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The Effect of Solvent Variation on Flavonoid Content of Purslane Herb (Portulaca grandiflora Hook.)

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ABSTRACT

Purslane is from the *Portulaceae* family which contains active phenolic compounds and has different antioxidant activities. *Portulaca grandiflora* Hook. is one of the species of purslane where all parts of the plant are useful. Purslane herb (*Portulaca grandiflora* Hook.) contains active compounds of alkaloids, flavonoids, tannins, terpenoids, and saponins. Flavonoids act as antioxidants. This study aims to determine the differences in flavonoid levels of extracts resulting from soxhletation of purslane (*Portulaca grandiflora*) herb with different solvents. This research is an experimental study with soxhletation extraction process and assay using a UV-Vis spectrophotometer with various concentrations of 5, 10, 15, 20, 25, and 30 ppm at a wavelength of 424.5nm. The flavonoid content in each extract of n-hexane, ethyl acetate, and 96% ethanol was 17.91% w/w; 23.27% w/w; 11.67% w/w.

Keywords: Flavonoid Levels, Purslane, Portulaca Grandiflora, Solvent, Soxhlet

INTRODUCTION

Purslane is a plant that grows wild in open places exposed to sunlight and is often found as a nuisance plant (weeds) in yards, plantations, roadsides, purslane can be consumed as fresh vegetables, cooking, herbal medicines which are empirically used by the community to treat diarrhea, breast inflammation, hemorrhoids, insect bites, ulcers, and eczema (Wijayakusuma, 2008).

Purslane comes from Brazil, herbaceous annuals with wet trunks and grows supine or up with a length of about 15-30 cm, and has flowers gathered 2-8 at the end of the stem. Choon's research (2014) shows that all parts of the plant are beneficial, especially the leaves which contain the highest phenolic compounds and have antioxidant activity. Flavonoid compounds are polyphenolic compounds that have 15 carbon atoms arranged in a C6-C3-C6 configuration, meaning that the carbon skeleton consists of two C6 groups (substituted benzene rings) joined by a three-carbon aliphatic chain (Tiang-Yang *et al.*, 2018).

Arifin & Ibrahim (2018) showed that Portulaca has properties as an antioxidant, antimicrobial, anti-inflammatory, and diuretic which is associated with the phytochemical content in purslane, including flavonoid acids, and alkaloids. This study aims to determine the difference in flavonoid levels of extracts from soxhletation of purslane (*Portulaca grandiflora*) herb with different solvents.

METHODS

A. Tools and Materials

The equipment used in this research is a soxhletation tool set, rotary evaporator, oven, UV-Vis spectrophotometer, micropipette, measuring flask, volume pipette, and beaker. The ingredients used were purslane herb (Portulaca grandiflora), 96% ethanol, ethyl acetate, n-hexane, quercetin, Mg powder, HCl, AlCl3, aqua dest, methanol, 1M potassium acetate, and10%AlC.

B. Extract Preparation

Purslane herb (Portulaca grandiflora Hook.) has been dried as much as 100 grams, each extracted with 96% ethanol, ethyl acetate, and 250 ml of n-hexane until the active substance is completely extracted. The dregs were then macerated using each solvent of 96% ethanol, ethyl acetate, and 250 mL of n-hexane. The results of the first and second maceration were mixed and evaporated with a rotary evaporator at a temperature of 40°C to obtain a thick extract.

C. Qualitative and Quantitative Test

In this study, the flavonoid test was carried out using a diluted thick extract, 1 ml was taken plus 0.1 grams of Mg powder and 2 ml of HCl, then an intensive red color change was observed for 25 minutes (Harbone, 1987). Quantitative tests were carried out to determine the levels of flavonoid compounds in purslane (Portulaca grandiflora Hook.) herbs using the UV-Vis spectrophotometry method. Quantitative test stages by making blank solutions, measuring lambda max, and making standard solutions of quercetin and sample solutions. Preparation of standard solutions by way of 50 mg quercetin dissolved in 25 ml of pro-analysis methanol then made variations in concentrations of 5 ppm, 10 ppm, 15 ppm, 20 ppm, 25 ppm, and 30 ppm. Two ml of the standard solution of quercetin pipette, add 0.1 ml of 10% $AlCl_3$; 0.1 ml Potassium acetate 1M; 2.8 ml of distilled water and 15 ml of methanol pro-analysis (Kemenkes RI, 2017). The solution was homogenized and left at room temperature for 30 minutes, the absorbance was measured at the maximum wavelength. Preparation of purslane herb sample solution from each thick extract using n-hexane, ethyl acetate, and ethanol extracts was treated the same as making the standard solution of quercetin and replicated 3 times.

