



Determination of Total Flavonoid Content of Ashitaba (*Angelica keiskei* Koidz.) Leaf Extract Using UV-Vis Spectrophotometry

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Abstract: Ashitaba (*Angelica keiskei* Koidz.) is a medicinal plant known to be rich in active flavonoid compounds, particularly chalcones, which exhibit antioxidant, anti-inflammatory, and antidiabetic activities. This study aimed to determine the total flavonoid content in Ashitaba leaf extract obtained from Sembalun Village using UV-Vis spectrophotometry. The extract was obtained by maceration using 96% ethanol. Quantitative analysis was performed at the maximum wavelength of 415 nm, with quercetin as the standard. Calibration curves were prepared using quercetin concentrations of 20, 30, 40, 50, and 60 ppm on the X-axis, with absorbance values of 0.297, 0.337, 0.436, 0.509, and 0.588 on the Y-axis, resulting in the regression equation $Y = 0.0094x + 0.0472$ with a correlation coefficient (r) of 0.9677, indicating a linear relationship between concentration and absorbance. Absorbance values of the sample measured in triplicate were 0.661, 0.634, and 0.676, yielding an average total flavonoid content of 25.948 mgQE/g in Ashitaba leaf extract.

Keywords: Ashitaba (*Angelica keiskei* Koidz.); Ethanol extract; Total flavonoid; Maceration; UV-Vis spectrophotometry.

Introduction

Ashitaba (*Angelica keiskei* Koidz.) is a medicinal plant native to Japan and is widely consumed as a functional vegetable (Singh et al., 2024; Wahyuni et al., 2024). In its country of origin, Ashitaba is valued for both its nutritional and therapeutic benefits. In contrast, Ashitaba remains relatively unfamiliar in Indonesia as an introduced plant species. Limited public awareness has contributed to its underutilization in local communities (Chowdhury et al., 2025). Several studies have demonstrated that Ashitaba contains various bioactive compounds. Among these compounds, chalcones are the most prominent and are classified as flavonoids (Ji et al., 2025; Siswanto et al., 2024).

Chalcones derived from Ashitaba have been reported to exhibit antitumor activity. In addition, these compounds possess antibacterial, antifungal, and anti-inflammatory properties (Silva et al., 2025). Ashitaba chalcones are also associated with stimulating red blood cell production and enhancing immune system function.

Furthermore, Ashitaba has been reported to exhibit antihypertensive effects and may contribute to the prevention of stroke. Beyond these effects, chalcones isolated from Ashitaba, including xanthoangelol and isobavachalcone, have been shown to exhibit strong antioxidant activity (Ji et al., 2022; Wang et al., 2024). These compounds also demonstrate antibacterial properties against various pathogenic microorganisms. In addition, pro-apoptotic activity of Ashitaba chalcones has been reported in neuroblastoma cells. Similar cytotoxic effects have also been observed in leukemia cell lines (Sheikh et al., 2025).

Phytochemical screening of different plant parts, including roots, stems, leaves, and flowers, revealed a diverse range of secondary metabolites (Nadeem et al., 2025). The identified compounds include alkaloids, flavonoids, phenolics, and tannins. Among these plant parts, the leaves exhibit the highest antioxidant capacity due to their superior free radical scavenging activity compared to stems and roots (Kellogg, 2022; Pardo & Paranada, 2025). Flavonoids are one of the largest

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groups of plant polyphenols and have been extensively studied for their health-promoting roles, including antioxidant, anti-inflammatory, antibacterial, and chemopreventive effects. Their antioxidant action primarily involves inhibiting reactive oxygen species (ROS) generation and enhancing endogenous antioxidant regulation. Several flavonoids have also been shown to lower blood glucose through mechanisms involving improved insulin sensitivity and inhibition of α -glucosidase enzymes. Thus, flavonoids are promising therapeutic agents for chronic diseases, including diabetes and cancer (Flores-Bocanegra et al., 2025; Thozhukkad Moosarippambal & Vadakkadath Meethal, 2025).

Given the lack of public awareness and limited scientific documentation on the phytochemical properties of Ashitaba cultivated in Indonesia, this study addresses an important research gap. In particular, information regarding Ashitaba grown in the Sembalun region remains scarce (Arshad et al., 2025). Environmental factors such as altitude, climate, and soil conditions may influence the accumulation of bioactive compounds in medicinal plants. Therefore, phytochemical profiles of Ashitaba cultivated in different regions may vary significantly (Mahmoud & Selim, 2025).

Flavonoids are among the most important bioactive compounds due to their antioxidant and therapeutic properties. Determining the total flavonoid content is essential for evaluating the medicinal potential of Ashitaba. This study specifically focuses on quantifying the total flavonoid content of Ashitaba cultivated in Sembalun. The findings are expected to provide baseline data for future pharmacological studies. Establishing flavonoid concentration can support the standardization of Ashitaba as a functional plant. Scientific evidence regarding flavonoid content may also enhance its acceptance in the health sector (Eghlima et al., 2025; Fahmy et al., 2025). Moreover, such data can contribute to the development of herbal-based products. The results may encourage wider utilization of Ashitaba among local communities. Increased scientific documentation can improve public awareness of its health benefits. This study also supports the conservation and sustainable cultivation of Ashitaba in Indonesia (Aminzai et al., 2025; Song et al., 2025). Overall, the research is expected to strengthen the scientific foundation for the potential health-related applications of Ashitaba.

Method

Research Design

This study employed a laboratory experimental design with full replication. All experimental

procedures were carried out under controlled laboratory conditions. Each analytical step was conducted in triplicate to minimize random error. Replication was applied consistently across all samples and measurements. This approach was intended to enhance data reliability and ensure the accuracy of the results (Bauer et al., 2025; Harmon-Jones et al., 2025).

