Journal of Carbazon
Vol. 02, (01) 2024
DOI: 10.24815/jocarbazon. v2i1.37890
eISSN : 2988-5744

Determination Of Total Flavonoids Extract of White (*Magnolia Alba* (Dc.) Figlar) Using Spectrophotometry Uv-Vis Method

MASTURA[1*], ULIL AMNA[1], SARAH NIACI[1], TIA PEBIOLA[1]

[1]*Chemistry Departement, Faculty of Engineering, Universitas Samudra 24416, Kota Langsa, Aceh, Indonesia*
[*]Corresponding Author E-mail: mastura@unsam.ac.id

Citation: Mastura; U. Amna; S. Niaci; T. Pebiola; Determination Of Total Flavonoids Extract Of White (Magnolia Alba (Dc.) Figlar) Using Spectrophotometry Uv-Vis Method, *J. Carbazon*, 2024, Vol. 2 No.1, 1-9.
DOI:10.24815/jocarbazon. v2i1.37890
Received: 24 May 2024
Accepted: 21 June 2024
Published: 24 June 2024

Abstract: Flavonoids are a group of secondary metabolites produced by plants that have the potential to influence pharmacological activity. This study aims to determine the total flavonoid content of the ethanol crude extract of white cempaka flowers (*Magnollia alba* (DC.) Figlar) using the UV-Vis spectrophotometry method. This study used colorimetric and spectrophotometric methods using quercetin as a comparison. The qualitative test research results proved that cempaka putih (*Magnolia alba* (DC.) Figlar) flowers contain flavonoid coumpound. Then a maximum wavelength of 432 nm is obtained with an absorbance of 0.1236, the absorbance calculation for each quercetin concentration and the sample is calculated using a maximum wavelength of 432 nm. Based on the linear regression equation obtained from the standard quercetin curve, the total flavonoid content of the crude ethanol extract of cempaka putih flowers with 3 different concentration variations, at a concentration of 100 ppm, the results obtained for the total flavonoid content were 276,83 ± 17,7 mgQE/g, 500 ppm. total flavonoid levels of 105,189 ± 8,16 mgQE/g, 1000 ppm obtained total flavonoid levels of 79,115 ± 8,33 mgQE/g.

Keywords: flavonoids, exstract, ethanol, spectrophotometry UV-Vis, *Magnolia alba* (DC.) figlar

INTRODUCTION

Due to Indonesia's favorable climate, a wide variety of plant species are able to thrive there [1]. Modernization links medicinal plants with the pharmaceutical world whose properties are slowly being recognized by the scientific community [2].

Modern medicine has scientific strength, because it has gone through clinical trials but lacks modern medicine which has big side effect. Therefore, people are starting to turn traditional medicine which has less side effects and is used as a medicinal plant [3].

One of the plants that has secondary metabolites that are commonly used as medicinal plants is cempaka putih (*Magnolia alba* (DC.) Figlar) [4]. With a simple process, this flower can treat ailments such as vertigo disorders, flatulence, sinusitas, vaginal discharge, respiratory tract and coughs. But there is still no more indepth research. Another benefit of this flower is a perfume and aromatheraphy. The content of secondary metabolities from white cempaka flowers are alkaloids, flavonoids [5].

Flavonoids are a class of secondary metabolites obtained from plants in the large group of polyphenols. This class of polyphenolic compounds has been known to have beneficial biochemical and antioxidant effects related to diseases such as cancer, Alzheimers, atherosclerosis, and so on [6].

Two substituted benzene rings are joined by a three-carbon alpha-chain to form the carbon skeleton of flavonoids. The grouping of flavonoids is based on an additional heterocyclic oxygen ring and the hydroxyl group of flavonoids has a pyran ring connecting three carbons with one benzene [7].

Based on the findings of the literature review, it can be seen that cempaka putih flowers contain one of the secondary metabolites, namely flavonoids, where flavonoid compounds have various pharmacological activities that can have potential as medicinal raw materials.

