The characteristic improvement of nano biomass

E Noor¹, F A K Haque¹, M Fachrizal¹ and W Niloperbowo²

¹Department of Agroindustrial Technology, Bogor Agricultural University, Bogor, Indonesia
² Bioscience and Biotechnology Research Center, Institute Technology Bandung, Bandung, Indonesia

E-mail: erlizanoor@yahoo.com

Abstract. The application of nanotechnology for agriculture products (biomass) is increasing recently, as it shown an improvement in product quality. The research aim was to determine the characteristic changing of some products Curcumin and Gingerol nanoemulsion. The production of nanoemulsion was obtained by high speed homogenization in the variation of rotation speed, time and solution compositions. The quality improvement of biomass in general include the increase of bioavailability, effectivity, stability and enhanced of solubility in the various solvents that would rise it application for various products. The addition of stabilizer and antioxidant in such level would prevent the agglomeration of particle and therefore improve the stability for some extend. Depending on the use of the application, the nanoemulsion of curcumin and gingerol. The characteristic determined were conductivity, stability, dispersability, bioavailability and polarity. In general, the characteristic of nanoemulsion are improved, therefore, the opportunity to expand its application especially for pharmaceutical, cosmetics and other industries.

1. Introduction

Nanotechnology is one of the most important and fast-growing engineering fields today. This is because nanotechnology can be applied widely in various fields including environment, industry, food, agriculture, biomedical etc. Technology applications contribute greatly to the field of the agricultural industry to produce healthcare, food and other products. In the field of food nanotechnology applications offer advantages in improving the bioavailability of active ingredients, controlling the release of active ingredients as well as improving sensory properties. Nano particles will bring up the new properties. In applications, novel properties include electronic, mechanical, optical and catalytic reactivity. The new properties would produce new products with stronger material performance, lighter and faster. Some research results related to the provision of nano-sized biomaterials using agricultural products presented in this paper include fabrication of nanocurcumin and nanogingerol. Particle nanoscale research as a carrier of active substances and the introduction of active substances in herbal products has evolved over the last few years. At the nanometer scale or one-billionth of a meter (10⁻⁹m), the material has unique and far superior features and performance and phenomena than on a micro-meter scale (10⁻⁶ m), so that the properties and the performance of the material can be engineered in such a way to be more effective and efficient. At emulsions, the performance of
nanoemulsions is far superior to that of ordinary emulsions [1]. Other food ingredients that have wide application include antioxidants. Flavonoids are the largest group of phenol compounds in plants and have strong antioxidant activity. The modification of antioxidant to nanoemulsion has been done by researchers including flavonoids glycosides from Gedy leaves (Abelmoschus manihot L. Medic) which can serve as hepatoprotector [2], nano particles of low pentin of the lime peel (Citrus aurantifolia Swingle) and for the encapsulation of probiotics [3]. This paper shows the results of research related to the advantage of nanocurcumin and nanogingerol solutions in increasing the solubility of nanoemulsion in polar and semi polar solvents, conductivity and bioavailability. Nanoemulsion particle size stability was also observed during storage at variations in storage temperature and addition of additives.

2. **Materials and methods**

The biomass used for the manufacture of nanocurcumin are temulawak (Curcuma xanthorrhiza Roxb), nanogingerol is ginger emprit (Zingiber officinale Rosc var. Amarum). The manufacture of nanoemulsion temulawak and gingerol begins with the manufacture of emulsions. The raw materials were extracted by maceration using 98% ethanol solvent. The emulsion solution was further nanoemulsified with homogenization using a high-speed homogenizer. The rotational speed and time spent varied from 8000-12000 rpm for 20-40 minutes for nanocurcumin, as well as setting the temperature between 30-50°C at 22000 rpm for nanogingerol. In the manufacture of nanoemulsion, a phosphate buffer solution for nanocurcumin, sodium dihydrogenphosphate buffer for nanogingerol, and addition of Tween 80 surfactant for all products. Measurement of particle size using PSA (Particle Size Analyzer), bioavailabilty using Franz diffusion cells [4].

