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Secondary Metabolite Content and Antioxidant Activity of Ethanol Extract of Tali Putri (*Cuscuta australis*) at Different Heights Places

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Abstract

Cuscuta australis (commonly known as Tali Putri) is a parasitic weed that extracts water, minerals, and nutrients from its host plants to sustain its life cycle. Among its shared hosts, *Asystasia gangetica* (Israel grass) is highly susceptible due to its soft stem structure, which facilitates haustorial penetration and efficient nutrient transfer. Although generally regarded as a harmful weed, *C. australis* has demonstrated pharmacological potential owing to its secondary metabolites and antioxidant properties, both of which are relevant in disease treatment. This study investigated the effect of altitude on the phytochemical profile, antioxidant activity, moisture content, and ash content of *C. australis* and its host plant. A survey method was applied across three altitudinal zones (lowland, midland, and highland) with seven replications per site. Laboratory analyses were performed at the Plant Physiology Laboratory, Faculty of Agriculture, Andalas University, and the Higher Education Service Institution (LLDIKTI) Laboratory. Data were analyzed using analysis of variance (ANOVA) with the F-test, and significant differences ($p < 0.05$) were further evaluated using Duncan's New Multiple Range Test (DNMRT). The results showed that *C. australis* grown at high altitude exhibited the highest antioxidant activity (173.35 µg/ml), moisture content (9.06%), and ash content (5.16%). Similarly, *A. gangetica* collected from highland sites demonstrated superior antioxidant activity (64.39 µg/ml), moisture content (8.82%), and ash content (7.07%). By contrast, altitude had no detectable effect on the phytochemical composition of either species, both of which consistently contained flavonoids, alkaloids, phenolics, terpenoids, and triterpenoids. These findings indicate that highland populations of *C. australis* and *A. gangetica* possess enhanced bioactive properties, suggesting that high-altitude habitats may provide the most suitable raw material sources for pharmaceutical applications.

Keywords: Altitude, Antioxidant, Secondary metabolites, Tali Putri

1. Introduction

Cuscuta australis (commonly known as Tali Putri or Talli Putri) is a parasitic weed that depends on its host plant for water, minerals, and nutrients. Morphologically, it is distinguished by its rope-like growth habit, golden-yellow coloration, and creeping stems that twine extensively around the host, often causing significant damage (Hidayat *et al.*, 2017). Among its preferred hosts, *Asystasia gangetica* (Israeli grass) is particularly susceptible due to its soft stem structure, which facilitates haustorial penetration and supports vigorous parasitic growth. Israeli grass itself contains diverse phytochemicals, including flavonoids, alkaloids, phenolics, steroids, terpenoids, and saponins (Utami *et al.*, 2025).

The parasitism of *C. australis* is mediated through haustoria, specialized multicellular organs that establish connections with the vascular tissues (xylem and phloem) of the host. This interface enables efficient transfer of water, nutrients, and secondary metabolites from host to parasite (Kokla & Melnyk, 2018). Haustoria, which resemble fine hairs anchoring to the host, are critical for the survival of *C. australis* but simultaneously weaken the host plant, often reducing productivity and, in severe cases, leading to host mortality.

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Despite its parasitic nature, *C. australis* possesses notable medicinal value. In traditional Chinese medicine, related *Cuscuta* species have been used for centuries to treat kidney disorders, back pain, and skin infections (Cyril *et al.*, 2020). Ethnobotanical reports further indicate that *C. australis* has been employed in the treatment of gonorrhea, kidney disease, and as a natural antioxidant (Mythili *et al.*, 2011). These therapeutic applications are attributed to its rich phytochemical composition and antioxidant activity, highlighting its potential as a source of bioactive compounds.

Phytochemical compounds, a component of plant chemistry, are produced through the biosynthesis of metabolite compounds, Which Are Primary metabolites offering benefits for both plant and human health (Maslahah & Nurhayati, 2024). Phytochemical compounds come from secondary metabolites produced by plants. Secondary metabolites are produced by plants in small amounts. However, plants themselves have several functions, including serving as an attractant (attracting other organisms), defense against pathogens, adaptive protection against environmental stress, protection against ultraviolet rays, acting as a growth regulator, and competing with other plants (Tampubolon et al., 2018). For health, secondary metabolites act as antioxidants because they contain phytochemical compounds such as flavonoids, saponins, triterpenoids, and phenolics (Giri *et al.*, 2021).

