JURNAL REKAYASA PROSES

Research article / Vol. 13, No. 2, 2019, hlm. 145-150

Journal homepage: http://journal.ugm.ac.id/jrekpros

Synthesis of Curcumin Nanoparticle from *Curcuma xanthorrhiza* Roxb. Extract by Solvent-Antisolvent Precipitation Method

Nur Rofiqoh Eviana Putri*, Annisa Amalia Ulfah, and Yuni Kusumastuti
Department of Chemical Engineering, Faculty of Engineering, Universitas Gadjah Mada
Jl Grafika No. 2 Kampus UGM, 55281 Yogyakarta
* Corresponding author: nur.rofiqoh.e@mail.ugm.ac.id

(Submission: October 25, 2019; Revision: November 17, 2019; Acceptance: November 19, 2019)

ABSTRACT

Curcumin is an active compound found in *temulawak* (*Curcuma xanthorrhiza* Roxb.) extract which is widely used for biomedical application. However, the utilization of curcumin is still limited due to its properties i.e. hydrophobicity, poor stability, and low water solubility. Modification of curcumin molecule and process optimization during the extraction and purification process is needed to minimize the aforementioned limitations. One of the approaches is producing curcumin in nano size. This present research aims to optimize the synthesis of curcumin nanoparticle from *Curcuma xanthorrhiza* Roxb. extract using solvent-antisolvent precipitation method. Curcumin colour stability was also enhanced by controlling the pH during raw materials preparation. The obtained curcumin nanoparticle was then characterized using particle size analysis (PSA). Result showed that *Curcuma xanthorrhiza* Roxb. extract colour could be controlled by maintaining acidic environment. At the pH of 3, yellow colour of extract was obtained, meanwhile at neutral pH, the colour of extract changed into dark brown. PSA result showed that optimum stirring condition of precipitation process was obtained using 500 rpm stirring rate for 45 minutes which resulted in curcumin nanoparticle in the size range of 164.37±3.29 nm. Thus, by controlling the pH of extract at 3 during extraction process and using optimum stirring condition at 500 rpm for 45 minutes during precipitation process, more stable and soluble curcumin was successfully produced.

Keywords: *Curcuma xanthorrhiza* Roxb.; curcumin nanoparticle; solvent-antisolvent precipitation, *temulawak*

ABSTRAK

Kurkumin merupakan salah satu senyawa aktif yang terkandung dalam ekstrak temulawak (Curcuma xanthorrhiza Roxb.) yang banyak digunakan untuk aplikasi biomedis. Meskipun demikian, pemanfaatan kurkumin masih terbatas dikarenakan sifatnya yang hidrofobik, stabilitas yang rendah, serta kelarutan di air yang rendah. Modifikasi kurkumin serta optimisasi proses ekstraksi maupun purifikasi perlu dilakukan untuk mengatasi kelemahan tersebut. Salah satu pendekatan yang dapat dilakukan adalah dengan membuat kurkumin dalam ukuran nano. Penelitian ini bertujuan untuk mengoptimasi pembuatan nanopartikel kurkumin dari ekstrak Curcuma xanthorrhiza Roxb. menggunakan metode presipitasi solven-antisolven. Stabilitas warna kurkumin dijaga dengan mengontrol pH saat persiapan bahan baku. Hasil nanopartikel kurkumin yang didapat kemudian dianalisis menggunakan particle size analysis *(PSA). Hasil menunjukkan bahwa warna ekstrak Curcuma xanthorrhiza Roxb. dapat dijaga pada kondisi asam. Pada pH 3, ekstrak berwarna kuning cerah sedangkan pada pH netral, warna ekstrak berubah menjadi coklat gelap. Hasil PSA menunjukkan bahwa kondisi pengadukan yang optimum pada saat proses presipitasi diperoleh*

DOI: 10.22146/jrekpros.50909

e-ISSN 2549-1490 p-ISSN 1978-287X

menggunakan kecepatan pengadukan 500 rpm selama 45 menit yang menghasilkan nanopartikel kurkumin dengan ukuran 164,37±3,29 nm. Dengan mengontrol ekstrak pada pH 3 selama proses ekstraksi dan menggunakan kondisi pengadukan optimum pada 500 rpm selama 45 menit pada proses presipitasi, kurkumin dengan stabilitas dan kelarutan yang baik dapat diperoleh.