METHOD

Sample, Tools and Materials

The sample used in this study was *M. alba* flowers taken in Meurandeh, Langsa City, Aceh, Indonesian

The tools used in this study were maceration vessels, blenders, scissors, rulers, pencils, alumunium foil, watch glass, plastic, jars, glass funnels, measuring flasks, reagent bottles, rotary evaporator HS-2005S, a set of glassware, analytical balance, stir bar, dropper pipette, spatula, test tube rack, gloves, mask, cutter, erlenmeyer, cuvette, micropipette, test tube clamp, suction ball, and Orion Aquamate 8100 UV-VIS Spectrophotometry.

The materials used in this study were 70% ethanol, 10% alumunium chloride, 1 M potassium acetate, quercetin, acetic acid anhydride, Mg powder, chloroform, concentrated sulfuric acid, diethyl ether, hydrochloric acid, filter paper and distilled water.

Sample Preparation

Samples of *M. alba* flowers were taken from the Meurandeh area, Langsa City, Aceh, Indonesian. The samples were separated from the stalks and air dried for 14 days then crushed to widen the contact area during the extraction process

Sample Extraction

1 kg of *M. alba* flowers which had been air-dried were successively macerated with 70% ethanol until the filtrate was clear. The maceration process was carried out for 3x24 hours.

After that the filtrate obtained was filtered, then extracted using a rotary evaporator and weighed the weight of the extract obtained.

Qualitative Analysis of Flavonoid Content
Testing for flavonoids was carried out by putting 2 mL of the extract sample into a test tube, adding a few drops of concentrated HCl and adding a little Mg powder. This result is positive if it produces an orange, dark red, pink or brick red color.

Quantitative Analysis of Flavonoid Content
Preparation of Main Solution (Quercetin 1000 ppm)
Quercetin was weighed as much as 10 mg with an analytical balance. Then quercetin was dissolved with 70% ethanol in a 10 mL volumetric flask up to the mark. for obtain a solution of 1000 ppm quercetin.

Preparation of Quercetin Solution for Making Standard Curves
Pipette the mother liquor as much as 0.5, 1, 1.5, 2, and 2.5 mL each into a 25 mL volumetric flask using a micropipette. Furthermore, 70% ethanol was added up to the limit mark, so that solutions with concentrations of 20, 40, 60, 80, dan 100 ppm were obtained. Pipette 0,5 mL of 70% ethanol, 0.l mL of 10% alumunium chloride, 0.1 mL of 1 M potassium acetate were added and 2.8 mL of distilled water was added until the mark was made three times, then shaken until homogeneous. Each solution was incubated at room temperature for 30 minutes.

Preparation of Sample Solution
To make a sample solution of 1000 ppm, 25 mg of cempaka putih flower extract was weighed, transferred to a 25 mL volumetric flask, and diluted in 70% ethanol to the mark. Dilution was carried out by pipetting a 1000 ppm sample solution of 2.5 and 12.5 mL. Them put each solution into a 25 mL volumetric flask and add 70% ethanol up to the mark, for obtain sample test solutions with concentrations of 100, 500, and 1000 ppm. Pipette each 0.5 mL of solution into a 5 mL volumetric flask, add 1.5 mL of 70% ethanol, 0.1 mL of 10% alumunium chloride, 0.1 mL of 1 M potassium acetate, add 2,8 mL of distilled water. To mark, shaken until homogeneous and made in three repetitions. Incubated at room temperature for 30 minutes.