D. Data Analysis

Purslane herb flavonoid levels from the three extracts were then analyzed using ANOVA with a 95% confidence level.

RESULTS AND DISCUSSION

Extraction is the process of extracting secondary metabolites from the purslane herb (*Portulaca grandiflora* Hook.). Soxhletation is a method of separating the components contained in samples in the form of solids through repeated filtration using certain solvents using a Soxhlet tool (Pratama *et al*., 2017). Yield is the comparison value between the weight produced and the weight of the raw material multiplied by 100%, the yield value affects the amount of bioactive extract produced (Sani *et al*., 2014). The fresh purslane (*Portulaca grandiflora* Hook.) herbs obtained from Madiun and surrounding areas were sorted and washed, then dried in an oven at a temperature of 50^0C. Dry simplicia was blended, and extracted using the soxhletation method to obtain a thick extract of n-hexane, ethyl acetate, and 96% ethanol as much as 0.61g; 1.23g; 2.22 g so that the yield is 2.3%; 4.1%; 7.4% (Table 1).

Table 1. Purslane (*Portulaca grandiflora* Hook.) Herb Extract Yield

Simplicia	Powder weight (grams)	Extract weight (grams)	Yield (%)
n-heksan extract	30,00	0,61	2,3
etil asetat extract	30,00	1,23	4,1
etanol 96% extract	30,00	2,22	7,4

The viscous extract obtained was then tested for flavonoid compounds qualitatively, each as much as 50 mg then diluted with methanol and filtered. Then 1 ml of the solution was added, 0.1 g of Mg powder, and 2 ml of concentrated HCl were added little by little, the result was that the solution turned red, meaning that the extract contained flavonoid compounds.

Table 2. Qualitative test results extract Purslane (*Portulaca grandiflora* Hook.)

Simplisia	Hasil Uji Kualitatif
N-hexane extract	+
Ethyl acetate extract	+
Etanol 96% extract	+

Extracts of n-hexane, ethyl acetate, and 96% ethanol showed positive results containing flavonoids (Table 2). Research by Budiawan *et al* (2021) confirmed that the content of total flavonoid compounds in purslane herb plays a role in antibacterial activity in the wound healing process in rabbits test animals. In another study conducted by Purwanto (2021), the flavonoid compounds found in purslane herbs have antibacterial activity. Meanwhile, research was conducted by Anghel *et al* (2013) from the *Portulaca grandiflora* Hook. herb showed the content of sterols, carotenes, polyphenolic acids, flavonoids, polysaccharides, and reducing agents. Quantitative test using a UV-Vis spectrophotometer, starting with optimization of the maximum wavelength, namely making a 20 ppm quercetin solution read at a wavelength of 400-500 nm (in the visible region). Flavonoids in spectrophotometry can show strong absorption because they have conjugated aromatic groups (Rais, 2015). Aromatic compounds can be read in the spectrum of ultraviolet light and visible light spectrum (Harborne, 1987).

