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ANTHOCYANIN ISOLATION METHOD OF RED DRAGON FRUIT *(Hylocereus polyrhizus)*: A REVIEW

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ABSTRACT

Background: The body needs antioxidants to reduce the number of free radicals in the body. The anthocyanin compounds in red dragon fruit can be a source of antioxidants to counteract free radicals. Many types of solvents can be used to extract anthocyanins, such as water, methanol, or ethanol. The extracts produced from this process usually contain solvents that can reduce the anthocyanin content in the extract. **Objective**: Researchers believe that a literature review is needed to find out the factors that affect the isolation of anthocyanin compounds from dragon fruit skin, such as extraction methods, types of solvents, temperatures, and pH. **Methods**: The methods used in this study were obtained from Google, Google Cendekia, and Pubmed using keywords such as anthocyanin isolation and red dragon fruit. The article used was selected in the range of 10 years, from 2014 to 2024. The inclusion criteria in the search for literature are the articles that explain the anthocyanin isolation method of red dragon fruit and articles are available in Indonesian or English, while the exclusion criteria in this study are articles that are an article review. **Results**: The methods of maceration, Ultrasonic Assisted Extraction, Microwave Assisted Extraction, and Microwave Assisted Hydro Distillation can be used to isolate anthocyanin compounds using solvents such as citric acid, ethanol, and acetic acid. **Conclusion**: The ultrasonic assistance method is known to be able to attract the most anthocyanin compounds from red dragon fruit compared to other methods, which is 29,640 mg/5 g. The solvent with the acid environment is more suitable for extracting anthocyanin compounds. Prolonged extraction time can cause a decrease in anthocyanin content, which is associated with anthocyanin degradation by citric acid. The optimal time is 45 minutes with the UAE method.

Keywords: Anthocyanin; Isolation method; Red dragon fruit

INTRODUCTION

Our body cells create ROS (Reactive Oxygen Species), commonly referred to as free radicals, during metabolism. Free radicals are produced by immune cells to destroy viruses and bacteria.[1] Environmental elements like radiation, cigarette smoke, pollution, and pesticides can also create free radicals. The organism generally tolerates free radicals, but oxidative stress results from either an overabundance of free radicals or a lack of antioxidants.[2] Environmental elements like radiation, cigarette smoke, pollution, and pesticides can also produce free radicals. The organism generally tolerates free radicals, but oxidative stress results from either an overabundance of free radicals or a lack of antioxidants.[3] Thus, studies on antioxidants derived from natural sources must be carried out.

An inexpensive alternative source of antioxidants that can enhance the quality

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of public health is natural materials.[4] Certain sections of some plants, including the red dragon fruit, naturally produce antioxidants. The plant known as pitaya, or red dragon fruit (*Hylocereus polyrhizus*), is native to arid tropical areas. One of the most well-liked fruits, it is grown extensively in several tropical nations. In the skin of red dragon fruit, compounds from the betalain, anthocyanin, vitamin E, and vitamin C groups can be found. Red dragon fruit contains anthocyanin chemicals, which can act as antioxidants to protect against free radicals.[5]

Anthocyanins are natural pigments that give red to purple colors to various parts of plants, including fruits and flowers. Anthocyanins are a type of flavonoid consisting of three carbon atoms bound by an oxygen atom that serves to connect two aromatic benzene rings. (C_6H_6). In addition to its function as a natural dye, anthocyanins are also known to have strong antioxidant properties. Some benefits of anthocyanins are that they are anti-inflammatory and anti-cancer, and they protect the cardiovascular system. A compound is said to be a very strong antioxidant if the IC_{50} value is less than 50, strong (50-100), moderate (100-150), and weak (151-200). The smaller the IC_{50} value, the higher the antioxidant activity.[6]

Many types of solvents can be used to extract anthocyanins, such as water, methanol, or ethanol. Factors such as excessively high extraction temperatures, prolonged extraction times, exposure to light and oxygen, and variations in the pH of the extraction environment can contribute to the degradation of anthocyanin compounds.[7]

Based on this background, the researchers believe that they need to conduct a literature review to gather literature to understand the factors influencing the isolation of anthocyanin compounds from dragon fruit peel, such as method factors, solvent factors used, temperature factors, and pH factors. This literature review can serve as a reference for the development of anthocyanin isolation methods in the future.

