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Effect of Different Extraction Method on Total Flavonoid Contents of Sansevieria trifasciata P. Leaves Extract

(Pengaruh Perbedaan Metode Ekstraksi Terhadap Kadar Flavonoid Total Ekstrak Daun Lidah Mertua (Sansevieria trifasciata P.)

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ABSTRACT.

Sanseviera leaves (*Sansevieria trifasciata* P.) is a plant that contains flavonoids. Flavonoid in the health sector act as antibacterial, antioxidant, anti inflammatory and anti diabetic. The extraction method will determine the amount of substance on the plant extract. The purpose of this study was to determine the effect of different extraction method on total flavonoid contents of sansevieria leaves extract. The method used is an experimental by comparing meceration and soxhletation extraction method to the total flavonoid contents of sansevieria leaves extract using UV-Vis spectrophotometric. The results showed that the total flavonoid content of the maceration extraction method was 13.934 mgQE/g or 1.39% higher than the soxhlet extraction method of 8.117 mgQE/g or 0.81%. The results of statistical tests showed the significant value of 0.001 (p<0.05), that means there is a significant effect between the contents of total flavonoids in maceration and soxhlet extraction methods.



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ABSTRAK

Daun lidah mertua (*Sansevieria trifasciata* Prain) merupakan salah satu tanaman yang mengandung flavonoid. Flavonoid dalam bidang kesehatan berperan sebagai antibakteri, antioksidan, anti inflamasi dan anti diabetes. Metode ekstraksi akan menentukan banyaknya zat yang dapat tersari sehingga dilakukan penelitian ini. Tujuan penelitian ini untuk mengetahui pengaruh kadar flavonoid total ekstrak daun lidah mertua dengan menggunakan metode meserasi yang berbeda. Metode yang digunakan adalah metode eksperimen dengan membandingkan metode ekstraksi meserasi dan sokletasi terhadap kadar flavonoid ekstrak daun lidah mertua menggunakan analisis spektrofotometri UV-Vis. Hasil penelitian menunjukan bahwa kadar flavonoid total metode meserasi yaitu 13,934 mgQE/g atau 1,3934% lebih tinggi dari metode sokletasi yaitu 8,117 mgQE/g atau 0,8117 %. Hasil uji statistik menunjukkan nilai sig 0.001 (p< 0.05), artinya terdapat pengaruh yang signifikan antara kadar flavonoid total pada metode maserasi dan metode sokletasi.

Kata kunci: Maserasi; Sokletasi; Flavonoid; Spektrofotometri UV-Vis; Sanseviera trifasciata P.

INTRODUCTION

Indonesia is a country that has abundant natural resources with various types of plants that can be used as traditional medicines (Rusdi, 2018). One of the plants that can be used is the sansevieria leaves. This plant is widely known as an ornamental plant and is believed to be able to absorb air pollution and is widely used as an indoor refreshing plant (Nurjanah, 2014). The sansevieria leaves can also be used as a medicinal plant (Suharsi & Andiani, 2013). By utilizing natural resources, the mother-in-law's tongue leaf is a form of local wisdom that must be maintained. The use of medicinal plants that have been passed down by ancestors then develops and produces a local wisdom, this local wisdom appears in the form of a culture of utilizing the values and efficacy of medicinal plants. Sansevieria leaves is a plant that contains flavonoid compounds. Flavonoids are natural phenolic compounds found in almost all parts of plants. Flavonoids in the health sector act as antibacterial, antioxidant, anti-inflammatory and anti-diabetic (Panche et al., 2016).

There are several method that can be used to isolate the active compounds from natural ingredients, including maceration extraction, soxhletation, reflux, sonication, distillation and others (Mukhtarini, 2011). The extraction method will determine the amount of substance that can be extracted, so this a study was conducted to compare the levels of flavonoids content in the ethanol extract of Sansevieria leaves. In this study, sansevieria leaves was extracted with ethanol 96% solvent with different method, namely maceration and soxhletation methods.

The maceration method was carried out by immersing the simplicia at room temperature with the appropriate solvent. The immersion of the sample is carried out for 3-5 days by stirring several times to accelerate the analyte dissolution process (Leba, 2017). The soxhletation technique is a method of separating substances from the mixture by heating, the solvent used will undergo circulation (Sri Irianty, 2014).

The total flavonoid contents was higher in the maceration technique because there were a class of flavonoid compounds that were not heat resistant and easily oxidized in the soxhletation method (Ramayani et al., 2021). Based on this description, it is interesting to examine the effect of maceration and soxhlet extraction methods to obtain the highest contents of flavonoids in sansevieria leaves with ethanol 96% solvent.

