JURNAL ILMU KEFARMASIAN INDONESIA
Indonesian Journal of Pharmaceutical Sciences
ISSN p-ISSN 1693-1831 (Print) e-ISSN 2614-6495 (Online) PUBLISHER : Faculty of Pharmacy Universitas Pancasila

The utilization of red dragon fruit peel extract (*Hylocereus polyrhizus*) in nanoemulsion as an antioxidant

Eriska Agustin[1*], Ega Zahara[1]

[1]*Department of Pharmacy, Faculty of Pharmacy, Universitas Kader Bangsa, Palembang, 30253, Indonesia*

*Corresponding Author: eriska.agustin@ukb.ac.id

Received: 12 June 2024 / Accepted: 25 April 2025

ABSTRACT: The red dragon fruit plant (*Hylocereus polyrhizus*) contains bioactive substances, one of which functions as an antioxidant from anthocyanin and betacyanin. The Dragon Fruit Peel (DFP) has not been widely used by the community in the health sector, and it is only becomes waste. The preparation of nanoemulsion aims to increase the low antioxidant activity of dragon fruit peels. The preparation of DFP nanoemulsion using 8% Tween 80 as emulsifier, 1% oleic acid, and 2% transcutol, with variations in extract concentrations of 1% and 2%. The globule size is reduced to nanoemulsion using a sonicator bath for 60 minutes. The testing of antioxidant activity using the DPPH method. The results of the organoleptic test showed that the DFP extract nanoemulsion had a transparent yellow color with a globule size and Polidispersity Index (PI) of 165±5.54 nm (PI=0.2±0.03) and 115±3.46 nm (PI=0.3±0.02) at an extract concentration of 1 % and 2%. The globule morphology using TEM shows a spherical shape. The results of the pH test showed a range of 4.5-5.3. In testing the antioxidant activity, the DFP extract nanoemulsion showed an increase in IC_{50} values at 1% and 2% extract concentrations, namely 85.39±4.69 and 64.46±3.45, compared to the extract without formula (pure extract), namely 167.34±8.98. The DFP nanoemulsion preparations produced globule sizes in the nano range, and antioxidant activity testing showed an increase in the IC_{50} value of 2 concentrations compared to conventional extracts ($p<0.05$).

KEYWORDS: Anthocyanin; betacyanin; DPPH; IC_{50}; nanotechnology.

INTRODUCTION

Red dragon fruit (*Hylocereus polyrhizus*) is rich in bioactive compounds such as anthocyanins and betacyanins, which exhibit strong antioxidant activity. However, the peel, which contains higher concentrations of these compounds than the pulp, remains underutilized. The red dragon fruit plant has been widely consumed by the public because it tastes sweet and has many benefits in the health sector [1]. The benefits of dragon fruit plants in the health sector include lowering blood sugar levels and cholesterol, preventing colon cancer, strengthening kidney function, strengthening brain performance, and increasing body immunity [2]. Red dragon fruit, with its high antioxidant content—including anthocyanins and betacyanins as natural pigments—is believed to bind carcinogenic substances that cause cancer, as supported by Haveni's study which reported an IC_{50} value of 58.35 ppm indicating strong antioxidant activity [3]. Anthocyanin is a phenolic compound that belongs to the flavonoid class and is water soluble [4]. Red dragon fruit skin also contains a lot of high anthocyanins, but it has not been widely used in the health sector, and often becomes waste. Most people consume only the flesh of the fruit [5]. In a study by Haveni, the anthocyanin level of red dragon fruit skin flesh was 104.58 ppm, higher than that of white dragon fruit skin flesh, 16.73 ppm [3]. Meanwhile, previous studies have shown that the ethanolic extract of red dragon fruit peel exhibits weak antioxidant activity (IC_{50} 329.7 ppm), suggesting the need for formulation improvement. [6].

