 141,963                  | 142,253         | C-H bend |
| 137,816                  | 137,816         | C-N |
| 115,731                  | 115,731         | C-O-C stretch |
| 1079                     | 102,518         | C-O |

**Table 2: The result of sodium tripolyphosphate wave number compared with the literature**

| Wavenumber (cm\(^{-1}\)) | Functional group |
|--------------------------|-----------------|
| Result                   | Literature\(^{[12]}\) |
| 121,132                  | 1210            | \(\nu_{sa}P=O\) |
| 115,731                  | 1130            | \(\nu_{as}O-P=O\) |
| 109,269                  | 1090            | \(\nu_{as}PO_3\) |
| 88,823                   | 888             | \(\nu_{as}P-O-P\) |

**Table 3: The result of Eudragit\(^{®}\) S100 wave number compared with the literature**

| Wave number (cm\(^{-1}\)) | Functional group |
|---------------------------|-----------------|
| Result                    | Literature\(^{[13]}\) |
| 299,743; 295,307          | 295,248         | O-H |
| 173,018                   | 172,917         | C=O |
| 148,426; 145,049          | 144,930         | \(-CH_3\) bend |

**Table 4: The result of alpha mangostin wave number compared with the literature**

| Wave number (cm\(^{-1}\)) | Functional group |
|---------------------------|-----------------|
| Result                    | Literature\(^{[14]}\) |
| 342,081; 325,204          | 3260            | O-H stretch |
| 298,875; 296,078; 292,317| 2989; 2962; 2924| C-H stretch |
| 164,338                   | 1642            | C=O |
| 145,435                   | 1454            | C-C |
| 119,782                   | 1199            | orto-OCH_3 stretch |
| 107,533                   | 1076            | C-O-C stretch |

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*Figure 1:* (a) FI solution before drying, (b) FII solution before drying, (c) FIII solution before drying
treatment failure is observed for most of the cytotoxic drugs. The main problem for the treatment failure is the drug’s inability to act particularly at the target site, which leads to a lack of site-specificity leading to the side effects to both healthy cells and tumor cells by the drug. Some of the other limitations associated with the anti-cancer drugs are their hydrophobic nature, improper biodistribution, and their susceptibility to developing drug resistance.[15] Alpha-mangostin is one of the hydrophobic nature that had pharmacological activity as anti-cancer which shows low solubility in the oral delivery system, this is one of the obstacles in utilizing alpha-mangostin therapeutic activity.[2]

Nanotechnology is currently attracting attention because it offers advantages and has been used widely in various fields of science. A branch of nanotechnology, called nanomedicine, is an application of nanotechnology that is applied to specific medical interventions for prevention, diagnosis, and treatment, thus enabling the development and improvement of controlled and specific drug delivery at the desired location.[16] Nanoparticles are made by using the ionic gelation method, where in this method, an electrostatic reaction occurs between chitosan and sodium TPP. Nanoparticles are formed spontaneously due to constant mechanical stirring at room temperature. Dissolution of chitosan in acetic acid will change the amine group (−NH₂) to ammonium (−NH₃⁺), the free −NH₃⁺ group will cause the formation of nanoparticles complex, which is unstable and weak. To stabilize the positive charge of chitosan and so that the nanoparticles that are formed are stronger so that they can absorb the active compound, then the addition of sodium TPP. Alpha-mangostin is added to the chitosan solution so that it can be absorbed then crosslinking with sodium TPP and coating with Eudragit® S100. The greater the ratio of the core volume to the coating, the better the core will be coated so that it results in better absorption.[15] Particles of small size generally have relatively higher intracellular uptake and broader biological targets. The various particle sizes of each formula can be caused by the ratio of polymers used, the speed in the process of dropping the solution is not constant, and in the atomizer, the process using an ultrasonic nebulizer.

The drying of nanoparticle solvents in this study was carried out by heating using the spray pyrolysis method. SP is the development of spray drying. Drying occurs due to the evaporation of the solvent from the sample transported by gas through a tube that has been heated. The SP process is good application for the synthesis of very fine, uniform, and pure particles.[17]

![Figure 2: Fourier transform infrared spectrum of raw materials and nanoparticles](image1)

![Figure 3: Morphology of nanoparticles obtained with scanning electron microscopy (a) core: coating (1:2), (b) core: coating, (c) core: coating (1:0.5)](image2)
The absorption band at the wave number 1720 cm\(^{-1}\) is a typical absorption band of Eudragit® S 100 which shows the presence of C=O (ester) group of the structure. The absence of new absorption bands in the three formulas indicates that the interactions that occur between alpha mangosteen and polymers are limited to physical interactions.

Based on the results of the graph, it can be seen that the entrapment efficiency values of the three formulas can be categorized as good because they are close to 100%, which indicates that the active compound has been well absorbed in the polymer. The higher the Eudragit® S100 polymer used, the higher the entrapment efficiency value of the nanoparticles because the stronger coating layer is formed.

**CONCLUSION**

It can be evident from the study that alpha-mangostin was successfully encapsulated in chitosan-tpp polymer by ionic gelation method and then coated with Eudragit S 100. Alpha-mangostin chitosan-eudragit nanoparticles (core: Polymer ratio of 1:2) yielded more entrapment efficiency when compared to the other polymer ratios.

**Acknowledgment**

This study was supported by RDDU Grants and academic leadership grants (ALG) no 1373b/UN6.O/LT/2020. We would like to thank Print-G Unpad and for supporting this study through the provision of radioactive compounds and research sites.

