**QUERCETIN AND PIPERINE CONTENT OF FRACTION N-HEXANE
SESEWANUA LEAF (*CLERODENDRUM FRAGRANS* WILD.)**Arlan K. Imran^{1*)}, Zulfiayu¹⁾, Ahamd Aswad²⁾¹⁾Department of Pharmacy, Gorontalo Ministry of Health Polytechnic.²⁾Department of Nursing, Gorontalo Ministry of Health Polytechnic.*Email: arlan_imran@poltekkesgorontalo.ac.id**Detail Artikel**

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Kata Kunci*Sesewanuwa**Piperine**Quercetin**n-Hexane**Fractionation***Penulis Korespondensi**

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Sesewanua (Clerodendrum fragrans Wild.) is used as an anti-inflammatory (anti-rheumatoid arthritis) topically and orally and as an antioxidant with very strong potential. The n-hexane fraction: ethyl acetate of sesewanua leaves contains secondary metabolites of flavonoids and alkaloids. This study aimed to measure the levels of the flavonoid quercetin and the levels of the alkaloid piperine in the n-hexane: ethyl acetate fraction of the ethanol extract of sesewanua leaves. This research method: The n-hexane:ethyl acetate fraction was determined for its flavonoid content at a wavelength of 256 nm and piperine alkaloids at a wavelength of 342.5 nm using the UV-Vis spectrophotometry method. The results obtained from the n-hexane:ethyl acetate fraction of the ethanol extract of

sesewanua leaves contained the flavonoid compound quercetin as much as 6.385 PPM and the alkaloid compound piperine as much as 5.854 PPM. Conclusion: The statistical results of the Independent T. Test show that there are significant levels of the flavonoid quercetin and alkaloid piperine, which means that the levels of the flavonoid quercetin in the n-hexane: ethyl acetate fraction of the ethanol extract of sesewanua leaves are higher than the alkaloid piperine compound.

INTRODUCTION

Sesewanua (*Clerodendrum fragrans* Wild.) is a plant that is commonly used empirically by people as an anti-inflammatory. Research related to sesewanua activity has been carried out in stages by researchers majoring in pharmacy at the Gorontalo Ministry of Health Polytechnic, including Sapiun and Pangalo 2019, they found that the ethanol extract of sesewanua leaves has activity as an anti-rheumatism. Sapiun et al also found that there were 4 major compound components in Sese Wanua's ethanol extract, including flavonoids and alkaloids. Subsequent research by Sapiun *et al.* (2020) found that the ethanol extract of sesewanua leaves has anti-cancer activity. In 2021 Sapiun et al were able to separate and find 1 of the most active compounds using varying volumes of the solvent *n*-hexane:ethyl acetate.

Fraction *n*-hexane:ethyl acetate. Sesewanua leaf ethanol extract has high antioxidant activity with an IC 50 value of 2.5 ppm (very strong potential). Research in 2022, Sapiun et al formulated this compound in a pharmaceutical preparation using nano technology, namely the *Self Nano Emulsifying Drug Delivery System* (SNEDDS), which is efficacious as an oral and topical anti-rheumatoid arthritis. In 2022 Sapiun et al discovered the *n*-hexane:ethyl acetate fraction. Sesewanua leaf ethanol extract contains 2 large compound components, namely flavonoids and alkaloids.

According to Maburroh *et al* (2019) and Mutmainah (2017) the use of solvent *n*-hexane:ethyl acetate is able to separate stains well. The compounds that can be extracted using solvent *n*-hexane:ethyl acetate include flavonoids and alkaloids (Rubiyanti et al., 2019). Flavonoids are a group of natural compounds that have a variable phenolic structure and alkaloids are compounds that have many nitrogen atoms (Widiasari 2018; Maisarah et al, 2023). To determine the levels of flavonoids and alkaloids in the *n*-hexane:ethyl acetate fraction of sesewanua (*Clerodendrum fragrans* Wild.) leaves, it is necessary to determine the levels using UV-Vis spectrophotometry.

Based on the above background, research was conducted to determine the levels of quercetin and piperine fraction *n*-hexane : ethyl acetate, ethanol extract of sesewanua leaves (*Clerodendrum fragrans* Wild.).

RESEARCH METHODE

This research method is a laboratory experiment where the simplisia of sesewanua leaves that have been extracted by maceration method using ethanol solvent has been fractionated using column chromatography with *n*-hexane solvent: ethyl acetate ratio (10:0); (9:1); (8:2); (7:3) combined into 1. *n*-hexane fraction: Ethyl Acetate ethanol extract leaves have been determined levels of flavonoids (quercetin) and alkaloids (piperine) with a standard curve $y=bx\pm a$ each of which has been made quercetin at 12 concentration points, namely at 2 ppm, 3 ppm, 4 ppm, 5 ppm, 6 ppm, 7 ppm, 8 ppm, 9 ppm, 10 ppm, 20 ppm, 24 ppm and 50 ppm. As well as the standard curve of piperine at 6 concentration points, namely 1 ppm, 2 ppm, 3 ppm, 4 ppm, 6 ppm and 10 ppm at wavelengths of 256 nm for quercetin compounds and 342.5 nm for wavelengths of piperine compounds using a spectrophotometer UV-Vis (Huliselan *et al.* 2015; Rusman 2020; Sari *et al.* 2021; Hikmawanti et al. 2021; Imran et al. 2019)