Given the low antioxidant potency of the peel extract, formulation approaches are needed to enhance its bioactivity, and one promising strategy is the use of nanotechnology-based delivery systems, which have long attracted researchers because materials at the nanoscale exhibit markedly different physicochemical and functional properties compared to their larger-scale forms [7]. One of the applications of nanotechnology in the health sector is the development of nanoemulsion formulations, which have been shown to increase the solubility, stability, and bioavailability of phenolic compounds due to their reduced droplet size and enlarged surface area that enhance dissolution rate, protect against degradation, and improve transmembrane absorption according to various studies. Nanoemulsion is a thermodynamically stable emulsion system with globule sizes in the nanometer range [8]. Encapsulation into nanoemulsion systems has been shown in recent studies to significantly improve the solubility, protect against chemical degradation

How to cite this article: Agustin EA, Zahara EZ. The utilization of red dragon fruit peel extract (*Hylocereus polyrhizus*) in nanoemulsion as antioxidant. JIFI. 2025; 23(2): 211-217.

https://doi.org/10.35814/jifi.v23i2.1614
ISSN: 2614-6495

(light/temperature/pH/oxidation), and increase the bioaccessibility of phenolic compounds including anthocyanins by reducing droplet/particle size, increasing surface area, and providing physical/steric protection during storage and simulated digestion. The formation of nanoemulsion can be done using the self-nanoemulsification (SNE) method. SNE is an isotropic mixture of oil, surfactants, and co-surfactants, which, when mixed with water, will spontaneously form nanoemulsions with light stirring. This system is thermodynamically stable, and the globule size in the nano range ensures high absorption efficiency, both absorption in the digestive tract and the topical route [9]. Antioxidant testing that is commonly carried out uses the DPPH radical scavenging method (2,2-diphenyl-1-picrylhydrazyl) with a mechanism based on electron donors from antioxidants to neutralize DPPH radicals, as seen from the color change of DPPH measured at a wavelength of 515-517 nm. The principle of measuring antioxidant activity quantitatively using DPPH is the change in the intensity of the purple color, which is proportional to the concentration of the DPPH solution. This color change will give a change in absorbance, which is expressed in the IC_{50} value or effective antioxidant concentration, which can reduce 50% of free radicals [10].

Despite the high anthocyanin content in the peel, its poor solubility and low stability limit its antioxidant potential. Therefore, developing a nanoemulsion system could enhance its antioxidant efficacy. In this research, the preparation of DFP extract nanoemulsion was carried out at concentrations of 1% and 2% with the composition of emulsifier, oil phase, and co-emulsifier using a sonicator bath for 60 minutes. This preparation is expected to increase antioxidant activity compared to the extract without the formula, which was tested using the DPPH method. This study aimed to formulate and evaluate a nanoemulsion of red dragon fruit peel extract to enhance its antioxidant activity compared to the conventional extract.

▪ MATERIALS AND METHODS

Materials

The material used in this study was red dragon fruit skin obtained from the plantation of Purworejo Village, Megang Sakti District, Musi Rawas, Indonesia. Other materials used included ethanol p.a (Merck, Germany), Tween 80 (BASF, Germany), oleic acid (Brataco, Indonesia), transcutol (BASF, Germany), aquadeion (Brataco, Indonesia), and DPPH (Sigma Aldrich, Merck, Germany).

Tools

The tools used in this study include an analytical balance (Mettler Toledo® XS205), Heldoph® rotary evaporator RV 10 (Germany), magnetic stirrer (IKA® RW 20 Digital, Germany), sonicator bath (Nagoya S Ultrasonic Cleaner GB-928), pH meter (Mettler Toledo® S20), Particle Size Analizer (Delsa™ Nano C Particle Analyzer, Beckman Coulter, USA), Transmission Electron Microscopy (TEM, JEOL JEM 1400, Japan), UV-Visible spectrophotometry (Beckman® DU 7500i, United States), and laboratory glassware.

The preparation of dragon fruit peel extract

The red dragon fruit skin is dried first, then the dry simplicia is extracted using the maceration method using ethanol p.a solvent with a ratio of simplicia and solvent 1:10 for 3 cycles (1 cycle for 24 hours). The maceration results were carried out by the process of thickening the extract with a rotary evaporator at a temperature of 50 °C [11].

The preparation of dragon fruit peel extract nanoemulsion

Preparation of dragon fruit peel extract nanoemulsion modified from Suciati's research using the SNE method [12]. The following formula is used:

Table 1. Formulation of nanoemulsion DFP.