METHODS

The methods used in this research were obtained from various library and literature sources. The data excavation process was carried out through searches from reliable literature sources such as Google, Google Scholar, and PubMed by looking for articles on anthocyanin isolation and antioxidant tests from red dragon fruit. The keywords used in the literature search were anthocyanin isolation, red dragon fruit, and antioxidant. The search data were then further selected within a 10-year range from 2014 to 2024. Several inclusion criteria in the literature search were that the articles were in Indonesian or English, the articles were fully available, and the articles explained the methods of anthocyanin isolation from red dragon fruit. In contrast, the exclusion criteria were literature reviews in the form of articles.

RESULTS

Based on the search results from the literature sources, 14 scientific articles were obtained. It was found that the obtained articles used various methods and solvents to isolate anthocyanins, one example being maceration, UAE (Ultrasonic Assisted Extraction), MAE (Microwave Assisted Extraction), and MAHD (Microwave Assisted Hydro Distillation).

Out of the 14 articles used, 10 articles employed maceration as the method for extracting anthocyanins from red dragon fruit peel powder samples.

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Table 1. Results of the Review of the Maceration Method of Red Dragon Fruit Peel Powder

Method	Preparation	Instrument	Result	Reference
Maceration	100 g red dragon fruit peel powder with aquadest and 10% citric acid (1:6) at room temperature for 3 x 24 hour with no stiring	Spectrophotometer UV-Vis with λ = 520.0 nm	The anthocyanin concentration (%) result is 30.4% b/v	[8]
	100 g red dragon fruit peel powder with 50 mL of 0.4 M citric acid solvent at a temperature of 20°C for 2 hours with no stiring	Spectrophotometer UV-Vis with λ = 510.0 nm and 700.0 nm	The anthocyanin content obtained was 12.447 mg/L	[9] [10]
	100 g red dragon fruit peel powder with 50 mL of 0.4 M citric acid solvent at a temperature of 20°C for 3 hours with no stiring	Spectrophotometer UV-Vis with λ = 510.0 nm and 700.0 nm	The anthocyanin content obtained was 1.225 mg/L	[10]
	10 g red dragon fruit peel powder with 30 mL aquadest and 10% citric acid at room temperature for 4 days with 30 second of stirring once	Spectrophotometer UV-Vis with λ = 510.0 nm and 700.0 nm	The anthocyanin content obtained was 4.73 mg/L	[11]
	10 g red dragon fruit peel powder with 40 mL aquadest and 10% citric acid at room temperature for 4 days with 30 second of stirring once	Spectrophotometer UV-Vis with λ = 510.0 nm and 700.0 nm	The anthocyanin content obtained was 2.40 mg/L	[11]
	50 g red dragon fruit peel powder with aquadest and 2% citric acid at room temperature for 2 x 24 hours with no stiring	Spectrophotometer UV-Vis with λ = 535.0 nm	The anthocyanin content obtained was 6.38±0.44 mg/50 g	[12]
	Red dragon fruit peel powder with ethanol and 10% citric acid (5:1) at room temperature for 24 hours with no replication and no stirring	Spectrophotometer UV-Vis with λ = 498.6 nm	The maximum anthocyanin content was obtained using a 10% ethanol and citric acid solvent	[13]
	Red dragon fruit peel powder with aquadest and 10% citric acid (1:6) at room temperature for 3 days with no stirring	Spectrophotometer UV-Vis with λ = 500.0 nm – 550.0 nm	The yield of anthocyanins was obtained, which is 62.68%	[14]
	50 g red dragon fruit peel powder with aquadest and 2% citric acid for 2 x 24 hours at room temperature with no stirring	Spectrophotometer UV-Vis with λ = 535.0 nm	The total anthocyanin content obtained is 38.33 mg/100 g	[15]
	20 g red dragon fruit peel powder with 96% Ethanol solvent at a speed of 15 rpm for 60 minutes at room temperature	Spectrophotometer UV-Vis by calculating %yield with λ = 530.0 nm	The %yield obtained is 0.000275%.	[16]
	25 g red dragon fruit peel powder with 0,5 M citric acid solvent stirred for 150 minutes at room temperature	Spectrophotometer UV-Vis with λ = 510.0 nm and 700.0 nm	The anthocyanin content obtained is 11.439 mg/L.	[17]