RESULTS AND DISCUSSION

The determination of the maximum wavelength has been carried out using a UV-Vis Spectrophotometer and maximum wavelength results are obtained. The maximum wavelength obtained is used to obtain maximum absorption sensitivity results to quercetin and piperine compounds. This is according to the Lambert-Beer law, the intensity of light absorbed will be directly proportional to the concentration read. Thus, the maximum absorbance that has been obtained has a very small error rate (Ibrani 2016; Ahriani *et al.* 2021). The results of the measurement of the raw absorbance of quercetin flavonoids obtained can be seen in Figure 1.

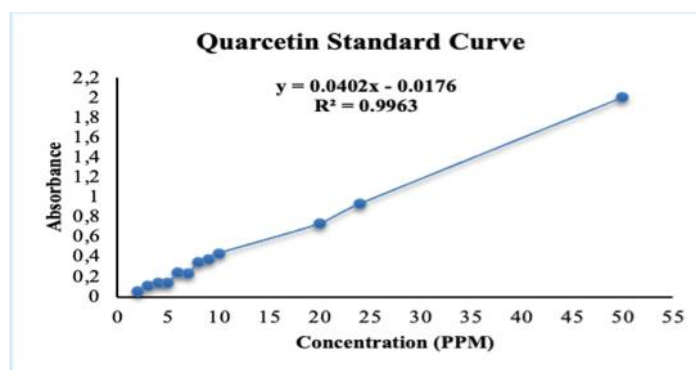


Figure 1. Quercetin Standard Curve

The regression equation $y = 24.79x + 0.481$ at the wavelength 256 nm obtained has a correlation coefficient value of 0.9963. This value shows that the standard curve of quercetin flavonoids obtained has good linearity. A value of r^2 close to 1 indicates that the linear calibration curve has a relationship between the concentration of quercetin solution and the absorbance value. The results of the standard absorbance measurement of piperine alkaloids obtained can be seen in Figure 2.

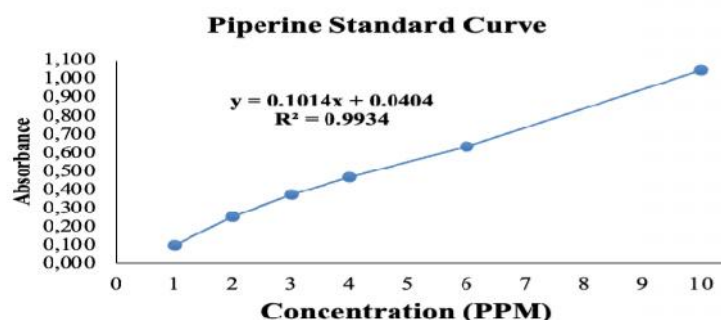


Figure 2. Piperine Standard Curve

The regression equation $y = 0.1014x + 0.0404$ at the wavelength of 342.50 nm obtained has a correlation coefficient value of 0.9934. This value shows that the standard curve of quercetin flavonoids obtained has good linearity. A value of r^2 close to 1 indicates that the linear calibration curve has a relationship between the concentration of quercetin solution and the absorbance value.

Table 1. Results of Flavonoid (Quercetin) and Alkaloid (Piperine) Levels

Compound	PPM rate	Percentage Rate
Quercetin	6.3856	0.000638557
Piperine	5.8540*	0.000585404

*Significant to Quercetin (Independent Sample T Test)

Based on table 1. The solvent *n*-hexane; ethyl acetate may attract quercetin flavonoids better than piperine alkaloids. The results of statistical tests using the Independent Sample T Test obtained a sig value of <0.05, which means that the difference in the results of quercetin and piperine levels that have been obtained in fractions is due to the ability of the solvent to attract compounds. The results obtained are in accordance with the research of Satria, Hakim, and Darsono (2022) *n*-hexan solvent is able to attract more flavonoid compounds by 2.563%, while in the research of Utami, Arruansaratu, and Jumaetri (2022) *n*-hexan solvent attracts alkaloid compounds only by 0.873% (Liu, Zhang, dan Yuan 2022; Usman, Abu Bakar, dan Mohamed 2016; Sapiun et al. 2024; Sapiun, Pangalo, Imran, et al. 2020)

CONCLUSIONS

The fraction *n*-hexane: ethyl acetate contains quarcetin 6,385 PPM (0.0006385%) more than piperine 5,854 PPM (0.0005854%). Suggestions for further research are to carry out structural identification of quarcetin and piperine compounds present in the fraction *n*-hexane:ethyl acetate of sesewanua leaves.

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