The content of secondary metabolites, specifically flavonoid compounds, offers benefits in treating various diseases, including cancer, hypertension, and stroke, and serves as an antioxidant (Ballard & Maróstica, 2019). Saponins are naturally occurring surface-active glycosides, mainly produced by plants, whose structure consists of sugar groups bound to hydrophobic aglycones. Saponin compounds have benefits in the world of pharmacology because they are known to have antibacterial, anti-inflammatory, anticancer, and antitumor activities (Marrelli et al., 2016). Triterpenoids have significant pharmacological activities such as antiviral, antibacterial, anti-inflammatory, and anticancer (Ramadhan *et al.*, 2023). Phenolic compounds function as anti-obesity, antibacterial, antimicrobial, and antioxidants. The antioxidant properties of phenolic compounds are related to their molecular structure, more precisely to the presence and number of hydroxyl groups, as well as conjugation and resonance effects (Hidayat *et al.*, 2017).

Antioxidants are compounds that play a crucial role in protecting body cells from damage caused by free radicals. They achieve this by capturing radical molecules, thereby inhibiting oxidative reactions that lead to various diseases. Testing of antioxidant activity and phytochemical content of plants was carried out using ethanol extract because it is an effective solvent in extracting various bioactive compounds such as phenolics, flavonoids, alkaloids, steroids, saponins, and phenolics (Riwanti & Izazih, 2019). Ethanol has good penetration ability, so it can dissolve active compounds that act as antioxidants (Widyasanti *et al.*, 2016).

The value of antioxidant activity can be measured using the DPPH method. (2,2- *diphenyl* -1- *picrylhydrazyl*) This method is one of the widely used approaches for determining antioxidants, producing more accurate results than other methods. Principles: The DPPH method is a hydrogen capture reaction by DPPH free radicals from antioxidants. Absorbance is measured using a spectrophotometer at the maximum wavelength. The antioxidant activity value can be seen from the IC50 value. (*Inhibitory Concentration*) This parameter, commonly used in biochemistry and pharmacology, evaluates the effectiveness of a substance in inhibiting enzyme activity or cell growth. Specifically, it indicates the concentration of a substance required to inhibit enzyme activity or cell growth by 50% compared to conditions without an inhibitor. (Caldwell *et al.*, 2012) . Meanwhile, the phytochemical content can be seen from the color change that occurs when the test solution is added.

Antioxidant and phytochemical activities are influenced by host type, host number, physical condition, genetics, environmental stress factors, and altitude. Arnelio's research (2024) stated that Tali Putri contains flavonoid, saponin, triterpenoid, and phenolic phytochemical compounds and has antioxidant activity displayed in the form of IC50 values, namely 163 µg/ml to 279 µg/ml. In addition, Sidhu *et al.'s (2022)* research obtained IC50 values of Tali Putri with ethanol solvent (118.5 µg/ml), methanol solvent (123.2 µg/ml), and n-Hexane solvent (117.9 µg/ml). Different environmental conditions greatly affect the production of secondary metabolite compounds in plants (Qaderi et al., 2023). Altitude is one aspect that causes differences in climate elements, such as air temperature, rainfall, humidity, and air pressure. According to Ohmura (2012), each altitude has its own environmental characteristics that will affect the phytochemical content, water content, ash content, and biochemical processes in plants that have an impact on plant antioxidant activity. Lestari *et al.* (2024) reported that the antioxidant activity of the Plant Hippobroma longiflora, growing at various altitudes, shows the highest activity found in the lowlands.

Environmental factors, particularly altitude, may influence the biochemical properties of parasitic plants and their hosts. Altitude in Indonesia is generally categorized into three zones: lowland (0–400 masl), midland (400–700 masl), and highland (>700 masl) (Istiawan & Kastono, 2019). Both *C. australis* and *A. gangetica* are capable of surviving across these altitudinal ranges, offering a natural gradient for studying environmental effects on plant biochemistry.

