DETERMINATION OF TOTAL FLAVONOID CONTENT OF YOUNG SUJI LEAF EXTRACT (*Dracaena Angustifolia*) USING THIN LAYER CHROMATOGRAPHY AND UV-VIS SPECTROPHOTOMETRY METHODS

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A B S T R A C T

Dracaena angustifolia Roxb or better known as the suji plant has long been known in Indonesia, in its use as medicine, generally the part that is used is the leaves. Suji is said to have several compounds such as saponins, flavonoids, steroids and phenolic compounds. The most abundant polyphenol compound in suji leaf plants is flavonoids. Even so, the levels of flavonoids in a plant have different levels, both in tissue, plant age and environmental factors, and such as temperature, ultraviolet light, nutrition, water availability, and C02 levels in the atmosphere. Suji leaves can also be used for anti-inflammatory drugs, anti-dysentery, beriberi, gonorrhea and menstrual pain medication. The purpose of the study was to determine the presence of flavonoid compounds and the total flavonoid content of young suji leaf extract. The research sample was taken from Lahumoko Village, Kambowa District. This type of research is qualitative and quantitative using thin layer chromatography and uv-vis spectrophotometry methods. In the Thin Layer Chromatography (TLC) method, the mobile phase used is n-hexane: ethyl acetate (3:7), the visibility of spots on the TLC plate is assisted by UV light 366 nm. Determination of the flavonoid content of young suji leaves using UV-Vis spectrophotometry 366 nm. The results of the study contained flavonoid compounds as seen from the change in greenish yellow color and the total flavonoid compound content of young suji leaves was 291,731 mgRE/g. by 366 nm UV light. The determination of flavonoid content of young suji leaves was determined using UV-Vis 366 nm spectrophotometry. The results of the study showed a variety of flavonoids with a greenish-yellow color change and the level of flavonoid compounds in the total extract of young suji leaves was 291,731 mgRE/g.

INTRODUCTION

Indonesia is a country with many islands which has thousands of islands with a forest area of up to 130.78 hectares. The total number of medical plants in Indonesia covers 90% of the number of medical plants in the Asian region. Indonesia has more than 20,000 types of medical plants, but only 1000 types of plants have been recorded and only around 300 types are used as medical plants (Yolandari & Mustiqawati, 2022). The use of one of the medical plants in Indonesia is the suji plant (*Draceana angustifolia*). The suji plant is a woody, grooved and transverse plant that has a pointed leaf shape. Suji leaves are the part most often used for medicinal plant materials, with a length of about 10 to 25 cm and a width of 0.9 to 1.5 cm (Narande *et al.*, 2013).

Dracaena angustifolia Roxb or better known as the suji plant has long been known in Indonesia, in its use as medicine, generally the part that is used is the leaves. Suji is said to have many compounds such as saponins, flavonoids, steroids and phenolic compounds (Handayani, 2018). Pandanus leaves have the highest chlorophyll content compared to other plants. Pandanus leaf water extract was found to contain alkaloids, flavonoids, glycosides, saponins, and triterpenoids. The flavonoid and chlorophyll content in pandanus leaves shows that pandanus leaves have antibacterial and anti-insecticide properties (Marleni *et al.*, 2018)

The use of plant compounds such as alkaloids, organosulfur, phenolic acids, flavonoids, carotenoids, coumarins, terpenoids and tannins is good for further research for the advancement of science and health (Mustiqawati, 2021). Flavonoids are secondary metabolites of polyphenols, found in abundance in plants and have medical effects including anti-virus, anti-inflammatory, cardioprotective, anti-diabetic, anti-cancer, anti-aging, antioxidant and others (Arifin & Ibrahim, 2018). The amount of flavonoids in plants is not the same, whether in parts, tissues, and age of plants, and also due to environmental factors. These factors include CO2 levels, water availability, ultraviolet light, nutrients and temperature. Based on research on anti-inflammatory activity and the total amount of flavonoids, the total amount of flavonoids 0.95% to 16.937% has inflammatory activity (Satria et al., 2022).

Flavonoid content is determined spectrophotometrically through the $AlCl_3$ colorimetric method, where the measurement principle is measured from color. This method is known to be able to

measure the amount of flavonoids. Quercetin is a flavonoid with a keto group at C-4 and a hydroxyl group at C-3 or C-5 atoms adjacent to flavones and flavonols, so quercetin is used as a comparison when making a calibration curve (Kartikasari et al., 2019).