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**Table 5: The result of nanoparticles wave number**

|   | Wave number (cm\(^{-1}\)) |   | Functional group                  |
|---|---------------------------|---|----------------------------------|
| FI| 340,243                   | FII| 340,629                         | O-H stretch dan N-H stretch |
|   | 299,352; 295,109          | FII| 299,738; 294,723; 292,794       | C-H stretch                 |
|   | 172,812; 164,335          | FII| 172,436                         | C=O                          |
|   | 155,848                   | FII| 164,721; 155,848                | Interaction P=O and -NH\(_3^+\) |
|   | 141,963                   | FIII| 143,504; 141,189                | C-H bend                     |

**Figure 4:** Graph of particle size distribution (a) core: coating (1:2), (b) core: coating (1:1), (c) core: coating (1:0.5)

**Figure 5:** The Entrapment Efficiency Values

The spectrum of FTIR of chitosan, sodium tripolyphosphate, Eudragit® S100 and alpha mangostin have similar absorption bands with literatures as show in Tables 1-4. The spectrum of FTIR in Figure 2 shows the spectrum of FI, FII, and FIII have similar absorption bands. The difference occurs in the band with a wavenumber of 1720 cm\(^{-1}\), as shown in Table 5, which is a typical absorption band of the Eudragit® S 100 polymer where the intensity decreases from FI to FIII, and this is directly proportional to the reduction in the Eudragit® S 100 polymer used. In addition, no new absorption bands appear from each formula. The decrease in intensity at wave number 1720 cm\(^{-1}\) from the FI spectrum to FIII is due to the less Eudragit® S 100 used.
Financial support and sponsorship
Thesis Doctorate of Unpad Grants (RDDU) and ALG no 1373b/UN6.O/LT/2019. We would like to thank to Print-G Unpad and for supporting this study through the provision of radioactive compounds and research sites.

Conflicts of interest
There are no conflicts of interest.

REFERENCES

1. Ji X, Avula B, Khan IA. Quantitative and qualitative determination of six Xanthones in *Garcinia mangostana* L. by LC-PDA and LC-ESIMS. J Pharm Biomed Anal 2007;43:1270-6.
2. Aisha AF, Abdulmajid AM, Ismail Z, Alrokayan SA, Abu-Salah KM. Development of polymeric nanoparticles of *Garcinia mangostana* Xanthones in Eudragit RL100/RS100 for anti-colon cancer drug delivery. J Nanomater 2015;2015:1-12.
3. Fabrega J, Fawcett SR, Renshaw JC, Lead JR. Silver nanoparticle impact on bacterial growth: Effect of pH, concentration, and organic matter. Environ Sci Technol 2009;43:7285-90.
4. Akbal Ö, Erdal E, Vural T, Kavaz D, Denkbaş EB. Comparison of protein- and polysaccharide-based nanoparticles for cancer therapy: Synthesis, characterization, drug release, and interaction with a breast cancer cell line. Artif Cells Nanomed Biotechnol 2016;45:193-203.
5. Abdullah M. *Pengantar Nanosains*. Bandung: Penerbit ITB; 2009.
6. Kumar MN. A review of chitin and chitosan applications. React Funct Polym 2000;46:1-27.
7. Lin WC, Yu DY, Yang MC. pH-sensitive polyelectrolyte complex gel microspheres composed of chitosan/sodium tripolyphosphate/ dextran sulfate: Swelling kinetics and drug delivery properties. Colloids Surf B Biointerfaces 2005;44:143-51.
8. Prajakta D, Ratnesh J, Chandan K, Suresh S, Grace S, Meera V, et al. Curcumin loaded pH-sensitive nanoparticles for the treatment of colon cancer. J Biomed Nanotechnol 2009;5:445-55.
9. Deladino L, Anbinder PS, Navarro AS, Martino MN. Encapsulation of natural antioxidants extracted from *Ilex paraguariensis*. Carbohydrate Polymers 2008;71:126-34.
10. Martien R, Adhyatmika ID, Irianto K, Farida dan V, Sari DP. Developments of Nano based drug delivery systems. Majalah Farmaseutik 2012;8:133-44.
11. Yasmeen S, Kabiraz MK, Saha B, Qadir R, Gafur A, Masum S. Chromium (VI) ions removal from tannery effluent using chitosan-microcrystalline cellulose composite as adsorbent. Int Res J Pure Applied Chem 2016;10:1-14.
12. Loutfy SA, El-Din HM, Elberry MH, Allam NG, Hasanin MT, Abdellah AM. Synthesis, characterization and cytotoxic evaluation of chitosan nanoparticles: *In vitro* liver cancer model. Adv Nat Sci Nanosci Nanotechnol 2016;7:1-9.
13. Manikandan M, Kannan K, Manavalan R. Compatibility studies of camptothecin with various pharmaceutical excipients used in the development of nanoparticle formulation. Int J Pharm Sci 2013;5:315-21.
14. Nelli GB, Solomon KA, Kilari EK. Antidiabetic effect of α-mangostin and its protective role in sexual dysfunction of streptozotocin induced diabetic male rats. Syst Biol Reprod Med 2013;59:319-28.
15. Tummala S, Kumar MN, Prakash A. Formulation and characterization of 5-fluorouracil enteric coated nanoparticles for sustained and localized release in treating colorectal cancer. Saudi Pharm J 2015;23:308-14.
16. Rawat M, Singh D, Saraf S, Saraf S. Nanocarriers: Promising vehicle for bioactive drugs. Biol Pharm Bull 2006;29:1790-8.
17. Panatarania C, Muharam DG, Wibawa dan BM, Joni IM. Blue luminescence of zno: zn nanocrystal prepared by one step spray pyrolysis method. Mater Sci Forum 2013;737:20-7.
18. Garti N, McClements DJ. Encapsulation Technologies and Delivery Systems for Food Ingredients and Nutraceuticals. UK: Woodhead Publishing Limited; 2012.