Solvents that can be used for extraction include aquadest, nitric acid, and ethanol. The calculation of anthocyanin content obtained from the extraction results is performed using a UV-Vis spectrophotometer.

Out of the 14 articles used, two articles employed ultrasonic-assisted extraction (UAE) as the method for extracting anthocyanins from red dragon fruit peel powder samples. Solvents that can be used for extraction include

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aquadest, nitric acid, and ethanol. The calculation of anthocyanin content obtained from the extraction results is performed using a UV-Vis spectrophotometer. Out of the 14 articles used, 1 article employed microwave-assisted extraction (MAE) as the method for extracting anthocyanins from red dragon fruit peel powder samples. Solvents that can be used for extraction include aquadest and nitric acid. The calculation of anthocyanin content obtained from the extraction results is performed using a UV-Vis spectrophotometer.

Out of the 14 articles used, 1 article employed maceration as the method for extracting anthocyanins from red dragon fruit peel powder samples. Solvents that can be used for extraction include aquadest and ethanol. The calculation of anthocyanin content obtained from the extraction results is performed using a UV-Vis spectrophotometer.

Table 2. Results of the Review of the Ultrasonic Assisted Extraction Method of Red Dragon Fruit Peel Powder

Method	Preparation	Instrument	Result	Reference
Ultrasonic Assisted Extraction (UAE)	5 g red dragon fruit peel powder with aquadest and 10% citric acid solvent for 45 minutes and 65% amplitude	Spectrophotometer UV-Vis with λ = 510.0 nm and 700.0 nm	The anthocyanin content obtained is 29.640 mg/L	[18]
	Red dragon fruit peel powder with ethanol, aquadest, and acetic acid (80:19:1) solvent for 15 minutes	Spectrophotometer UV-Vis with λ = 520.0 nm and 700.0 nm	The anthocyanin content obtained is 0.0241 mg/L	[15]

Table 3. Results of the Review of the Microwave-Assisted Extraction Method of Red Dragon Fruit Peel Powder

Method	Preparation	Instrument	Result	Reference
Microwave Assisted Extraction (MAE)	100 g red dragon fruit peel powder with aquadest and citric acid solvent (4:1) for 6 minutes	Spectrophotometer UV-Vis with λ = 535.0 nm	The anthocyanin content obtained was 28.11 mg/100 mg extract	[19]

Table 4. Results of the Review of the Microwave Assisted Hydro Distillation Method of Red Dragon Fruit Peel Powder

Method	Preparation	Instrument	Result	Reference
Microwave Assisted Hydro Distillation (MAHD)	100 g red dragon fruit peel powder with ethanol and aquadest solvent (4:1) for 4 minutes	Spectrophotometer UV-Vis with λ = 510.0 nm and 700.0 nm	The anthocyanin content obtained was 52.184 mg/100 g sample	[20]

DISCUSSION

Based on the search results from the literature sources, 14 scientific articles were obtained. It was found that the obtained articles used various methods and solvents to isolate anthocyanins, one example being maceration, UAE (Ultrasonic Assisted Extraction), MAE (Microwave Assisted Extraction), and MAHD (Microwave Assisted Hydro Distillation).