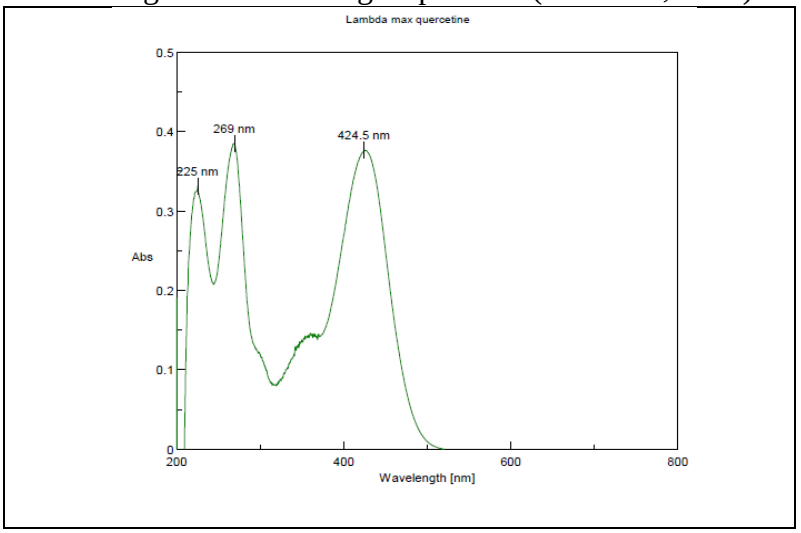

Figure 1. Maximum Wavelength

Preparation of the quercetin series standard to obtain a standard curve. The results of making the standard series can be used to calculate the quercetin content of the sample (Azizah *et al*., 2014).

Each concentration of the standard series made was measured on a UV-Vis spectrophotometer. Average absorbance to obtain the standard curve equation y=bx+a.

Table 3. Preparation of the quercetin series standard

Concentration (ppm)	Absorbance			
	1	2	3	Average
5	0.0540	0.0537	0.0536	0.0538
10	0.1845	0.1821	0.1811	0.1826
15	0.3257	0.3257	0.3253	0.3255
20	0.4589	0.4588	0.4588	0.4588
25	0.5714	0.5715	0.5719	0.5716
30	0.7178	0.7180	0.7178	0.7179

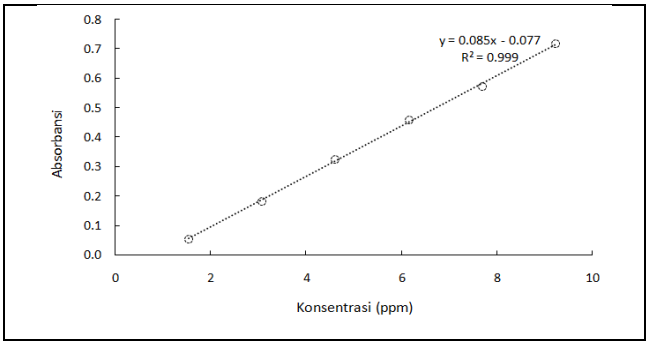

Figure 2. Standard Curve Equation

Treatment of samples of 96% ethanol extract, ethyl acetate, and n-hexane in the same way as the preparation of comparison standards then read the absorbance at a maximum wavelength of 424.5 nm and replicated three times (Kemenkes RI, 2017).

Table 4. Results of sample flavonoid content

Sample	Replication	Absorbance	Content (% w/w)
etanol 96% extract	1	0,3010	11,636
	2	0,3026	11,684
	3	0,3027	11,688
etil asetat extract	1	0,6775	23,222
	2	0,6797	23,290
	3	0,6803	23,310
n-heksan extract	1	0,5042	17,890
	2	0,5052	17,920
	3	0,5056	17,930

This study showed the highest levels of flavonoids in purslane herb in the ethyl acetate extract of 23.27% w/w, while the flavonoid content of the n-hexane extract was 17.91% w/w and the ethanol extract was 11.67% w/w. The polarity of the solvent at the time of extraction affects the levels of flavonoids so that solvents with simplicia that have the same polarity will easily absorb secondary metabolites in the solvent (Winahyu *et al.*, 2018). Purslane herb flavonoid levels from the three extracts were then analyzed using ANOVA which resulted in all data having significant differences with sig < 0.05.

CONCLUSION

The flavonoid content of purslane (*Portulaca grandiflora* Hook.) herb using 96% ethanol, ethyl acetate, and n-hexane as solvent was 11.67% w/w; 23.27% w/w; 17.91% w/w, all of which are significantly different.

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