Based on these considerations, the present study, titled *"Identification of Phytochemical Content and Antioxidant*

Activity of Ethanol Extract of Cuscuta australis at Different Altitudes", was conducted to evaluate the influence of altitude on the phytochemical profile, antioxidant activity, moisture content, and ash content of *C. australis* and its host plant, *A. gangetica*.

2. Material and Methods

2.1. Time and place

This research was conducted from January to June 2025. Sampling of Tali Putri and host plants was done in the same place with three different altitudes, namely in Nagari Alahan Panjang, Lembah Gumanti sub-district, Solok Regency (highlands >1000 masl), Nagari Malalak Timur, Malalak sub-district, Agam Regency (middle plains 400-700 masl), and Nagari Sungai Sarik, VII Koto sub-district, Padang Pariaman Regency (lowlands 0-400 masl). The drying and extraction process was carried out at the Plant Physiology Laboratory, Faculty of Agriculture, Andalas University. The phytochemical testing process was carried out at the Organic Chemistry Laboratory, Faculty of Mathematics and Natural Sciences, Andalas University, and the antioxidant activity test was carried out at the Laboratory of the Higher Education Service Institute (LLDIKTI), region 10. The following are the coordinates of the locations where the Talli Putri and host plants grow, with different terrains and seven replications in each terrain, as detailed in Table 1.

Table 1. Coordinates of the locations where the Talli Putri and host plants grow

Test	Low	Currently	Tall
1	0°34'27.5"S	0°25'37.3"S	1°04'41.4"S
	100°13'16.9"E	100°15'54.5"E	100°47'13.5"E
2	0°34'21.5"S	0°25'23.9"S	1°04'57.9"S
	100°13'08.8"E	100°16'16.9"E	100°47'19.2"E
3	0°34'28.3"S	0°25'03.4"S	1°05'02.9"S
	100°13'08.2"E	100°16'19.1"E	100°47'15.8"E
4	0°34'33.0"S	0°24'53.9"S	1°05'00.0"S
	100°13'10.7"E	100°16'29.1"E	100°47'03.5"E
5	0°34'33.5"S	0°24'41.5"S	1°04'51.0"S
	100°13'00.6"E	100°16'32.5"E	100°46'47.8"E
6	0°34'23.1"S	0°24'30.8"S	1°04'41.8"S
	100°13'22.0"E	100°16'36.4"E	100°46'46.8"E
7	0°34'29.0"S	0°24'13.0"S	1°04'31.1"S
	100°13'27.2"E	100°16'51.0"E	100°46'28.2"E

2.2. Tools and materials

The tool used in this research is a UV-Vis spectrophotometer (Hitachi), Buchi rotary evaporator (HeildopH Laborota 4000), oven (Haraeus), *micropipette* (Dragonlab), furnace, plate, dropper, stirring rod, hotplate, test tube, vortex *(DLAB vortex mixer MX-S*), desiccator, analytical balance (*analytical balance Fujitsu*), 300 ml glass bottle, 10 ml glass bottle, five volumetric flask ml, 25 ml measuring flask, 10 ml measuring flask, reagent bottle, measuring cup, test tube, porcelain cup, blender, *whatman* paper no. 1, funnel, knife, tray, mobile phone and stationery.

The materials used were host plants, namely Israeli grass (*Asystasia gangetica*), and Talli Putri (*Cuscuta australis*), water, distilled water and chemicals used in this study were DPPH (2,2- *diphenyl-1-picrylhydrazyl*), methanol *pa, ethanol* $_{96}$ %, CH3OH, HCl, H_2SO^4, chloroform, Mg powder, acetic anhydride, ammonia, Meyer, Dragendorff, FeCl3, cotton, tissue and *aluminum foil.*

2.3. Research methods

This research used a survey method, which included surveys at three altitude categories: lowlands, midlands, and highlands. Seven replicates were taken at each altitude, resulting in 21 samples for Tali Putri and 21 samples for Israel grass.

2.4. Implementation of research

The implementation of the research can be seen in Figure 1.

2.4.1. Host plant exploration

This research employed an experimental method, starting with an exploration of the presence of host plants. The host plant identified was Israel grass. (*Asystasia gangetica*) The study was conducted at three elevations, with seven replicates at each elevation. The criteria for Israeli grass used as a host were a height of 50-60 cm, thriving, and resistance to pests and diseases.