Kata kunci: Curcuma xanthorrhiza Roxb.; nanopartikel kurkumin; presipitasi solven-antisolven; temulawak

1. Introduction

Curcuma xanthorrhiza Roxb. oil is one of traditional medicines which is widely used for biomedical application. It is extracted from *Curcuma xanthorrhiza* Roxb. or known as Javanese Turmeric (*temulawak*) in Indonesia. *Curcuma xanthorrhiza* Roxb. oil contains curcumin which has anticancer (Aggarwal et al., 2003), antioxidant (Aftab and Vieira, 2010), and anti-inflammatory (Jurenka, 2009) properties. The use of *Curcuma xanthorrhiza* Roxb. as the source of curcumin for medicine has been increasing due to the increasing awareness for nature-based medicine and the rising price of chemicals (Nihayati et al., 2013). In the world, curcumin demand was fulfilled from turmeric extract which is coming from India. About 80% of world turmeric production was obtained from India and 60% of world export was also obtained from India (Lal, 2012). Whereas, Indonesia as tropical country has an abundance *Curcuma xanthorrhiza* Roxb. as the source of curcumin. The production of *temulawak* in Indonesia has been increasing. In 2014 to 2015, its growth is rising 12.33% with the harvested area (m^2) of 13,178,025 to 14,803,423 (BPS, 2015).

The utilization of curcumin is still limited because of its characteristics i.e. hydrophobicity, poor chemical stability, low bioavailability and low water solubility (Anand et al., 2007). Curcumin is also unstable to the change of pH, light, temperature and it exhibits poor chemical stability in aqueous solution and in oil-in-water emulsion. As a medicinal compound, curcumin needs to be more stable and to have high water solubility so that its therapeutic effectiveness can be enhanced.

One of the promising approaches to stabilize curcumin is by producing curcumin in the form of nanoparticles. The size of curcumin nanoparticles is less than 1 micron so that its solubility and bioavailability are increased (Devalapally et al., 2007). The size of nanoparticles varies but it is commonly in the range of 100-500 nm (Rizvi and Saleh, 2018). Nanoparticle can be made through both top-down and bottom-up methods (Pattekari et al., 2011). Development of synthesis process is still explored to find the easiest and low-cost method which produces homogeneous nanoparticle. Solvent-antisolvent precipitation method is quite attractive for its simplicity and affordability. This method is carried out by dissolving compound into its solvent and then adding the antisolvent rapidly under constant stirring to reach super saturated condition. Precipitation will occur and nanoparticle can be obtained (Kakran et al., 2012).

Some experiments have been conducted to optimize the solvent-antisolvent methods by varying its ratio, stirring speed, and solvent types. Kakran et al., 2012 successfully produced curcumin nanoparticle by studying the effect of antisolvent addition, ratio of solvent to antisolvent, and also stirring speed. Yadav and Kumar (2014) was also successfully synthesized curcumin nanoparticle using solvent-anti solvent precipitation method and adding stabilizer to obtained more homogeneous particles. However, those previous researches were done with pure curcumin. Meanwhile, in order to utilize the local source of *Curcuma xanthorrhiza* Roxb., many aspects of curcumin nanoparticle formation from *Curcuma xanthorrhiza* Roxb. extract need to be explored. Pure curcumin has different chemical and physical characteristic than curcumin in *Curcuma xanthorrhiza* Roxb. oil. During the extraction process, the stability of *Curcuma*

xanthorrhiza Roxb. extract also needs to be controlled.

This research was focused on examining the synthesis of curcumin nanoparticles from *Curcuma xanthorrhiza* Roxb. extract by using solvent-antisolvent method. The experiment consists of two parts, i.e. extraction of curcumin from *Curcuma xanthorrhiza* Roxb. and production of curcumin nanoparticles from *Curcuma xanthorrhiza* Roxb. extract. During the extraction process, the pH will be controlled to prevent unwanted reactions and hence to produce more stable extract. In addition, during the nanoparticle synthesis, the effects of stirring time and stirring rate were studied. The resulted curcumin nanoparticles were then characterized using particle size analysis (PSA) to clarify the effect of those parameters on particle size distribution. By applying the optimum conditions suggested by this study, curcumin nanoparticles with good stability and homogeneous nano particle size distribution can be produced.