In this study, the extraction method used the maceration method, which is a simple filtration method that does not cause damage to secondary metabolite compounds in the sample due to strong heating. The use of multi-stage maceration techniques aims to extract compounds based on their polar properties and maximize the extraction process (Safrudin et al., 2022). Based on the description above, the researcher is interested in conducting this research with the aim of determining the total flavonoid content of young striped leaf plants (*Dracaena angustifolia*).

METHODOLOGY

Time and Place of Implementation

This research was conducted from July 3 to July 22, 2024 at the Pharmacy Laboratory of Halu Oleo University (UHO) Kendari.

Tools

The tools used in this study were stirring rods, blenders, porcelain cups, chambers, measuring cups, filter paper, dropper pipes, rotary evaporators, test tubes, analytical scales, TLC and glass jars.

Materials

The materials used are young pandan leaf extract, 96% ethanol, EEDC, methanol, AICI3 and potassium acetate.

Work Procedures

Sample Preparation

Young suji leaves were taken from Lahumoko Village, Kambowa District. The samples were cleaned until all the dirt that was attached was separated. To facilitate the drying process, the samples were chopped and then dried in the sun until the water content decreased and were completely dry. The dried samples were then blended until smooth and stored in a tightly closed container. The sample extraction process is ready to be carried out.

Sample Extraction

Dry simplisia of young suji leaves amounting to 413 grams was soaked with 2 liters of 96% ethanol. The mixture was soaked for approximately 3 days while stirring periodically. Separated between the dregs and the existing solvent. The dregs obtained were then re-macerated again 2 times with 1 liter of 96% ethanol. The maceration results obtained were then thickened using a rotary evaporator.

Thin Layer Chromatography Test

Pandanus leaf extract was mixed in 96% ethanol, dropped on the stationary phase, and eluted with the mobile phase (n-hexane::ethyl acetate (3::7)). The spot length was observed at 254 and 366 nm under UV light. Sprayed with flavonoid compound reagent in the form of sitroborate. A good fluorescent color is greenish yellow. After that, heating was continued for 1 to 5 minutes at a temperature of 1000°C, and the yellow/green-yellow fluorescence was observed under 366 nm ultraviolet light, and the Rf value of each visible spot was finally determined.

UV-Vis Spectrophotometry Test

Preparation of Quercetin Standard Solution

A 1000 ppm standard solution was prepared by weighing 10 mg of quercetin mixed in 10 mL of methanol. The solution was diluted into several concentration series: 37.5 µg/ml, 62.5 µg/ml, 75 µg/ml, 87.5 µg/ml and 100 µg/ml. Optimize the maximum wavelength of the routine standard before measurement. The absorbance was then measured by UV-Vis spectrophotometry with a wavelength of 418 nm.

Making an ethanol extract solution from young suji leaves (*Dracaena angustifolia*)

EEDC extract of 10 mg was weighed and mixed in 10 mL of methanol. Pipette 0.5 mL each, add 1.5 mL of methanol, 0.1 mL of AlCl3, 0.1 mL of potassium acetate, and add distilled water until it becomes 5 mL, repeat three times. Vortex and incubate for 30 minutes. The last step is to measure the absorbance at a wavelength of 418 nm.

RESULTS & DISCUSSION

In this study, fresh young suji leaves (*Drancaena agustifolia*) were used as samples on leaves that were not too old because they contained flavonoid chemical compounds. As many as 5000 grams of suji leaves were taken, sorted wet and then washed with running water. Then, they were dried in an open place covered with a cloth so that they were not exposed to direct sunlight. This was done to reduce damage to the chemical components in the suji leaves due to exposure to sunlight. The suji leaves were blended until they became a simple powder of suji leaves weighing 431 grams, then extraction was carried out by maceration with 96% enatol solvent of 2000 mL within 3x4 hours. (Marleni et al., 2018). After maceration, filtration is carried out to separate the filtrate from the residue. After that, the filtrate that has been obtained is then used in a rotary evaporator to produce a thick extract of young suji leaves. This extract is used for testing with phytochemical screening and with the thin layer chromatography method against a number of secondary metabolite compounds in young suji leaves (*Drancaena agustifolia*) (Putri & Lubis, 2020).