A) Maceration Method

Maceration is a method of separating a compound that involves soaking it in an organic solvent at a specific temperature.

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The maceration process is considered advantageous for isolating a natural compound due to its low cost and ease of execution. The maceration technique relies on pressure changes inside and outside the cell to break the cell wall and membrane, dissolving secondary metabolites in the cytoplasm.[21]

The amount of solvent used is one of the factors in the amount of compounds extracted in maceration. According to the research results in Table 1, maceration using 30 mL of citric acid and aquadest produced an anthocyanin content of 4.73 mg/L, while maceration using 40 mL of citric acid and aquadest produced an anthocyanin content of 2.40 mg/L. The addition of citric acid gives the solvent acidic properties. Under acidic conditions, plant cell membranes can denature, causing the pigments present in the plants to dissolve more easily into the solvent, making the compounds easier to extract.[13, 22]

Based on the results of the literature study, the more solvent volume used for maceration, the lower the concentration of anthocyanin compounds obtained from the maceration process. The decrease in anthocyanin concentration occurs because the cyanidin groups in the red dragon fruit skin have been extracted by the citric acid solvent, resulting in a lower anthocyanin concentration. so that with the increase in volume, the amount of anthocyanin obtained does not increase. This is in accordance with the extraction theory, which states that the concentration of the product decreases with the amount of solvent present.[23]

The acidity level of the solvent also affects the extraction of anthocyanins because anthocyanin compounds are pH-sensitive. In the extraction using citric acid, higher yields were obtained compared to the use of ethanol. This is because anthocyanins are more stable in acidic solutions compared to alkaline or neutral solutions. In acidic solutions, anthocyanins are stable, and in strong acidic solutions, anthocyanins are very stable, whereas in neutral and basic solutions, anthocyanins become unstable. In a solvent with a high pH, anthocyanins will undergo deprotonation, which weakens the bonds within the molecule, making it more susceptible to degradation.[9]

The duration of the maceration treatment also affects the concentration of anthocyanin compounds extracted during maceration. According to the research results in Table 1, maceration for 2 hours produced an anthocyanin content of 12.447 mg/L. On the other hand, maceration for 3 hours resulted in an anthocyanin content of 1.225 mg/L. The anthocyanin content decreases as the extraction time increases.

Based on the literature review, the longer the extraction time, the higher the amount of anthocyanin compounds obtained. This is because the opportunity for contact between the material and the solvent increases, resulting in a greater yield. However, the increase in extracted compounds will stop when the material reaches saturation point. The decrease in anthocyanin content occurs due to the degradation of anthocyanins, causing the contained anthocyanin levels to decrease. An extraction time that is too long will cause the extract to hydrolyze, while an extraction time that is too short will result in not all active compounds being extracted from the material.[14, 18]

B) Ultrasonic Assisted Extraction (UAE) Method

Ultrasonic-assisted extraction (UAE) is a promising extraction method because it yields higher yields and shorter processing times. UAE has the principle of acoustic cavitation, which can damage plant cell walls, allowing bioactive compounds to be released into the environment. UAE has been proven to improve extraction efficiency and extraction time. Additionally, UAE can also be performed at lower temperatures

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to avoid damage due to heating.[18, 24]

The use of aquadest and citric acid as solvents in Table 2 has proven to be more effective in extracting anthocyanin compounds, with a yield of 29.640 mg/5 g in the study. In contrast, UAE using ethanol, aquadest, and acetic acid yielded an anthocyanin content of 0.0241 mg/30 g. Compared to maceration, UAE produced a higher yield, with maceration yielding an anthocyanin content of 38.33 mg/100 g. This is because UAE utilizes ultrasonic waves to enhance the penetration of the liquid into the cell walls, resulting in a faster mass transfer rate and, consequently, a higher anthocyanin content.[25]

C) Microwave Assisted Extraction (MAE)