3. **Results and discussion**

The characteristics improvement of nanocurcumin and nanogingerol extracts of temulawak and ginger has long been used in traditional and modern medicinal herbs. Extensive uses include functional foods, medicines, flavorings, fresheners, cosmetics, perfumes and a variety of other industries. Increasing the characteristics of the extract can be done by converting the active ingredient to nanometer size (less than 100 nm) and nanoemulsion solution. In the curcumin nanoemulsion, additional of Tween 80 surfactants and maltodextrin were able to stabilize the solution, while in gingerol nanoemulsion the addition of Tween 80 also added with 96% ethanol. The important characteristic changes of the two nanoemulsion solutions compared to the emulsion solutions are the solubility in polar and semi-polar solvents, increased product stability and increased bioavailability. Increased bioavailability is important because it will increase the absorption of the material in the body so as to reduce the dosage of use. Comparison of nanoemulsion and emulsion characteristics of curcumin and gingerol is given in table 1.
Table 1. Characteristic of emulsion and nanoemulsion of Curcumin and Gingerol.

| Parameters          | Curcumin emulsion | Nanocurcumin | Gingerol emulsion | Nanogingerol |
|---------------------|-------------------|--------------|-------------------|--------------|
| Colour              | Dark brown        | Yellow       | Yellow cloudy     | Clear yellow |
| Particle size       | ≥ 6 µm            | 86 nm        | 496 nm            | 80 – 89 nm   |
| pH                  | 5.2               | 6.8          | 6.98              | 7.2          |
| Viscosity (cP)      | 7.1               | 3.9          | 2.82              | 1.85         |
| Solubility          |                   |              |                   |              |
| Heksane             | 0                 | 90.17        | 24.7              | 8.6          |
| Aceton              | 90.96             | 95.70        | 48.1              | 93.0         |
| Ethanol             | 96.34             | 96.74        | 57.7              | 92.2         |
| Methanol            | 0                 | 91.40        | 55.2              | 93.6         |
| Water               | 0                 | 90.00        | 53.6              | 94.0         |
| Conductivity        |                   |              |                   |              |
| Bioavailability (%) | 13.79             | 20.13        | 17.58             | 49.50        |

Decreasing the particle size will decrease the viscosity of the nanoemulsion solution (table 1). This decrease in viscosity could reduce the aggregation of particles so that dispersion of particle would improve. The smaller size of the particle diameter also will increase the surface area and increase the emulsion resistance to flow thus increasing the viscosity [5]. The pH value of the nanoemulsion solution also increases and is in the neutral pH range. So, both nanoemulsions solution is safe to use as a base of lotion, cream or ointment and commercial applications such as for food, beverage and pharmaceutical products. Both nanoemulsions of the material exhibits a change in properties compared to the emulsion form i.e. increased solubility at various levels of solvent polarity. Especially in semi-polar and polar solvents. In this solubility test both nanoemulsions can change the solubility properties, high solubility in water is very advantageous in the application for the drug so it is safer for the body.

3.1. Bioavailability of nanoemulsion

Bioavailability is the rate and relative amounts of drugs that reach the general circulation of the body or circulatory system. The benefits of bioavailability include the time required for a drug to have a therapeutic effect and how much it can be absorbed by the body. Penetration ability of nanoemulsion is faster and greater than the emulsion for curcumin and gingerol were observed. The emulsion dan nanoemulsion of nanocurcumin dan nanogingerol demonstrated low cumulative concentration in two hours of penetration process. Then increase rapidly afterward, the concentration measurement by using the Franz diffusion apparatus showed after 8 hours of receptor fluid of nanocurcumin emulsion was penetrated 30.19 mg/cm², whereas in the emulsion only prepared by 20.43 mg/cm². Percentage of curcumin penetrated after 8 h on nanoemulsion and emulsion forms were 20.13% and 13.79% (figure 1). This result shows an increase in penetration of nanoemulsion solution by 46%. For gingerol after 8
hours of testing with Franz diffusion on the receptor fluid the nanoemulsion solution was penetrated 2.58 mg/cm², whereas in the emulsion solution only 1.43 mg/cm². The percentage of emulsion penetration after 8 h for nanogingerol and gingerol emulsion were 49.50% and 17.58% respectively.