2.4.2. Taking samples of host plants

Host plant samples were collected by taking plant parts, namely leaves and stems. The host plant's growing season is January and February. Afterward, drying was carried out, and the plant's simplicia and extract were prepared. Furthermore, water content, phytochemical content, and antioxidant activity were tested, similar to those used for the Tali Putri (lace plant).

2.4.3. Inoculation of the daughter cord

Inoculation of the Talli Putri was carried out in March, when the host plant, Talli Putri (*Cuscuta australis*), was sampled and inoculated on Israeli grass. (*Asystasia gangetica*) . The process of inoculating the Talli Putri involves taking a rope-shaped object and cutting it to a length of 40-50 cm. It is then wrapped around the stem or base of the plant's leaves. After 2 days of wrapping, the filamentous plant will automatically wrap around the host plant and continue to grow. Checks are carried out weekly on the filamentous plant to ensure it is growing well and not wrapping around other plants. After 7-8 weeks, the filamentous plant will have grown well and abundantly on the host plant, and samples of the filamentous plant are taken.

2.4.4. Sampling

Plant samples were collected at three different altitudes in April, with 300 g of each host plant part (stems and leaves) taken. For the Tali Putri sample, 300 grams were collected from each of the three locations with different heights. The criteria for selecting the Talli Putri for testing are that it has grown significantly on the host, is sufficiently mature, and is aged 7-8 weeks post-inoculation. We obtained 42 test samples from both the Talli Putri and the host plants.

2.4.5. Process of Making Simple Drugs

The process of making simplicia from host plants involves taking the stem and leaves of the host plant. In contrast, for the female Tali Putri, all parts of the Tali Putri are taken. After that, the host plant and Tali Putri are taken to the Laboratory. The next step is to separate the dirt attached to the plant that was carried during sample collection before washing. Then, the study conducted washing, cutting, and drying at a temperature of 50°C for 24 hours.

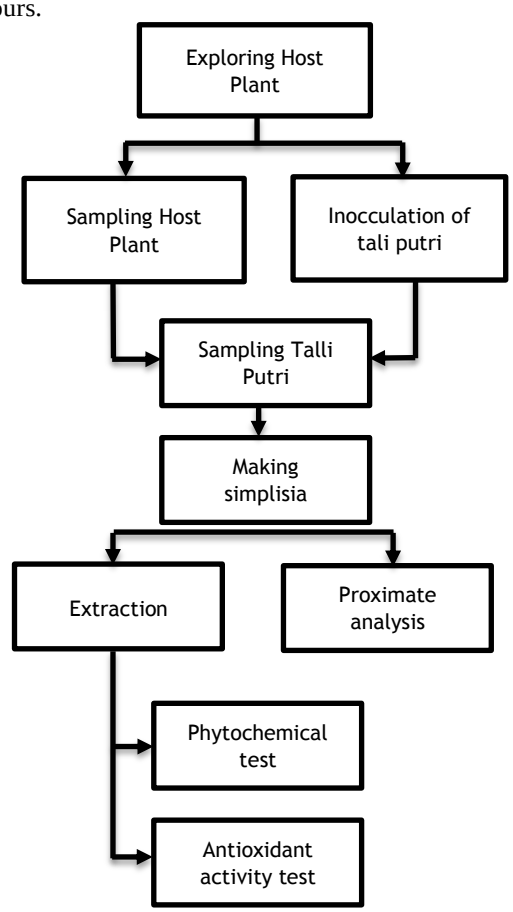

Figure 1. Research implementation process.

Dry sorting involves separating foreign objects that were carried along during the drying process. Host plants and princess tali are sorted, dried, and then blended in a blender to grind. The powdered herbs are then placed in glass bottles, covered with *aluminum foil*, and labeled according to the treatment.