A new method that has received attention in recent times to be developed in the isolation of active compounds, the name of this new method is microwave-assisted extraction (MAE). This method has advantages over conventional extraction methods, namely the presence of molecular transfer involving microwave energy. The MAE method has the advantage of obtaining extraction yields quickly, as microwave energy causes molecular movement through ion migration and dipole rotation. This very rapid movement generates friction, which ultimately produces thermal energy within the material, causing the cell walls or tissue to break down allowing the solute to be released.[26]

A mixture of aquadest and citric acid, using the MAE method in Table 3, produces an anthocyanin content of 28.11 mg/100 mg, compared to maceration using the same solvent mixture, which yields an anthocyanin content of 4.73 mg/L. This shows that the MAE method is more effective in extracting anthocyanin compounds compared to maceration because MAE uses microwave waves to generate friction, which ultimately produces heat energy in the material, causing the cell walls or tissue to break down, making it easier for the solvent to attract the compounds.[27]

The UAE method yields higher results than MAE, with a solvent of aquadest and citric acid yielding 29.640 mg/L, which is not much different from MAE. The UAE method uses a principle almost identical to MAE, which involves using waves to break down cell walls, allowing more compounds to be extracted. Based on the literature review, MAE (Microwave-Assisted Extraction) is stated to be superior to UAE (Ultrasound-Assisted Extraction) because MAE is capable of extracting a larger amount of compounds compared to UAE under optimized extraction conditions, even when using a longer extraction time. This is evidenced by the UAE requiring 45 minutes, while MAE only takes 6 minutes.[27]

D) Microwave Assisted Hydro Distillation (MAHD)

Microwave Assisted Hydro Distillation (MAHD) is an advancement of Microwave-Assisted Extraction (MAE) that uses electromagnetic waves to heat the solvent. This method is superior because it produces a high extract yield, is easy to use, and is time-efficient. MAHD can be an alternative extraction method for compounds with results similar to those of conventional water distillation. MAHD produces higher anthocyanin content than MAE and maceration, as well as color characteristics that correspond to pH changes.[28]

The MAHD method using ethanol and aquadest solvent (4:1) in Table 4, yields an anthocyanin content of 52.184 mg/100 g. This result is significantly higher compared to maceration, which yields an anthocyanin content of 38.33 mg/100 g. This is because the MAHD method uses microwaves similar to the MAE method to break down the

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compound cell walls, thereby increasing the content. The MAHD method is considered more efficient, requiring only 4 minutes, while maceration requires 2 x 24 hours.[26]

The MAHD method yields higher results compared to the MAE method, which yields 28.11 mg/100 mg. This is because the MAHD method is an improvement of the MAE method with the addition of an integrated distillation process during extraction, resulting in a higher yield.

The results obtained by the UAE method were 29.640 mg/5 g, compared to the MAHD method, where the UAE method yielded a higher concentration. This is because the solvent used in the MAHD method is ethanol. The solvent used in the UAE method is citric acid, which creates an acidic condition in the solvent. This can denature plant cell membranes, making it easier for the pigments in the plants to dissolve into the solvent, thus allowing the compounds to be easily extracted. This results in a lower concentration produced by the MAHD method using ethanol.[9]

CONCLUSION

Factors that influence the success of the anthocyanin isolation of red dragon fruit include the type and volume of the solvent, the length of time the extraction, and the extraction method used. The ultrasonic assistance method (UAE) is the most effective in isolating anthocyanin compounds compared to other methods, with anthocyanin content obtained by 29,640 mg/l. Extraction in acid conditions proved effective in attracting anthocyanin, but the extraction of extraction in acid conditions can cause anthocyanin compounds to degrade. The length of time for anthocyanin extraction in the most optimal acid conditions is 45 minutes with the UAE method. A longer extraction time using an acidic solvent will cause the anthocyanin compounds to degrade and reduce the anthocyanin.

CONFLICT OF INTEREST

There is no conflict of interest in this paper. This article was written independently without affiliation with other parties.

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