Ingredients	Concentration (%)		
	F0	F1	F2
The DFP Extract	0	1	2
Tween 80	8	8	8
Transcutol	1	1	1
Oleic Acid	2	2	2
Aquadeion	ad 100	ad 100	ad 100

The process of making nanoemulsion with the oil in water system preparations was carried out by first homogenizing the DFP extract, oleic acid, and Tween 80 with a magnetic stirrer for 15 minutes at 500 rpm. Next, the mixture was added with transcutol and homogenized again for 15 minutes. After all the mixtures were homogeneous, globule size reduction was carried out using a sonicator bath for 60 minutes. The formed nanoemulsion was diluted with distilled water and homogenized for 15 minutes with a magnetic stirrer.

The evaluation of dragon fruit peel extract nanoemulsion

Globule size test

Globule size testing was carried out using a Particle Size Analyzer (PSA). The sample used as much as 1 g of the oil phase of the nanoemulsion extract was dispersed into 5 mL of aquadeion and put in a cuvette with a refractive index of 1.3332 at a temperature of 24.9 ℃/25 ℃ [13].

Globule morphology test

The morphology of the DFP extract nanoemulsion was analyzed using TEM. Before analysis, as much as 1 g of extract was dispersed in 5 mL of deionized water. The mixture was then stirred, and 10 µL was dripped onto the specimen. The 400 mesh grid tool was placed over the specimen containing nanoemulsion droplets and left for 1 minute. The remaining nanoemulsion droplets on the grid were cleaned using filter paper, then 10 µL of uranyl acetate was dripped onto the groid and the remaining droplets were cleaned again using filter paper. The grid was left for 30 minutes to dry and then inserted into the TEM tool to take an image [14].

pH test

The pH test was carried out by dissolving 1 mL of the sample into 10 mL of distilled water with a pH meter. Previously, the pH meter was calibrated using a buffer with a pH of 7, 4, and 9 by inserting the pH meter into the buffer for each pH, and then the nanoemulsion preparation was tested [15].

Antioxidant activity test

The determination of the antioxidant activity of extracts and DFP nanoemulsion preparations using DPPH (2,2-diphenyl-1-picrylhydrazyl) using the Blois (1958) method, as described by Fidrianny with several modifications [14]. The preparation begins with the preparation of 50 ppm DPPH stock solution as a free radical reagent. The standard solution or positive control used was ascorbic acid with varying concentrations of 0.5-2.5 ppm, the DFP extract solution was varied in concentration from 50-450 ppm, and the DFP nanoemulsion preparation solution was varied in concentration from 30-100 ppm for F1 and F2 . Testing the DPPH damping activity on samples (extracts and DFP nanoemulsion preparations) and standard solutions using a 1:1 ratio, which were incubated at 25℃ for 30 minutes in a dark light-tight vial. The absorption absorption was measured using a UV-visible spectrophotometer at a wavelength of 517 nm.

The percentage of inhibition is calculated by the following formula:

$$DPPH\ Inhibition\ Percent = \frac{control\ absorbance - sample\ absorbance}{control\ absrobance} x\ 100\%$$

Data analysis

All data shown is presented in the form of an "mean ± SD". Data collection was carried out in triplo (n=3) and statistical analysis using Anova One-Way Test was carried out for each experiment.

RESULTS

Globule size test

In this study, the DFP is formulated in a nanoemulsion preparation because the extract obtained from the maceration extraction process using ethanol p.a. produces a thick oil extract, so DFP is made in a nanoemulsion formulation. The color is transparent/clear (see Figure 1) and there is no precipitate on the preparation due to the utilization of nanotechnology by reducing the size of the globule with optimal emulsifier and co-emulsifier composition with a globule size of 165±5.54 nm (PI = 0.2±0.03) and 115±3.46 nm (PI: 0.3±0.02) at 1% and 2% formula.

Figure 1. The result of Nanoemulsion DFP, base (left), F1 (middle), F2 (right).

Globule morphology test

Polydispersity Index (PI) value of below 0.5 indicating that nanoemulsions DFP have the homogeneous dispersion system, so it could have a better stability. Figure 2 shows that the oil globules in the nanoemulsion are evenly distributed.