2.4.6. Extraction

Simplicia powder was weighed to a maximum of 30 grams for each sample and placed in a glass bottle. Subsequently, 300 ml of 96% ethanol solvent was added, and the mixture was left in a dark place for 24 hours, with stirring every 6 hours. Next, the solution was filtered and separated into dregs and extracts using Whatman paper no. 1. This process yielded the filtrate and dregs of the extract from the Talli Putri and host plants. Then, the extract was evaporated with *a rotary evaporator* at a temperature of 40°C until a thick extract was obtained. The extract is a thick preparation made using the maceration method. The thick extract is stored in a glass bottle and covered with *aluminum foil* (Rahayu *et al.*, 2020).

2.5. Data analysis

Phytochemical test results data are displayed quantitatively, while water content, ash content, and IC50 data are analyzed using the Statistical Tool for Agricultural Research (STAR) version 2.0.1. The observation data were analyzed using analysis of variance with the F test. If the calculated F value exceeds the F table value at 5%, the analysis proceeds with *Duncan's New Multiple Range Test* (DNMRT).

2.6. Observation variables

2.6.1. Water content

Empty porcelain cups were dried in an oven for 30 minutes or until a constant weight was obtained, then cooled in a desiccator for 15 minutes and weighed. Each sample and its replication were weighed up to 2 grams, placed in a cup, and then heated in an oven for 2 hours at 110°C. They were then weighed again and heated for an additional 2 hours at the same temperature. The sample was heated until a constant weight was obtained. If the sample had not reached a constant weight, the sample was ovened again for 1 hour until a constant weight was obtained (Wandira *et al.*, 2023). The percentage of water content can be measured as follows:

$$\%\text{Water content} = \frac{(W1-W0)-(W2-W0)}{(W1-W0)} \text{x } 100\%$$

Information:
W2 : sample weight + porcelain cup after heating
W0 : weight of empty porcelain cup
W1 : initial sample weight (grams)

2.6.2. Ash content

The porcelain cup is dried in an oven at a temperature of 110 °C for 1 hour, then cooled in a desiccator for 15 minutes until a constant weight is obtained. Each sample and replicate was weighed at 2 grams, placed in a porcelain cup, and then heated in a furnace at 500 °C for 5 hours to obtain the ash. The ash was then placed in a desiccator for 30 minutes, after which it was weighed (Sudarmadji et al., 1984). The ash content calculation is carried out as follows:

$$\%\text{Ash content} = \frac{W2-W0}{W1-W0} \text{x } 100\%$$

Information:
W0 : weight of empty porcelain cup
W1 : initial sample weight (grams)
W2 : sample weight + porcelain cup after heating

2.6.3. Phytochemical content

The testing of secondary metabolites, in the form of phytochemical content, was conducted on extracts of Tali Putri and Israeli grass, which were added to test solutions according to the test protocol. 5 mg of plant extract was placed in a test tube, then added with Aquadest and chloroform in a 1:1 ratio, up to a total volume of 5 ml. After that, it was shaken to form two layers of water and chloroform. The top layer is the water layer, and the bottom layer is the chloroform layer. The water layer was used to test flavonoids and phenolics, and the chloroform layer was used to test triterpenoids and steroids.

Flavonoid testing is performed by taking a 5 mL water layer using a dropper and placing it in a test tube. Then, add hydrochloric acid and Mg powder. Observe the color change; if orange or red forms, it indicates the presence of flavonoid compounds. (Nor *et al.*, 2023) .

Phenolic testing involves layering water, with up to 5 ml added to a test tube. Ferric chloride (FeCl3) is then added, and the color change is observed. If a green or blackish-blue color forms, it indicates the presence of phenolic compounds. (Nor *et al.*, 2023) .

Saponin testing was carried out using each host plant and Talli Putri extract. 10 mg of the sample was placed in a test tube, shaken for 1 minute, and then 10 ml of distilled water was added. If foam forms that persists for 30 seconds after adding concentrated HCl, this indicates the sample contains saponins. (Wijaya *et al.*, 2014) .

Triterpenoid and steroid testing was carried out. How to insert a layer of water into the hotplate hole. Wait until dry, then add concentrated sulfuric acid for the triterpenoid test. Observe the changes that occur if a red ring forms, indicating the presence of triterpenoid compounds. For steroids, acetic anhydride and concentrated sulfuric acid are added to the hotplate hole. Observe the color change that occurs if a green ring forms, indicating the presence of steroid compounds (Nor *et al.*, 2023).