2. Research Methodology

2.1 Materials

Fresh *Curcuma xanthorrhiza* Roxb. known in Indonesia as *temulawak* was obtained from traditional market in Bantul, Yogyakarta, Indonesia. Citric acid (analytical grade, Sigma Aldrich) was used for raw material preparation, meanwhile technical grade of ethanol (76%, General Labora) was used for extraction process.

2.2 Procedures

Curcuma xanthorrhiza Roxb. rhizome was peeled, cleaned, and then wet-ground into small pieces using blender. An amount of 150 gram of the ground rhizome was adjusted to different pH values, i.e 3, 5, and 7, by adding citric acid. After that, *Curcuma xanthorrhiza* Roxb. rhizome was macerated with 400 mL ethanol 76% for 1 hour and continued to the extraction process in 70 ^{o}C for additional 1 hour using the extraction tools set as described by Harjanti (2008). The extract was then separated by filtration proccess and concentrated using rotary vaccum evaporator until the volume was reduced to 100 mL. Concentrated curcumin extract was added slowly to the aquadest and kept in stirring for 2 hours. Every 15 minutes, sample was taken and analyzed using PSA to observe the particle size. Different stirring rate (500, 750, and 1000 rpm) and different stirring time (15, 30, 45, 60, and 120 minutes) were tested to see the effect of those parameters on particle size. After 2 hour of stirring process, curcumin nanoparticle was obtained and then separated using centrifuge. It was dried up in 50 ^{o}C until curcumin nanoparticle powder was obtained. The resulted nano curcumin was then analysed using PSA (Horiba Scientific, Nanoparticle Analyzer SZ-100, Japan). The measurement was performed at 24.9 ^{o}C of holder temperature and 90^{o} of scattering angle.

2.3 Statistical Analysis

One-way analysis of variance (ANOVA) was used to see the significant differences and Tukey's post hoc test was performed to compare the data. The significant difference was considered when the p value was less than 0.05.

3. Results and Discussion

3.1 Optimization of Extraction Process

Citric acid was added to *Curcuma xanthorrhiza* Roxb. before extraction process to prevent the browning reaction. Citric acid was used to control the acid condition because it is an acid which is safe for food and medical application as it is listed in Food and Drug Administration (FDA) (Shukla et al., 2017). From the appearances, the final extracts have different colour as shown in Figure 1. The change of colour is obtained from the reaction of nucleophilic groups in the curcumin which act as a proton acceptor to the positively charged hydrogen from dissociated citric acids.

These finding highlights that altering the pH of curcumin extract into acid condition could decrease the posibility of curcumin degradation and prevent browning reaction. Curcumin stability decreases as the pH of the solution increases which further results in the formation of its degraded products such as ferulic acid

(brownish-yellow) and vanillin (Kumavat et al., 2013). Figure 1 shows that increasing pH could change the colour of curcumin to be more brownish as a sign of curcumin degradation. When the colour remains yellow, curcumin stability in the presence of light is increased.

(a) (b) (c)

Figure 1. Appearances of *Curcuma xanthorrhiza* Roxb. extract in pH of 3 (a), 5 (b) and 7 (c) during extraction process

Figure 2 shows curcumin nanoparticle average size at different pH. At pH of 3, 5, and 7, the average sizes of curcumin nanoparticle were 129.0 nm, 109.5 nm, and 180.0 nm, respectively. The size of curcumin nanoparticle at pH 3 was larger than curcumin nanoparticle at pH 5. It might be because in low pH, there was an increase in crystal formation. The tendency of crystal formation will be higher when the solution was mixed (Kharat et al., 2016). Meanwhile, the size of curcumin nanoparticle at neutral pH was larger than at acidic condition. It was likely because at neutral pH, curcumin was highly unstable (Kharat et al., 2016).