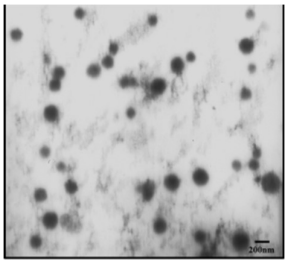

Figure 2. The Morphology of nanoemulsion globule DFP using TEM (magnification 40.000 times).

Antioxidant activity test

The results of testing the antioxidant activity of the nanoemulsion preparations compared to the DFP extract showed a significant decrease in the IC_{50} value of the nanoemulsion preparations ($p<0.05$).

Table 2. The result of antioxidant activity testing.

Sample Name	The IC_{50} value (ppm)
Vitamin C (positive control)	2.76±0.056
Ekstract DFP	299.34±34.56
F1 Nanoemulsion DFP 1%	85.39±4.69
F2 Nanoemulsion DFP 2%	64.46±3.45

(n=3±SD)

DISCUSSION

The extract was prepared using the maceration method with 96% ethanol solvent to produce a viscous oil purple-brown extract, a sticky texture, and an extract yield of 18.985%. The purpose of making DFP viscous extract using 96% ethanol is because antioxidant compounds such as anthocyanins are soluble in polar solvents and are able to extract more other phenolic compounds [16]. In the process of making nanoemulsion with a concentration of 1% and 2% DFP extract, it produces a yellow transparent preparation, without precipitate, and has a distinctive aroma. The color is transparent/clear and there is no precipitate on the

preparation due to the utilization of nanotechnology by reducing the size of the globule with optimal emulsifier and co-emulsifier composition 0.03) and 115±3.46 nm (PI= 0.3±0.02) at 1% and 2% formula. These results prove that the DFP nanoemulsion preparation enters the nano range, namely <1000 nm. The difference in globule size can be caused by the addition of extract concentration with a fixed emulsifier and co-emulsifier composition [17].

The globule size that becomes nano can be affected during the homogenization or mixing process using a magnetic stirrer, emulsifier and co-emulsifier composition, and globule size reduction process with a sonicator bath.

The composition of the DFP nanoemulsion preparation uses Tween 80 emulsifier and transcutol as co-emulsifiers. Tween 80 is a non-ionic surfactant with a steric inhibition mechanism. The mechanism of action of a nonionic emulsifier is that when absorbed at the interface between oil and water, the hydrophobic (water-insoluble) part of the emulsifier interacts with oil, while the hydrophilic (water-soluble) emulsifier will interact with water. The hydrophilic interaction of the interfacial water is able to form a layer that will hinder the interaction between oil droplets with each other or an electric double layer so that the oil droplets do not coalesce into one or become large droplets [18]. The use of oleic acid as the oil phase has the ability to form nanoemulsions and has a high dissolving capacity for oil. The use of transcutol can help solubilize hydrophilic emulsifiers and active substances in oil bases by filling in the surface gaps of the globules so that they are denser so that the surface tension can decrease [19].

The sonication method utilizes mechanical vibrations resulting from ultrasonic waves which cause cavitation. During the sonication process vapor bubbles will appear which can burst violently at a certain crunch size. The bursting of these vapor bubbles creates a very high energy that can make micro-sized particles break into globules [20]. Formula F1 and F2 have a polydispersity index value below 0.5 which indicates the uniformity of globule size in the preparation. The lower the polydispersity index value, the higher the globule size uniformity [21].

The results of the TEM analysis showed that the nanoemulsion globules of DFP F2 extract had a globule size of less than 200 nm with a spherical spherical shape. The Figure 2 shows that the oil globules in the nanoemulsion are evenly distributed. This is consistent with the results of the low polydispersity index of the DFP nanoemulsion extract (<0.05).

The examination of the pH value of the nanoemulsion preparations resulted in a pH value of F1 in the range 6.8-7.36 and F2 in the range 6.73-7.21. These results were obtained from pH testing from day 1-28. From the test results, there was no significant difference between the pH of F1 and F2 tested for 28 days ($p>0.05$).

The determination of antioxidant activity using the DPPH method has been widely used as a preliminary method because it is relatively easy and simple. The results of the antioxidant activity test obtained the Inhibitory Concentration (IC_{50}) value which indicates the concentration of a substance that can inhibit 50% of free radicals.