Alkaloid testing was conducted using each method. For this, 40 mg of the host plant was extracted, and the Talli Putri was placed in a test tube. Subsequently, 2 mL of chloroform and 2 mL of ammonia were added. 3-5 drops of concentrated $H2SO_4$ were added to the filtrate and then shaken until two layers were formed. The acid fraction was taken, then 4-5 drops of Mayer and Dragendorf reagents were added to each. If a precipitate forms, it indicates that the sample contains alkaloids, with Mayer's reagent giving a white precipitate, and Dragendorf's reagent giving a yellow-red precipitate (Nor *et al.*, 2023).

2.6.4. Antioxidant activity test

Antioxidant activity was tested using the DPPH method. Antioxidant activity was tested on Tali Putri and Israel grass. Antioxidant activity was also tested on vitamin C as a positive control.

The initial step is to prepare a DPPH (2,2-diphenyl-1-picrylhydrazyl) solution, weighing 4 mg, and then transfer it into a 25 ml measuring flask. Dissolve the solution with 25 ml of methanol *to* obtain a concentration of 160 ppm.

Preparation of Test Solution, made by dissolving each host plant extract and Talli Putri, weighing as much as 10 mg, into a measuring flask, then dissolved with methanol solvent in a 10 ml measuring flask so that the concentration of the stock solution is 1000 mg / L (1000 ppm), then made variations in the concentration of the test solution. The test solution in each replication varies based on the absorbance obtained during the test and the percentage of inhibition, which reaches 50%.

Sample measurements were performed using various prepared concentrations. The absorbance of each test solution and positive control concentration was then measured at a wavelength of 516 nm. This step resulted in the percentage of inhibition at each test concentration. Determination of the IC50 value of antioxidant activity is calculated using the following formula:

$$\% \text{ inhibition: } \frac{A\ Kontrol - A\ sampel}{A\ Kontrol} \text{x } 100\%$$

Information:
% inhibition : Percentage of antioxidant activity power
A Control : Solvent absorbance + DPPH
A Sample : Solvent absorbance + DPPH sample

After obtaining the % inhibition value from the calculation, the IC50 (Inhibitory Concentration) value can be determined from each variation of the test solution concentration by using the regression equation that has been obtained. By plotting the abscissa (X-axis) against the % inhibition value (antioxidant) on the ordinate (Y-axis), a linear regression equation is created using Microsoft Excel, and the equation is obtained.

$$y = ax + b$$

Information :
y : Percentage of free radical scavenging activity
a : Intercept
x : Level (ppm)
b : Slope

The IC50 value describes the concentration of a test compound that can trap 50% of free radicals. The lower the IC50 value of a sample, the higher its free radical-scavenging ability, thus indicating a high antioxidant capacity.

3. Results and Discussion

3.1. Water content

Water content is measured to determine the amount of water in the herbal medicine. The purpose of measuring water content is to determine the quality of simple ingredients, such as whether they are susceptible to contamination by microbes that can cause physical damage

(Handayani et al., 2017). The average water content of Tali Putri and Israeli grass at three altitudes with seven replications for each area is shown in Table 1.

Table 1 Percentage of water content of the simplicia Tali Putri and Israel grass at different altitudes

Plains (asl)	Water content (%)	
	Tali putri	Israeli grass
R low(0-400)	9.96 ± 0.33 a	9.97 ± 0.67 a
Medium (400-700)	9.67 ± 0.58 a	9.75 ± 0.33 b
High (>700)	9.06 ± 0.32 b	8.82 ± 0.29 c

Note: Numbers in the columns followed by different lowercase letters indicate significant differences based on the DNMRT test at a 5% significance level.

The water content of the Tali Putri herb in the highlands showed a significant difference compared to the medium and lowlands. Meanwhile, the water content between the lowlands and the medium did not show a significant difference. The highest water content of Tali Putri was found in the lowlands, at 9.96%. The water content of Israeli grass in all areas showed a significant difference, with the highest water content found in the lowlands, at 9.97%. The standard water content of the herb is below 10%. (Ministry of Health, 2017) So that the simplicia Tali Putri and the grass of Israel meet these standards. This finding indicates that the simplicia Tali Putri and grass are of good quality, with a low risk of microorganisms that can damage their quality, and have a long shelf life. The lower the water content of the simplicia, the better the quality of the simplicia.