A simple method is used in this extraction process. The extraction obtained was 431 grams. Maceration is one of the extraction methods through the process of soaking the powdered simplicia with liquid, and does not utilize the heating process or is also known as cold extraction (Dewatikasari, 2020). During the maceration process, the solvent used is 96% ethanol because its use is selective, non-toxic, and has good absorbency, which can inhibit the growth of bacteria and fungi (Damanis et al., 2020). The purpose of extraction is to remove chemical components present in natural materials. This extraction uses the principle of mass mobilization of material components into the solvent. The transition begins at the interface and diffuses into the solvent (Meigaria et al., 2016).

Phytochemical Identification

Table 1. Results of Phytochemical Screening Test of Young Pandan Leaf Extract (*Drancaena angustifolia*)

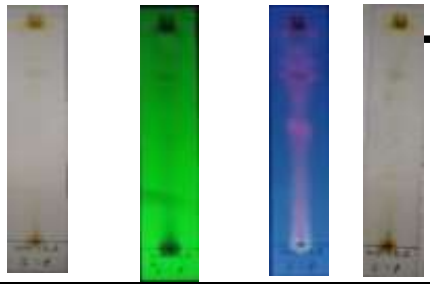
Compound	Reagent	Results	Information
Flavonoid	Mg powder + concentrated HCl	+	Greenish yellow

Source: Primary Data (2024)

Description:
+ = Positive

Flavonoid TLC Testing

The following are the results of the flavonoid compound test using the Thin Layer Chromatography method:

After Elution	UV 254nm	UV 366nm	Citroborate Reagent	Information
			Spot 1	Greenish yellow in color

Source: Primary Data (2024)

TLC profile observations showed that all young leaf extracts of Drancaena agutifolia contained various compounds, this was evidenced by the presence of colored spots on the research plate.

Flavonoid studies utilize thin layer chromatography (TLC) methods. TLC itself is a method of separating compounds based on differences in the distribution of two phases, namely stationary and mobile. The stationary phase used is UV 254 and the graphene phase is n: hexane: ethyl (3: 7). When viewed under UV 366 nm light, the spots are slightly fluorescent or fluorescent, and after spraying and

heating $AICI_3$ and Dicitrobar, the greenish yellow fluorescent spots become more concentrated when observed under UV 366 nm, indicating the presence of flavonoids (Winariyanthi, 2017). Based on several previous studies, it is known that flavonoid compounds can be used as antidiarrheal drugs. Flavonoids as antidiarrheals work by inhibiting intestinal motility (Teheni et al., 2024).

Table 2. Results of Thin Layer Chromatography Testing Analysis of Young Pandan Leaves (*Drancaena agustifolia*)

Sample Name	Compound	Reagent	Rf value	Information
Total extract of young suji leaves (*Dracaena angustifolia*)	flavonoid	sitoborat	0,97	Greenish yellow in color

Source: Primary Data (2024)

The research was conducted using 5000 grams of young pandan leaves (*Drancaena agustifolia*) which were macerated using 2000 liters of 96% ethanol, finally obtaining 431 grams of dry extract.

The yield percentage obtained is:

$$\%\ Yield = \frac{Extract\ weight}{Weight\ of\ Simplex} \times 100\%$$

$$= \frac{431\ gram}{5000\ gram} \text{ x } 100\%$$

$$= 8{,}62\ \%$$

Quarcetin Standard Curve 433.5 nm

The results of measuring quarcetin as a comparison of 37.5 62.5 75 87.5 and 100 mg/L at a wavelength of 433.5 nm and a comparison of five concentrations obtained y = 0.0065x + 0.3478 with an R2 value of 0.9946.

Table 3. Measurement of Absorbance Value of Quercetin at 4.335

Concentration (ppm)	Absorbance
37.5	0.6033
62.5	0.7360
75	0.8369
87,5	0.9180
100	1.0065

Source: Primary Data (2024)

The following is a graph of the results curve of measuring the absorbance value of quercetin:

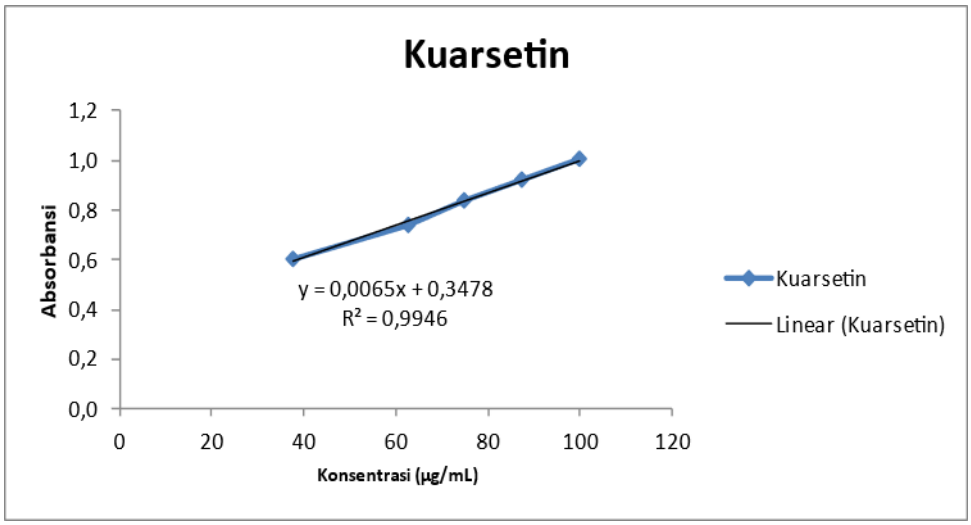


Figure 2. Curve Graph Results at 433.5 nm

The treatment given to the sample to obtain the curve and correlation coefficient value of the sample and the appropriate wavelength, then the sample is measured with the following wavelength:

Table 4. Maximum Wavelength Measurement of Quartzite

Standard	Concentration (ppm)	Maximum Wavelength (nm)	Absorbance	Linear Regression
Quercetin	37,5	433,5	0,6033	y = 0,0065x + 0,3478 R^2 = 0,9946
	62,5		0,7360	
	75		0,8369	
	87,5		0,9180	
	100		1,0065	

Source: Primary Data (2024)

Table 5. Three-fold Absorbance Results of Young Pandan Leaf Samples at 433.5 nm

Absorbance 1st	Absorbance 11th	Absorbance 111th	Flavonoids Total(%)
1.1065	1.1.078	1.1061	291.731

Source: Primary Data (2024)

Determination of total flavonoid content of EEDC using UV-Vis spectrophotometry. The measurement method based on the principle of spectrophotometry is based on the absorption of light with a specified wavelength by a solution containing pollutants, and the concentration of the pollutants is determined. The method has been validated and declared selected as a method of flavonoid analysis using UV-Vis spectrophotometry which is calculated routinely because it provides good parameters. Flavonoids also have a carbonyl system conjugated to the aromatic ring, so flavonoids can be characterized spectrophotometrically (Lestari et al., 2023).

Based on the results of testing flavonoid compounds of 96% ethanol extract of pandan leaves, after elution, yellow spots were formed. Observation with visible light UV 254 nm and at UV 366 nm. After spraying using sitroborate reagent, the spots were yellow, greenish. This is in accordance with research conducted by (Ramadhan *et al*., 2021). This confirms the presence of 96% flavonoids in the ethanol extract of young striped seaweed. Flavonoids are polar secondary metabolites. Flavonoids are thought to form bonds with a mixture of boric acid and citric acid when heated, and are better known as the sitroboric acid reagent. Researchers have found flavonoid compounds that act as antioxidants, anti-inflammatory agents, and have antitumor and antiviral effects (Bangun *et al*., 2021).

UV-Vis 366 lamps work the same way as other types of lamps. The UV lamp types have similar operating principles. Ultraviolet light passes through a lamp that is protected by clear glass. There are two types of UV-Vis lamps in the entire UV-Vis light trap. First, the lamp emits light at a wavelength of less than 366 nm (Irawan, 2019).

How UV-Vis 254 works is that low-pressure light sources are specifically designed to produce the maximum amount of UV radiation, with 90% of the energy normally produced at 254 nm. This wavelength of radiation is very close to the peak of the germicidal effectiveness curve of 265nm. The most lethal wavelength for microorganisms. 254 nm UV light has been shown to destroy bacteria and viruses by altering the DNA of harmful cells, even microorganisms that are difficult to see. (Azza et al., 2021).

CONCLUSION

Based on the results of the research conducted, it can be concluded that there is a flavonoid compound content in the extract of young suji leaves marked by the presence of greenish yellow spots in the test. It is known that the total flavonoid compound content in the extract of young suji leaves is 291.731 mgRE/g.

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