The results of testing the antioxidant activity of the nanoemulsion preparations compared to the DFP extract showed a significant decrease in the IC_{50} value of the nanoemulsion preparations ($p<0.05$). The principle of antioxidant testing using the DPPH method is a change in the intensity of the purple color of DPPH as a radical. The DPPH radical has an unpaired electron which is due to the quenching of free radicals produced by the reaction of the DPPH molecule with the hydrogen atom released by the sample compound molecule so that a diphenylpicryl hydrazine compound will be formed [14]. DFP nanoemulsion preparations were able to increase antioxidant activity 3.5 times for F1 nanoemulsion preparations, namely 85.39±4.69 ppm and 4.6 times for F2 nanoemulsion preparations, namely 64.46±3.45 ppm compared to conventional extracts DFP, namely 299.34±34.56 ppm, was included in the low antioxidant category (> 100 ppm) (see Table 2). The antioxidant activity of the DFP nanoemulsion is included in the strong antioxidant category (50-100 ppm). The increase in antioxidant activity of the DFP nanoemulsion preparations can be caused by several things, one of which is the use of oleic acid as a 2% oil base which can act as an antioxidant at certain concentrations [16]. In addition, the anthocyanin content in the extract plays a role in antioxidant activity due to its high redox potential and is a strong reducing agent [22].

The increase in antioxidant activity in nanoemulsion preparations can also be due to the extract being dispersed in an oil base of oleic acid and emulsifier tween 80 so that the increased surface area will increase

and accelerate contact with DPPH free radicals [21]. The scavenging of DPPH free radicals with hydrogen atoms from compound molecules in DFP nanoemulsion preparations will also increase so that it will increase the percentage of free radical inhibition expressed in the IC_{50} value [14].

Based on the results of statistical tests, testing the antioxidant activity of conventional DFP extract and nanoemulsion preparations there was a significant difference in the IC_{50} value ($p<0.05$), while there was no significant difference in the IC_{50} values of F1 and F2 ($p>0.05$). From the manufacture of DFP nanoemulsion preparations, it can be said that there is an increase in antioxidant activity compared to conventional DFP extracts.

▪ CONCLUSION

The results of the evaluation of the manufacture of DFP nanoemulsion preparations obtained the globule size values of F1 and F2 entering the nano range, pH evaluation obtained a pH that was included in the pH range of 6-7. Globule morphology testing produces a spherical spherical shape. Testing the antioxidant activity of the DFP F1 and F2 nanoemulsion preparations resulted in an IC_{50} value that was included in the strong antioxidant category and an increase in the antioxidant activity of the DFP nanoemulsion preparations compared to conventional DFP extracts.

Acknowledgements: The authors and researchers would like to thank all parties involved in the research and in writing this journal, and to the academic community at Kader Bangsa University Palembang.

Funding: This research received no external funding.

Conflict of interest statement: This research has no conflict of interest.

REFERENCES

[1] H. Hermansyah, A. Febrianto, H. Hermansyah, dan F. Barchia, "Growth response of red dragon fruit (Hylocereus costaricensis) stem cuttings to concentration and duration of immersion in young coconut water," J. Ilmu-Ilmu Pertanian Indonesia, vol. 21, no. 1, pp. 22–26, 2019.

[2] K. Y. S. Ni, "Morphological structure of flowers and pollen grain anatomy of super red dragon fruit (Hylocereus costaricensis)," J. Media Sains, vol. 1, no. 2, pp. 71–76, 2017.

[3] D. Haveni, Mastura, dan R. P. Sari, "Ethanol extract of super red dragon fruit (Hylocereus costaricensis) peel as an antioxidant using the DPPH method," Katalis J. Pendidik. Kimia dan Ilmu Kimia, vol. 2, no. 2, pp. 30–37, 2019.

[4] S. Q. Putridhika, D. Ratnasari, dan V. A. Gatera, "Antioxidant activity test of lip balm preparation from red dragon fruit peel (*Hylocereus polyrhizus*)," J. Pendidik. dan Konseling, vol. 4, no. 5, pp. 5845–5851, 2022.

[5] T. Aryani dan I. A. U. Mu'awanah, "Antioxidant activity and vitamin C content of fruit flesh and syrup of dragon fruit (Hylocereus costaricensis)," Biomedika, vol. 12, no. 2, pp. 149–157, 2019.