The penetration ability of the nanoemulsion is better than that of the emulsion due to the smaller size of nanoemulsion droplets compared to the emulsion. The droplet size of a drug affects absorption in the body, as evidenced by a study comparing the bioavailability of nanogel and gel preparations [6]. In addition to small size, viscosity also affects the release of the active ingredient from the base to the membrane surface. The low viscosity of nanoemulsion compared with the emulsion will increase the penetration speed, thereby increasing the mobility of the active substance to the membrane surface. In addition to the size, electrical conductivity also affects the penetration ability as the ionic properties contained in nanoparticles are effective in the system because it will be very easily removed from its matrix rather than a cationic preparation [7].

![Figure 1. Bioavailability of Curcumin in the emulsion and nanoemulsion solutions.](image1)

![Figure 2. Bioavailability of Gingerol in the emulsion and nanoemulsion solutions.](image2)

3.2. Stability of nanoemulsion

The change in particle size of nanocurcumin during storage at various temperatures is shown in Figure 3. At room temperature, the nanocurcumin particle size is relatively stable until 30 days, while the curcumin emulsion only lasts for 9 days. The size of the nanocurcumin particles observed at 4, 30 and 50 °C showed the absorption increase rapidly according to increasing of particle size at higher storage temperatures. Visually the increase of this size is accompanied by the clarity of the solution. During
storage at 4 °C, the flocculants formed in the solution after 60 days, and the color changes cloudier. An increase in size above 100 nm was achieved after 50 days storage, in 60 days storage the particle size reached 152 nm and continued to increase until 90 days to 384 nm. At temperature of 30 and 50 °C the size increases faster. The particle size greater than 100 nm are achieved after 10 days and 2 days for temperatures of 30 and 50°C, respectively. This increase in the size of r nanocurcumin is due to the formation of granular aggregations as the nature of the oils which tend to form large grains in the water phase [8].

The storage at 50°C, the breakdown of nanoemulsion is characterized by the creaming formed on day 15. The addition of Tween 80 as a stabilizer at this temperature is no longer effective due to a decrease in Tween 80 ability to maintain stable emulsion interface stability. Tween 80 is reported to have good emulsifying power at temperatures <250°C [9]. In general, the emulsifying temperature is more effective when it is in conditions around its optimum temperature because it has a larger surface activity.

![Particle size of nanocurcumin during storage at various temperature.](image)

**Figure 3.** Particle size of nanocurcumin during storage at various temperature.

The addition of citric acid to the nanocurcumin emulsion could prevent the loss of its concentration during the storage. The addition of 3% citric acid is ineffective to prevent the increase in particle size. However, this effective to inhibit the decrease in curcumin concentration during storage (figure 4). Citric acid as a secondary antioxidant that would prevent damage through the mechanism of capturing oxygen into non-radical products [10]. The ability of citric acid in reducing degradation of curcumin has also been proven [11].
The storage temperatures would considerably affect the particle size of nanoemulsion. The particles size of nanogingerol emulsion increased during storage for all temperature 4, 30 and 50°C. Graph 5 shows that at low temperature (4°C), particle size gradually changes from initial size. Whereas at higher temperature (30 and 50°C), the particle size changes substantially on the first day. This shows that the temperature is very influential on the stability of nanogingerol and also nanocurcumin.