Absorbance Meansurement of Quercetin Solution and sample Solution Using UV-Vis Spectrophotometry
70% ethanol is determined as a blank. One of the quercetin standards was measured at a wavelength of 400-600 nm, so that the maximum quercetin wavelength was obtained. The absorbance of quercetin standard solutions at concentrations of 20, 40, 60, 80 and 100 ppm, was measured at the maximum wavelength. Then graph the quercetin standard curve by comparing the absorbance (A) to the concentration (C, ppm). After that, the absorbance of the sample solutions at 100, 500, and 1000 ppm was measured at the maximum wavelength. The concentration of quercetin in the sample solution and the total flavonoid content were determined by plotting the absorbance of the sample solution against the graph of the standard quercetin solution.

RESULTS AND DISCUSSION
Sample of *M. alba* flowers were separated from the stalks and airdried to reduce the water content, then the samples were crushed and ground into powder to expand the contact area during the extraction process [8].

The maceration technique was used to extract the dried materials. Since maceration doesn't include any kind of heating procedure, its usage reduces the likelihood that any of the chemical compound's constituents will be damaged in the process. The maceration process was carried out repeatedly to obtain a clear colored filtrate. The purpose of doing this is to maximize the extraction results [9].

The extraction process was carried out by soaking the simplicia powder with 70% ethanol solvent, which aims to remove all secondary metabolite components in the *M. alba* flower sample, because ethanol solvent is as universal solvent that can attract compounds that dissolve in non-polar to polar solvents [10]. Then concentrated using a rotary evaporator to obtain concentrated ethanol extract. After obtaining a concentrated 70% ethanol extract, the yield of *M. alba* flower extract was calculated, which can be seen in Table 1.

Table 1. Yield of *M. alba* Flower extract

Extract	Concentrated extract weight (g)	yield (%) (b/b)	Concentrated extract
Ethanol 70 %	50 g	16,8 %	Dark brown

Qualitative analysis was carried out to determine the chemical components of plants using Mg powder and a few drops of concentrated HCl. The results obtained indicated that the ethanol extract of *M. alba* flowers contained flavonoid compounds, the change in color is indicated by turning red to orange, the orange color indicated that were flavonoids from the flavones, auron, and chalcone groups [11].

Samples that are proven positive for containing flavonoids are then determined by the UV-Vis spectrophotometric method because the results are more accurate and faster. Flavonoid analysis was carried out by UV-Vis Spectrophotometry because flavonoids contain conjugated aromatic systems so that they can show strong absorption bands in the ultraviolet and visible spectral regions [7].

This test aims to determine the total flavonoid content in the sample through the line equation formula. The determination of the quercetin absorbance curve was carried out at concentrations series is used to determine the absorbance value, then the blank used is only 70% ethanol solvent without using the addition of reagents because the wavelength that is read is only the quercetin wavelenth. Quercetin is used as a standard solution because quercetin is flavonoid of the flavonol group which has keto group at C-4 and has a hydroxyl group at C-3 or C-5 atoms which are neighbors of flavones and flavonols [12].

Measurement of the maximum wavelength of quercetin was carried out at wavelength in the range of 400-600 nm. Based on the results of measurement of the quercetin wavelength obtained, namely 432 nm. Can be seen in Table 2.

Table 2. Results of absorbance measurement of the quercetin standard solution at a maximum wavelength of 432 nm