[6] W. Utami, E. Mardawati, dan P. H., "Antioxidant activity testing of red dragon fruit peel (*Hylocereus polyrhizus*) as a peel-off gel mask," J. Industri Pertanian, vol. 2, no. 1, pp. 95–102, 2020.

[7] D. Thassu, M. Deleers, dan Y. Pathak, Nanoparticulate drug delivery systems. New York: Informa Healthcare, 2007.

[8] S. V. R. Rao dan J. Shao, "Self-nanoemulsifying drug delivery systems (SNEDDS) for oral delivery of protein drug: formulation development," Int. J. Pharm., vol. 362, pp. 2–9, 2008.

[9] A. U. Kyatanwar, K. R. Jadhav, dan V. J. Kadam, "Self micro-emulsifying drug delivery system (SMEDDS): review," J. Pharm. Res., vol. 3, no. 83, pp. 75–83, 2010.

[10] F. Shahidi dan Y. Zhong, "Measurement of antioxidant activity," J. Funct. Foods, vol. 18, pp. 757–781, 2015.

[11] Widyastuti, "Formulation of topical preparation of dragon fruit peel extract," J. Iptek Terapan, vol. 13, no. 1, pp. 20–33, 2017.

[12] T. Suciati, A. Aliyandi, dan Satrialdi, "Development of transdermal nanoemulsion formulation for simultaneous delivery of protein vaccine and artin-M adjuvant," Int. J. Pharm. Pharm. Sci., vol. 6, no. 6, pp. 536–546, 2014.

[13] W. A. Ningrum, Y. W. Permadi, F. F. Himmah, dan F. Ulfa, "Evaluation of lotion preparation containing nanoparticle extract of terong belanda as an antioxidant," J. Media Sains, vol. 14, no. 1, pp. 99–104, 2021

[14] I. Fidrianny, N. A. S. Anggraeni, dan M. Insanu, "Antioxidant properties of peels extracts from three varieties of banana (*Musa sp*.) grown in west java-Indonesia," Int. Food Res. J., vol. 25, no. 1, pp. 57–64, 2016.

[15] M. D. Y. Ambarita, E. S. Bayu, dan H. Setiado, "Identification of morphological characteristic of banana (*Musa spp*.) in Deli Serdang district," J. Agroekoteknologi, vol. 4, no. 586, pp. 1911–1924, 2015.

[16] N. S. Mathew dan P. S. Negi, "Traditional uses, phytochemistry and pharmacology of wild banana (*Musa acuminata Colla*): A Review," J. Ethnopharmacol., vol. 10, p. 10605, 2016.

[17] M. Jaiswal, R. Dudhe, dan P. K. Sharma, "Nanoemulsion: an advanced mode of drug delivery system," Biotech, vol. 5, no. 2, pp. 123–127, 2015.

[18] G. Zhang et al., "Enhanced stability of vitamin a palmitate microencapsulated by gamma-cyclodextrin metal-organic frameworks," J. Microencapsulation, vol. 35, pp. 249–258, 2018.

[19] E. Agustin, M. Insanu, dan R. Mauludin, "Improvement in pharmacological activity of mahkota dewa (*Phaleria macrocarpa (Schef. Boerl)*) seed extracts in nanoemulsion dosage form: in vitro and in vivo studies," Pharm. Nanotechnol., vol. 12, no. 1, pp. 90–97, 2023. http://dx.doi.org/10.2174/2211738511666230602100045

[20] I. Fidrianny, Y. Johan, dan Sukrasno, "Antioxidant activities of different polarity extracts from three organs of makrut lime (*Citrus hystrix DC*) and correlation with total flavonoid, phenolic, carotenoid content," Asian J. Pharm. Clin. Res., vol. 8, no. 4, pp. 239–243, 2015.

[21] G. Chhabra, K. Chuttani, A. K. Mishra, dan K. Pathak, "Design and development of nanoemulsion drug delivery system of amlodipine besilate for improvement of oral bioavailability," Drug Dev. Ind. Pharm., vol. 37, no. 8, pp. 907–916, 2011.

[22] S. A. Devi, "Aging brain: prevention of oxidative stress by vitamin E and exercise," Sci. World J., vol. 9, pp. 366–372, 2009.