At high storage temperature, more energy is transferred to the water phase and cause the molecule movement and increase the surface tension of the water [12]. The increase of water-oil surface tension would cause the nanoemulsion unstable [13]. This is in line with the report that states under different conditions the temperature of the environment causes the reduction of the interaction strength between the phase granules to be dispersed with the dispersing phase thus causing emulsion instability [14]. At high temperatures droplet-droplet interactions are stronger and particle joining occurs. This influence is reinforced by changes in the properties of Tween 80 on emulsions which change from hydrophilic properties at low temperatures to lipophilic with an increase in temperature due to dehydration of polyoxyethylene chains [15]. At high temperature the particle size continue to increase after 3 days. Whereas, at lower storage temperature (4 and 25°C) after 3 days the particle size are relatively stable.
4. Conclusion
The result conclude that small particle size of Curcuma and Gingerol nanoemulsion has performed better emulsion characteristics. Nanoemulsion of formation by reducing the size of the active curcumin and gingerol ingredient to less than 100 nm has improved the characteristics of the solution. Character enhancement is characterized by higher solubility rates in polar and semi-polar solvents, as well as improving the bioavailability of the active ingredient. Low temperature of 4°C is best to stabilize the particle size of nanocurcumin and nanogingerol emulsion.

References
[1] Devarajan V and Ravichandran V 2011 Nanoemulsions: As Modified Drug Delivery Tool. Int J Pharm Globale (IJCP) 4
[2] Pranowo D, Noor E, Haditjaroko L and Maddu A 2015 The ethanol extract nanoemulsion production of Abelmoschus manihot L. Medic by the combination of homogenization and solvent displacement technique International Journal of Sciences: Basic and Applied Research (IJSBAR) 24
[3] Usmiti S, Mangunwidjaja D, Noor E, Richana N and Prangdimurti E 2016 Fabrication of nanoparticles of sulfated low methoxyl pectin by cross-linking reaction on ionotropic gelation technique International Journal of Bio-Science and Bio-Technology (IJBSBT) 8
[4] Baskar V, Selvakumar K, Madhan R, Srinivasan G, Muralidharan M 2012 Study on Improving Bioavailability Ratio of Anti-inflammatory Compound from Ginger Through Nano Transdermal Delivery Asian J of Pharm and Clin. Research 5
[5] Koocheki, A, Kadkhodae R 2011 Effect of alsyssum homolocarpum seed gum, Tween 80 and NaCl on droplets characteristics, flow properties and physical stability of ultrasonically prepared corn Oil-in-Water emulsions Food Hydrocoll 25
[6] Lanimarta Y 2012 Pembuatan dan Uji Penetrasi Nanopartikel Kurkumin-Dendrimer Poliamidoamin (Pamam) Generasi 4 dalam Sediaan Gel dengan Menggunakan Sel Difusi Franz [Skripsi]. (Depok (ID): Universitas Indonesia)
[7] Hasyim A A 2012 The Delivery of Nanoparticles Jeneza Trdine 9, 51000 Rijeka Croatia
[8] Fingas M 2008 Oil spil dispersion stability and oil re-surfacing [Internet]. [Download 2015 Jan 15]. Available at: http://www.iosc.org/papers/2008%20111.pdf
[9] Chou D K, Krishnamurthy R, Randolph T W, Carpenter J F, Manning M C 2005 Effects of Tween 20 and Tween 80 on the stability of Albutropin during agitation. J Pharm Sci 94 1368–81
[10] Yousef S 2012 Evaluation of Antioxidants Stability by Thermal Analysis and Its Protective Effect in Heated Edible Vegetable Oil. Ciência e Tecnologia de Alimentos ISSN 0101-2061
[11] Kusumawardhani A N 2006 Kajian Penambahan Antioksidan terhadap Mutu Simplisia Temulawak (Curcuma xanthorrhiza Roxb.) [Skripsi] (Bogor (ID): Institut Pertanian Bogor)
[12] Shinoda K, Saito H 1969 The Stability of O/W Type Emulsions as Functions of Temperature and the HLB of Emulsifiers: The Emulsification by PIT- method J Colloid and Interf Sci, 30 258-63
[13] Hadiwiyoto S 2011 Product Meat Emulsion [Internet]. Download 2015 Jan 15. Available at: httpfoodreview.co.id.
[14] McClement D J 2011 Edible Nanoemulsion: Fabrication, Properties and Functional Performance. Journal Soft Matter 7 2297-316
[15] Herrera ML 2012 Analytical Technique for Studying the Physical Properties of Lipid Emulsion; Nano and Micro Food Emulsions. Springer