Quercetin	Absorbansi rata-rata
20	0,007
40	0,028
60	0,074
80	0,099

100	0,149

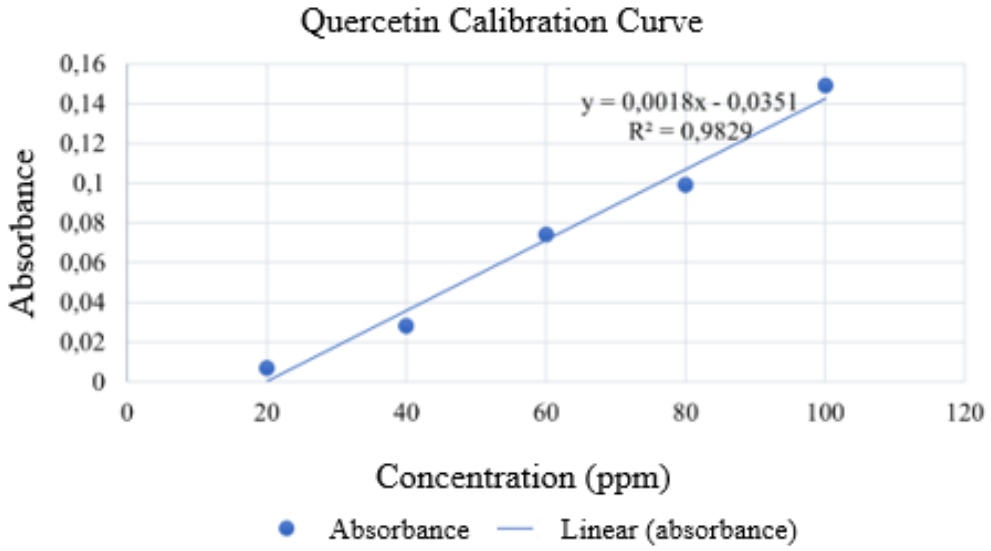

Figure 1. Quercetin calibration curve at a maximum wavelength of 432 nm

Based on these measurements (Figure 1), it can be seen that the value of R^2 and the equation of the linear line. The calibration curve is obtained from the relationship between quercetin ppm concentration and absorbance so that a regression equation is obtained, namely y=0,0018x-0,0351 with a correlation coefficient (R^2) of 0,9829 which is close to 1 so that it shows a linear calibration curve. The equation that has fulfilled the slope (b) and intercept (a) and absorbance can be used to determine the total flavonoids of *M. alba* flowers.

The total flavonoid compounds were determined by adding $AlCl_3$, a complex-forming molecule, to the sample solution, which caused a change in the solution's color from blue to yellow. In order to keep the wavelength in the visible range, potassium acetate is added [13]. Before taking a reading, incubating the sample for 30 minutes is done to ensure a smooth response. This results in the highest possible hue saturation. So that the yield of total flavonoids from *M. alba* flowers had flavonoid levels successively at a concentration of 100 ppm of 27,683 ± 1,77 mgQE/g, 500 ppm of 52,597 ± 1,98 mgQE/g, 1000 ppm 79,115 ± 8,33 mgQE/g as in Table 3.

Table 3. Results of determining the total flavonoid content % (w/w) in the ethanol extract of white cempaka flowers (*Magnolia alba* (DC.) Figlar).

Sample	Concentration	Absorbance	Total flavonoids level	The average level of total flavonoids
White cempaka flower (*M.alba*)	100	0,01790	294,44	27,683±1,77
		0,01153	259,04	
		0,12078	277	
	500	0,05745	102,836	52,597±1,98
		0,05352	98,266	
		0,06774	114,266	
	1000	0,12078	86,6	79,115±8,33
		0,11002	80,622	
		0,09116	70,144	

According to the table above, it is stated that the crude ethanol extract of white cempaka flowers (*M. alba*) can be tested positive for containing flavonoid, where flavonoids have many benefits in the health sector including as antioxidants, antidermatosis, chemopreventive, anti-cancer and antiviral [14].

CONCLUSION

The qualitative analysis of flavonoids supported the findings of the research, demonstrating that *M. alba* flower extract contained flavonoids and Determination of total flavonoid levels of *M. alba* flowers extract by UV-VIS spectrophotometry, namely successively at a concentration of 100 ppm of 27,683 ± 1,77 mgQE/g, 500 ppm of 52,597 ± 1,98 mgQE/g, 1000 ppm 79,115 ± 8,33 mgQE/g.

REFERENCES

[1] Aksara, R., Weny, J. A. M., dan La, A. Identifikasi Senyawa Alkaloid Dari Ekstrak Metanol Kulit Batang Mangga (*Mangifera Indica* L). *Jurnal Entropi*. 2013. Vol 3 (1): 1-6.