MATERIAL AND METHODS

Materials

Sanseviera leaves (*Sansevieria trifasciata* P.) were collected from garden in Tasikmalaya and identified at the Biology Education Laboratory, Faculty of Teacher Training and Education, Galuh University, ethanol 96% (Brataco®), methanol p.a (Brataco®), potassium acetat (Brataco®), quercetin (Brataco®), AlCl₃ (Brataco®), chloroform (Brataco®), aquadest.

Methods

Collection and Processing of Sansevieria Leaves

The simplicia that had been collected was then washed with running water, drained, then cut into small pieces and weighed as wet weight, then dried in an oven which is heated at 70° C for 12 hour, then weighed as dry weight. The dried samples were mashed using a blender, then stored in a plastic container to prevent the influence of moisture and other impurities.

Maceration extraction method

A total of 100 grams of powdered samples of sansevieria leaves were weighed and put in a maceration container. The ethanol 96% was added until the simplicia was completely submerged. Then left for 24 hours, stirring occasionally. Then filtered and separated the dregs and filtrate. Furthermore, the dregs were macerated again using a new ethanol filter. This was done for 3 consecutive days. The ethanol 96% extract of sansevieria leaves obtained was concentrated by means of a rotary evaporator (Amelinda et al., 2018).

Soxhlet extraction method

The soxhletation of sansevieria leaves was performed by using ethanol 96% as a solvent. 100 grams sansevieria leaves powder was wrapped in filter paper, then tied at both ends with thread, inserted into a soxhlet, put 2.5 L of 96% ethanol solvent into a soxlet flask (round bottom flask). Then, soxhletation was carried out at 70°C until the cycle droplets were colorless. The obtained filtrate is then concentrated using a rotary evaporator at a temperature of no more than 50°C and concentrated again using a water bath to become a thick extract (Amelinda et al., 2018).

Preparation of quercetin standard solution

A total of 50 mg of quercetin was weighed and dissolved in 50 ml of methanol p.a as a stock solution of 1000 ppm. Then made dilutions of quercetin with various concentrations as a comparison solution. Then 1 ml pipette was added and 1 ml of 1% AlCl₃ was added, the absorbance was measured at a length of 400-800 nm (Amelinda et al., 2018).

Maximum wavelength determination

The stock solution was taken as much as 1 ml and then the volume was made up using methanol p.a to 10 ml. The absorbance was measured using a UV-Vis spectrophotometer at the various wavelength. The maximum wavelength was determined by choosing the maximum absorbance obtained (Tarafder et al., 2007).

Quercetin Standard Curve Generation

The solution was prepared by dissolving 50 mg of quercetin in 100 methanol p.a until the volume was sufficient in a 100 ml volumetric flask to obtain 100 ml of 1000 ppm stock liquor. From a standard solution of 1000 ppm quercetin, then several concentrations were made, namely 5 ppm, 10 ppm, 15 ppm 20 ppm, 25 ppm and 30 ppm. From each concentration of standard solution of quercetin 1 mL was pipetted and 1 mL of 1% AlCl₃ and 1 mL of 120 mM potassium acetate were added. Samples were incubated for 30 minutes at room temperature. The absorbance was determined using the UV-Vis spectrophotometric method at the maximum wavelength (Aminah et al., 2017).

Determination of Total Flavonoid Contents

The samples of the ethanol extract of the maceration and soxhlet method were weighed 50 mg each. Each sample was dissolved with 5 mL of methanol p.a into a glass beaker, put into a 10 mL volumetric flask, add ethanol to the mark, then filtered with filter paper. The sample solution was pipetted as much as 1 mL and then put into a 10 mL volumetric flask and ethanol was added to the mark. 0.5 mL of the test solution was taken, then reacted with AlCl, 1 mL and 0.1 mL of potassium acetate and 1.5 mL of ethanol 96% and allowed to stand for 30 minutes. The solution is read the absorbance value at maximum. Each extract was determined for 3 times replication. The average absorbance is entered in the standard quercetin curve equation as the y value, where the x value obtained is the equivalent of milligrams of quercetin in every 100 milligrams of sample (Quercetin Equivalent, QE) (Aminah et al., 2017).

Data analysis

After all the data has been collected, the next step is to analyze the data using SPSS software using the one-way variation analysis (ANOVA) method.

RESULTS AND DISCUSSION

Extraction of Sansevieria Leaves

Tabel 1. Extraction Yield Form Maceration and Soxhletation Method

Extraction Method	Simplisia Weight Grams (g)	Extract Weigth Grams (g)	Rendement %
Maceration	100	6.18	6.18
Soxhletation	100	7.64	7.64

The results of the extraction process showed that more extract rendement were obtained by the soxhletation method of 7.64% because the soxhletation process was carried out repeatedly. Soxhletation was carried out for approximately 7 cycles or until the cycle drops were colorless which indicated the cessation of the extraction process because all the compounds in the simplicia had been completely extracted. Heating with this method can extract compounds that are insoluble at room temperature, can liberate and activate low molecular weights from high molecular weight polymer molecular subunits so that compound withdrawal activity is maximized (Hatam, 2013). Extraction method obtained by maceration yield 6.18% smaller than the results of soxhletation. This is because the maceration method using room temperature that it cannot extract compounds that are insoluble at room temperature and the extraction process is not perfect.