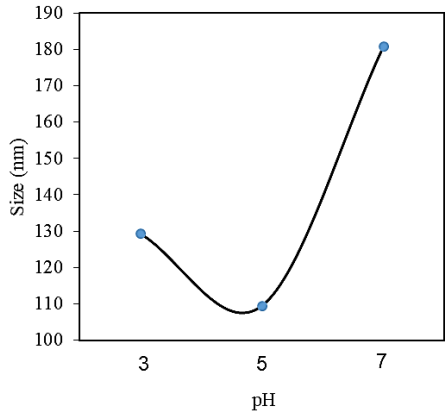

Figure 2. Average size of curcumin nanoparticle in different pH

3.2 Optimization of Curcumin Nanoparticle Preparation

Curcumin nanoparticle average sizes in different stirring rates and stirring times are shown in Figure 3. Trends of curcumin nanoparticle size against time for sample which was prepared by 750 rpm and 1000 rpm stirring rate are similar. Curcumin nanoparticle average size was decreased as the time increased from 15 to 45 minutes and reached minimum curcumin nanoparticle size at 45 minutes. Beyond 45 minutes, the size of curcumin nanoparticle rapidly increased and formed stable size at 120 minutes. On the contrary, at 500 rpm stirring rate, the curcumin nanoparticle size tends to decrease as time increases until reaching the stable size at 120 minutes. It might be caused by low stirring rate which lower the posibility of curcumin to collide and could not make an aggregate of particle as exhibited in samples prepared at higher stirring rate.

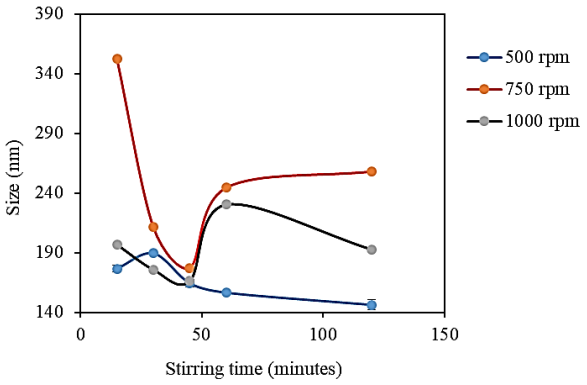

Figure 3. Size of curcumin nanoparticle synthesized at different stirring time and stirring rate

The average size at 120 minutes stirring time for sample which was prepared using 1000 rpm stirring rate is smaller than sample which was prepared using 750 rpm stirring rate as shown in Figure 4. It indicates that the possibility of particle to collide to each other in 1000 rpm is higher than 750 rpm. High particle collision tends to cause broken particles. In the high rate and long time stirring period, curcumin tends to form agregate in a specific time. In this case, the agregate curcumin formed bigger size molecule with around 60-minute stirring process as shown

in Figure 3. Those specific time did not occurred in samples which was prepared using 500 rpm.

Figure 5 shows the size of nano curcumin in different stirring rate at the optimum stirring time, i.e. 45 minutes. At that time, the average sizes of all samples which were prepared with different stirring rate were not significantly different.

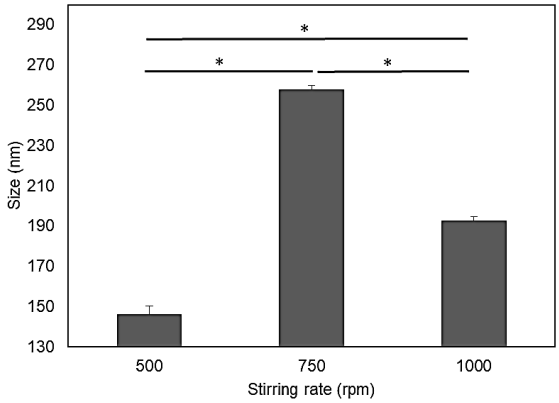

Figure 4. Size of curcumin nanoparticle synthesized at different stirring rate at 120 minutes of stirring time

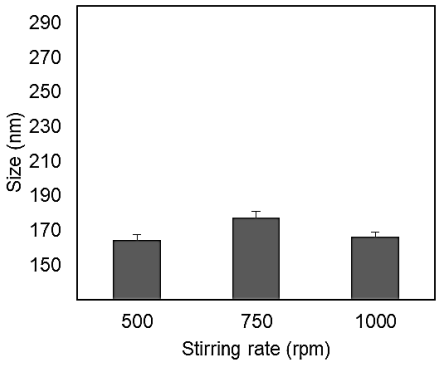

Figure 5. Size of curcumin nanoparticle synthesized at different stirring rate at 45 minutes of stirring time

4. Conclusions

Utilization of local *Curcuma xanthorrhiza* Roxb. (*temulawak*) to produce curcumin nanoparticle using solvent-antisolvent precipitation method was explored. Curcumin which has better stability and better nanoparticle size distribution can be produced by optimizing the extraction process and nanoparticle synthesize process. At pH value of 3, curcumin extract exhibited the expected yellow colour and the extract was also more stable to be compared to the extract obtained with other pH values. PSA result showed that all samples which were prepared at pH of 3, 5 and 7 contained nanoparticles in the range of 109.5 to 180.8 nm average size. Optimum stirring time for precipitation process was obtained at 45 minutes meanwhile the optimum stirring rate was obtained at 500 rpm. At the optimum stirring time, there was no significant difference of curcumin nanoparticle sizes when the process was run with different stirring rates.