Participants / Sample

The study sample consisted of 200 g of dried Ashitaba leaf powder (*Angelica keiskei* Koidz.). The plant material was collected from Sembalun Village, East Lombok, Indonesia. The leaves were selected based on uniform maturity and physical condition. Prior to analysis, the plant material was processed to ensure homogeneity. This sample preparation was intended to obtain representative and reliable analytical results (Xu et al., 2025).

Extraction Procedure

Two hundred grams of Ashitaba leaf powder were extracted by maceration in 96% ethanol (ratio 1:10) for 72 hours with occasional stirring. The filtrate was collected and remacerated for 24 hours to maximize yield. The combined filtrates were concentrated with a rotary evaporator at 50 °C and further evaporated in a water bath until a thick extract was obtained (Halid & Rahmawati, 2023).

Identification of Flavonoids

Flavonoids were qualitatively confirmed using the Mg-HCl test, indicated by the appearance of a red or orange color (Shraim et al., 2021).

Determination of Total Flavonoid Content:

Quantitative analysis was carried out by preparing a quercetin standard curve (20–60 ppm). One milliliter of standard or sample solution was mixed with 1 mL of 10% $AlCl_3$ and 8 mL of 5% acetic acid, then measured at λ_{max} 415 nm using a UV-Vis spectrophotometer. The regression equation obtained was used to calculate total flavonoid content expressed as mg quercetin equivalents per gram extract (mgQE/g) (Shraim et al., 2021).

Result and Discussion

Extraction Yield

The maceration of 200 g of Ashitaba leaf powder yielded 29 g of thick extract, corresponding to a yield of 14.5%. This yield indicates efficient extraction of soluble compounds from the plant material. The obtained value falls within the commonly reported range for herbal maceration processes (Auli et al., 2025; Castellanos et al., 2025). Previous studies have documented extraction yields ranging from 10% to 15% for similar medicinal

plants. Therefore, the yield achieved in this study can be considered satisfactory. Factors such as solvent type, extraction duration, and particle size may influence extraction efficiency. Overall, the results suggest that the maceration method used was appropriate for extracting bioactive compounds from Ashitaba leaves (Siswanto & Andarisuta, 2025; Tchamgoue et al., 2023).

Table 1. Extraction Yield of Ashitaba Leaf Extract

Sample weight (g)	Extract weight (g)	Yield (%)
200	29	14.5

Total Flavonoid Content

The quercetin calibration curve showed a linear relationship with the regression equation $Y = 0.0094x + 0.0472$ ($r = 0.9677$). The average absorbance values of Ashitaba extract replicates (0.661, 0.634, 0.676) corresponded to a total flavonoid content of 25.948 mgQE/g extract.

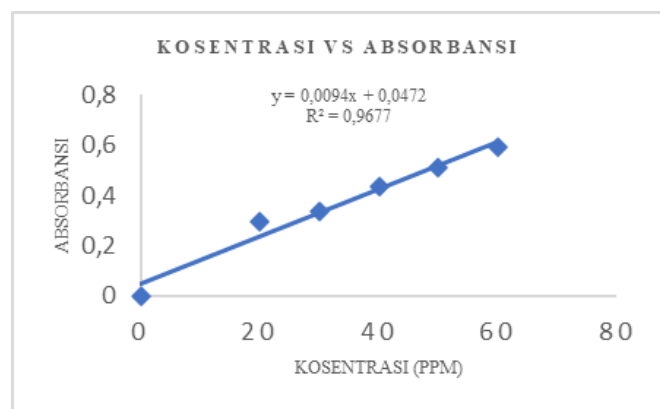


Figure 1. Quercetin Calibration Curve (20–60 ppm)

The flavonoid content obtained (25.948 mgQE/g) was significantly higher compared to the reported flavonoid content in Batak onion extract (1.5467 mgQE/g), suggesting that Ashitaba is a rich natural source of flavonoids (Dhikale et al., 2026; Singh et al., 2025). Differences in flavonoid content across studies may be attributed to environmental factors, extraction solvents, and post-harvest handling (Xu & Wang, 2025).

The use of 96% ethanol proved effective for flavonoid extraction from Ashitaba leaves. High-concentration ethanol is known to dissolve polar compounds, such as quercetin, more efficiently than lower concentrations (Lestari et al., 2025; Rocha et al., 2025). In contrast, 70% ethanol may extract a broader range of compounds but with reduced selectivity for flavonoids. The higher polarity of 96% ethanol enhances the solubility of flavonoid aglycones (Kubiak et al., 2025; Sun et al., 2025). This finding is consistent with previous studies reporting that ethanol concentration

significantly influences flavonoid yield. Optimal solvent selection is therefore a critical factor in phytochemical extraction. Overall, the use of 96% ethanol contributed to improved efficiency in isolating flavonoid compounds in this study (Bose et al., 2025; Lemanowicz et al., 2025).

These findings highlight the pharmacological relevance of Ashitaba leaf extract. Given flavonoids' well-documented antioxidant and antidiabetic properties, Ashitaba holds promise as a potential raw material for herbal medicines and functional foods (Miyamoto et al., 2025).

Conclusion

This study successfully quantified the total flavonoid content in Ashitaba (*Angelica keiskei* Koidz.) leaf extract using UV-Vis spectrophotometry. The extract yielded 25.948 mgQE/g, confirming that Ashitaba leaves are a significant natural source of flavonoids. These results support the potential application of Ashitaba as a functional ingredient with antioxidant and therapeutic properties for degenerative disease prevention.

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Conflicts of Interest

The authors declare no conflict of interest.

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