Talli Putri and Israeli grass that grow in the highlands have the lowest percentage of water content compared to the middle and lowlands. Plants grown in highlands have lower water content. Highland environmental conditions result in lower air pressure and relative humidity, as well as lower temperatures. Low air pressure lowers the boiling point of water, making it easier for water in plant tissues to evaporate. Furthermore, plants grown in highlands undergo physiological adaptations, such as reduced stomatal activity and decreased transpiration rate, which causes the water content in plant tissue to be lower. The combination of these factors causes simplicia originating from highlands to have a lower water content. Research (Safrina & Priyambodo, 2018) conducted at three altitudes and several drying methods stated that the higher the altitude, the lower the water content. Arnelio's research (2024) found that the water content of Tali Putri in the host grass israel was 8.25% and Adli's research (2014) found that the water content of Tali Putri and grass israel was 7-9% in different places, this is in accordance with research conducted where the water content obtained in simplicia Tali Putri and grass israel was 8.82% -9.97%.

3.2. Ash content

Content is a parameter that provides an overview of the internal and external mineral content originating from the initial process until the formation of the extract. The ash content of the simplicia Tali Putri and Israel grass was tested at three altitudes and seven replications, as shown in Table 2.

Table 2 Percentage of ash content of Tali Putri and Israel grass at different altitudes.

Plains (asl)	Ash content (%)	
	Tali putri	Israeli grass
Low (0-400)	8.55 ± 0.78 a	10.70 ±1.19 a
Medium (400-700)	10.12 ±2.15 a	9.83 ± 0.86 b
High (>700)	5.16 ± 0.79 b	7.07 ± 0.55 c

Note: Numbers in the columns followed by different lowercase letters indicate significant differences based on the DNMRT test at a 5% significance level.

The ash content of the Tali Putri herb in the highlands showed a significant difference compared to the medium and lowlands. Meanwhile, the medium and lowlands did not show a significant difference. The highest ash content of the Tali Putri herb was found in the mediumlands, namely 10.12%. Testing the ash content of Israeli grass revealed a significant difference across all areas, with the highest ash content found in the lowlands at 10.70%. The ash content of the Tali Putri herb met the standard, as the ash content of the simplicia is below 25% (Ministry of Health, 2017). The ash content of the Tali Putri herb and Israeli grass met the existing standards.

The purity of minerals in the simplicia is characterized by its low ash content, and vice versa, high ash content describes a low level of mineral purity in plant simplicia. Mineral purity affects the content of secondary metabolites and the antioxidant activity of plants (Saraf & Ajazuddin, 2010). The best ash content for Tali Putri and rumput israel is found in the highlands. Altitude affects the ash content of simplicia so that the higher a plain, the lower the ash content it has. This result is influenced by the climate and plant adaptation to the surrounding environmental conditions.

Antioxidant activity testing shows that the best antioxidant activity is also obtained at high altitudes. This finding indicates that the higher the altitude, the lower the ash content of the plant's simplex and the better the antioxidant activity obtained. Research by Arnelio (2024) found that the ash content of Tali Putri in Israeli grass was 7.51%, and research by Gautam et al. (2015) found that the ash content of Cuscuta reflexa was 4%. Research by Jose et al. (2018) found that the ash content of Tali Putri obtained in the host Alstonia scholaris was 7.96% and Ficus virens was 5.97%. The ash content of Israeli grass was 3% (Okolo et al., 2022). The ash content obtained in this study was almost the same as that of previous research, namely a

range of 3-7.96%.Phytochemical content

Phytochemical screening is a method for identifying secondary metabolite compounds in Tali Putri and Israeli grass. Identification of secondary metabolite compounds can be performed using color change reactions. Phytochemical content offers several benefits, including antioxidant, anticancer, detoxification, immune-boosting, and neuropharmacological properties (Kurmukov, 2013). The results of phytochemical tests of Tali Putri at different altitudes and seven replications at each plain are presented qualitatively in Table 3.