Determination of Flavonoid Contents

The measurement of the total flavonoid contents of the leaf extract of *Sansevieria trifasciata* carried out using the UV-Vis spectrophotometer method because this method is quite simple and the results obtained are quite accurate. Quercetin was used as a comparison because quercetin is the most widely distributed compound found in plants and is one of the flavonoid compounds that can react with AlCl₃ to form complexes. The result of determining the maximum wavelength is seen from the highest absorbance value, which is 415 nm (Table 1). This is in accordance with the research of (Ipandi et al., 2016) which determined the total flavonoid content using AlCl₃ with a wavelength of 415 nm. Operating time obtained is 30 minutes.

Table 1. Determination of Maximum Wavelength

wevelength (nm)	absorbance
385	0,217
400	0,22
415	0,241
430	0,211
445	0,106

The results obtained from the absorbance value of the quercetin standard solution can be seen on the quercetin calibration curve (Figure 1). It is in accordance with the Lambert-Beer law, where the concentration is directly proportional to the absorbance where the higher the absorbance value, the higher the concentration of the substance contained in a sample. The results show a straight relationship between absorbance and analyte content.

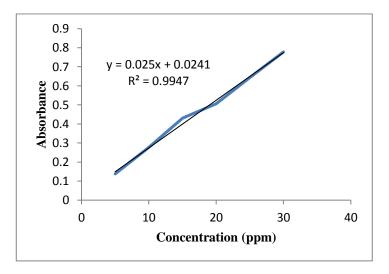


Figure 1. Quercetin Calibration Curve

Table 2. Calculation of total flavonoid content

Exctraction Method	Absorbance	Average flavonoid content (mgQE/g sample) ±SD
Maceration	0.372	13.934 ± 0.200
Soxhletation	0.227	8.117 ± 0.168

According to the 2014 Director General of Food and Drug Administration, the range of total flavonoid contains based on their absorbance values ranged from 0.2 to 0.8. And the absorbance values obtained in the maceration technique of the sansevieria leaves extract were 0.376, 0.369 and the soxhletation method 0.230, 0.227. The equation of the linear line y=0.025x + 0.0241 is obtained with an R2 value of 0.9947. The correlation coefficient value close to one indicates that the regression equation is a linear equation of the quercetin calibration curve that can be used as a comparison to determine the concentration of total flavonoid compounds in the sample extract so that the total flavonoid content for the maceration method extraction is 13.934 mg QE/g or 1.3934%. and soxhletation 8.117 mg QE/g or 0.8117%. Table 2 shows that the maceration method produces higher flavonoid levels than soxhletation. This is caused by the heating in the drying process, the concentration of the extract using a rotary evaporator so that it affects the decrease in the flavonoid contents. Due to heating. It can result a decrease

in flavonoid levels. There is a relationship between temperature and phenolic contents. An increase in temperature can cause an increase in phenolic content up to a certain temperature and then decrease with an increase in higher temperature. At a temperature of 50°C, it is relatively safe to prevent damage to certain secondary metabolites, especially flavonoids. Flavonoids are phenolic compounds that have a conjugated aromatic system. The conjugated aromatic system is easily damaged at high temperatures (Sa'adah et al., 2017).

Based on the statistical test carried out is the One-way Anova test, the data to be tested must be normally distributed and have the same variance (homogeneous). Therefore, before testing with the ANOVA test, the data must be tested for normality and homogeneity test using SPSS. Based on the normality test, the data tested were normally distributed, this was evidenced by a significance value > 0.05 so that it was proven that the data were normally distributed. Based on the homogeneity test, the data obtained have the same variance, because the significance value is 0.07 > 0.05 so it is proven that the data is homogeneous. From the One-way Anova test, a significance value of 0.001 < 0.05 was obtained, so the results were significant. This means that the extraction method of the sansevieria leaves influences the total flavonoid contents of the extract.

CONCLUSION

The total flavonoid contents of the maceration extraction method were 13.934 mgQE/g or 1.39% higher than the soxhlet extraction method of 8.117 mgQE/g or 0.81%. The results of statistical tests showed that the sig value of 0.001<0.05, which means that there is a significant effect between the levels of total flavonoids in maceration and soxhlet extraction methods.

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CONFLICT OF INTEREST

The authors declare that there is no conflict of interest.

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