[2] Ismawan, B. 2010. *Herbal Indonesia Berkhasiat Bukti Ilmiah dan Cara Racik Volume 8.*. Trubus Swadaya.: Bogor.

[3] Ipandi, I., Triyasmono, L., dan Prayitno, B.. Penentuan Kadar Flavonoid Total dan Aktivitas Antioksidan Ekstrak Etanol Daun Kajajahi (*Leucosyke capitellata wedd*). *Jurnal Pharmascience.* 2016. Vol. 3, No.1: 99-107.

[4] Pensuk, W., Tanit, P., Chayan, P. Comparison of The Chemical Consistuents in Michelia alba Flower Oil Extracted by Steam Distillation, Hexane Extraction and Enflourage Method. *Journal of That Traditional and Alternative Medicinel*. 2017. Vol 5. No.1

[5] Robby, F. A., Iman, S. P., Galuh T. Pediculidical Activity of White Cempaka Flower (*Michelia alba* DC.). Againts Pediculus Humanus Capitis. *Ad-Dawa Journal of Pharmaceutical Sciences*. 2021. Vol. 4. No 1: 9-16.

[6] Lan-yu, X., Jia Li, Yanqing, Y., Jiayu, Z., Yongwen, J. Comprehensive Investigation on Flavonoids Metabolites of Longjin Tea in Different Cultivas, Geographical Origins, an Strongetime. *Heliyon*, 2023, Vol 9.

[7] Harbone, J.B., *Metode Fitokimia; Penuntun Cara Modern Menganalisa Tumbuhan*. Terbitan Kedua, Terjemahan Kokasih Padmawinata dan Iwang Soeidiro. 1987. Bandung: ITB.

[8] Isnaini, L. Ekstraksi Pewarna Merah Cair Alami Berantioksidan Dari Kelopak Bunga Rosella (Hibicus Sabdriffa L) dan Aplikasinya Pada Produk Pangan. *Jurnal Teknologi Pertanian*. 2010. Vol 11 (1).

[9] Utami, T. S., Arbianti, R., Hermansyah, H., dan Reza, A. Perbandingan Aktivitas Antioksidan Ekstrak Etanol Daun Simpur (Dillenia indica) dari Berbagai Metode Ekstraksi dengan Uji ANOVA. *Prosiding Seminar Nasional Teknik Kimia Indonesia.* Bandung: 19-20 Oktober 2009. Hal: 1-6.

[10] Synder, C. R., J.J. Kirkland, and J. L. Glajach. Pratical HPLC Method Develoment, Second Edition. New York: John wiley and Sons. Lnc. 1997. Pp. 722-723

[11] Oktavia, D. F., Suyatno, S. Skrining Fitokimia, Kandungan Flavonoid Total, Dan Aktivitas Antioksidan Ekstrak Etanol Tumbuhan. *Jurnal Kimia Riset*. 2021. Vol 6. No 2: 141-153.

[12] Azizah, D. N., Kumolowati, E., Faramayuda, F. Penetapan Kadar Flavonoid Metode $AlCl_3$ Pada Ekstrak Metanol Kulit Buah Kakao (*Theobroma cacao L.*). *Kartika Jurnal Ilmiah Farmas*i. 2014. Vol 2 (2): 47

[13] Chang, C. C., Yang, M. H., Wen, H. M., dan Chern, J.C. Estimation of Total Flavonoid Content in Propolis by Two Complementary Colometric Methods. *Journal of Food and Drug Analysis*. 2002. Vol 10 (3): 178-182
[14] Saraswati, N. F. Uji aktivitas Antibakteri Ekstrak Etanol 96% Limbah Kulit Pisang Kepok Kuning (*Musa balbisiana*) Terhadap Bakteri Penyebab Jerawat (*Stapylococcus epidermidis, Staphylococcus aureus, dan Propionibacterium acne*). *Skripsi*, 2015