Acknowledgements

The authors acknowledge Universitas Gadjah Mada's internal grant scheme "Optimasi Pembuatan Nanopartikel Kurkumin dari Rimpang Temulawak (*Curcuma xanthorrhiza*)" with research grant number 4010/UN1/DITLIT/DIT-LIT/LT/2019.

References

Aftab, N., and Vieira A., 2010, Antioxidant activities of curcumin and combinations of this curcuminoid with other phytochemicals, Phytother. Res., 24, 500–502.

Aggarwal, B. B., Kumar, A., and Bharti, A. C., 2003, Anticancer potential of curcumin: preclinical and clinical studies, Anticancer Res., 23, 363–398.

Anand, P, Kunnumakkara A. B., Newman R. A., and Aggarwal B. B., 2007, Bioavailability of curcumin: Problems and promises, Mol. Pharm., 4, 807–818.

Badan Pusat Statistik (BPS), 2015, Statistics of Medical Plants Indonesia, BPS, Jakarta, pp 9.

Devalapally, H., Chakilam, A., and Amiji M. M., 2007, Role of nano- technology in pharmaceutical product development, J. Pharm. Sci., 96, 2547–2565.

Harjanti, R. S., 2008, Pemungutan kurkumin dari kunyit *(Curcuma domestica val.)* dan pemakaiannya sebagai indicator analisis volumetri, J. Rek. Pros., 2(2), 49-54.

Jurenka, J. S., 2009, Anti-inflammatory properties of curcumin, a major constituent of Curcuma longa: a review of preclinical and

clinical research, Altern. Med. Rev., 14, 141–153.

Kakran, M., Sahoo N. G., Tan, I., and Li, L., 2012, Preparation of nanoparticles of poorly water-soluble antioxidant curcumin by antisolvent precipitation methods, J. Nanopart. Res., 14, 1-11.

Kharat, M., Du, Z., Zhang, G., and Mcclements, D. J., 2016, Physical and chemical stability of curcumin in aqueous solutions and emulsions: Impact of pH, temperature, and molecular environment, J. Agr. Food Chem., 65(8), 1525-1532.

Kong, G., Braun, R. D., and Dewhirst, M. W., 2000, Hyperthermia enables tumor-specific nanoparticle delivery: effect of particle size, Cancer Res., 60, 4440-4445.

Kumavat, S. D., Chaudhari, Y. S., Borole P., Mishra, P., Shenghani, K., and Duvvuri, P., 2013, Degradation studies of curcumin, IJPRR, 3(2), 50-55.

Lal, J., 2012, Turmeric, curcumin and our life: A review, Bull. Environ. Pharmacol. Life Sci., 1(7), 11-17.

Nihayati, E., Wardiyati, T., Retnowati, R., and Soemarno, 2013, The curcumin content of temulawak (*Curcuma xanthorriza Roxb.*) rhizome as affected by N, K and micronutrients B, Fe, Zn, Agrivita, 35(3), 218-226.

Pattekari, P., Zheng, Z., Zhang, X., Levchenko, T., Torchilin, V., and Lvov Y., 2011, Top-down and bottom-up approaches in production aqueous nanocolloids of paclitaxel, Phys. Chem. Chem. Phys., 13, 9014–9019.

Rizvi, S. A. A., and Saleh, A. M., 2018, Applications of nanoparticle systems in drug delivery technology, SPJ, 26 (1), 64-70.

Shukla, P., Sharma, A., and Sharma, A., 2017, Food additives from an organic chemistry perspective, MOJBOC, I (3), 70-79.

Yadav, D., and Kumar, N., 2014, Nanonization of curcumin by antisolvent precipitation: process development, characterization, freeze drying and stability performance, Int. J. Pharmaceutics, 477, 564-577.