Table 3 Average results of phytochemical tests on Talli Putri at different altitudes

Phytochemicals	Low	Currently	Tall
Flavonoid	+	+	+
Phenolic	+	+	+
Saponin	+	+	+
Triterpenoid	+	+	+
Steroid	+	+	+
Alkaloid	-	-	-

Note : Contains compound (+), does not contain compound (-)

Altitude did not affect the content of flavonoids, phenolics, saponins, triterpenoids, and steroids. This result is because Tali Putri and Israeli grass already contain these phytochemicals, so altitude did not affect qualitative observations. This finding is in line with research conducted by Arnelio (2024), which found Tali Putri contained flavonoids, phenolics, saponins, triterpenoids, and steroids. Research by Saputri *et al.* (2024) also found flavonoids, phenolics, saponins, triterpenoids, and steroids in Israeli grass.

Phytochemical compounds contained in Tali Putri and Israeli grass have many health benefits such as acting as antioxidants, for the human body which can ward off various diseases such as cancer, hypertension and stroke, the mechanism of action is by suppressing the formation of ROS (*Reactive Oxygen Species*) which can be done by inhibiting enzymes or by binding trace elements related to the process of free radical formation, identifying the presence of ROS, and by increasing the regulation or protection of antioxidant defenses (Ballard & Maróstica, 2019).

Phytochemicals also offer numerous benefits to plants. The secondary metabolites they release are a form of self-defense for the plant. These secondary metabolites can protect against stress, such as flavonoids, terpenoids, and alkaloids, which are released during drought stress, and allelopathic compounds that act as antifungals, insecticides, anti-predators, antibacterials, and antivirals (Furtado et al., 2017).

3.3. Antioxidant activity

Antioxidant activity testing was carried out using the DPPH (2,2-diphenyl-1-picrylhydrazyl) method. The sample and DPPH were dissolved in methanol. The DPPH and sample were mixed, and then the test concentrations were varied accordingly. The test solution was analyzed using a UV-Vis spectrophotometer with a wavelength of 516 nm. The color change of the DPPH solution after being added to the dissolved Tali Putri extract is shown in the Figure.

Figure 2. Color change of the antioxidant activity test

Color changes occur when mixing the sample solution with DPPH in the antioxidant activity test. The color of the sample and DPPH ranges from blank to a concentration of 300 ppm, which initially changes from purple to yellowish. The color change is caused by electrons in DPPH pairing with electrons in the sample. Consequently, the inhibition value will increase as the sample concentration increases, due to the rising content of antioxidant compounds that can inhibit DPPH free radicals. (Molyneux, 2004) .

Table 4. IC50 values of Talli Putri and Israeli Grass.

Plains (asl)	Tali putri (µg/ml)	Israeli grass (µg/ml)
Low (0-400)	212.08 ± 77.04 b	348.96 ± 62.39 a
Medium (400-700)	316.30 ± 71.94 a	142.61 ± 48.71 b
High (>700)	173.35 ± 29.64 b	64.39 ± 14.71 c

Note: powerful category (0-50 µg/ml), strong (50-100 µg/ml), moderate (100-150 µg/ml), weak (150-200 µg/ml), and very weak (>200 µg/ml) (Yuniarti *et al*, 2020)

The antioxidant activity of Tali Putri, as indicated by IC50 values, did not exhibit significant differences between lowlands and highlands; however, it did show significant differences in medium plains. The highest IC50 value was found in Tali Putri grown in medium plains, namely 316.30 µg/ml. The antioxidant activity of Israeli grass in all plains showed significant differences, where the highest IC50 value was found in Israeli grass grown in lowlands, namely 348.96 µg/ml.

The antioxidant activity of the Talli Putri, which grows in both lowlands and highlands, does not show significant differences in statistical tests; however, it is categorized differently. The IC50 value in the highlands is categorized

as weak, while in the lowlands it is categorized as very weak. Princess Tali, grown in the highlands, exhibits better antioxidant activity when categorized accordingly. This result is due to the varying altitudes of the places, which in turn affect rainfall, air pressure, humidity, and temperature (Ohmura, 2012). Data from the West Sumatra BMKG Station from January to February can support that climate factors influence the antioxidant activity of plants, as seen in Table 5.

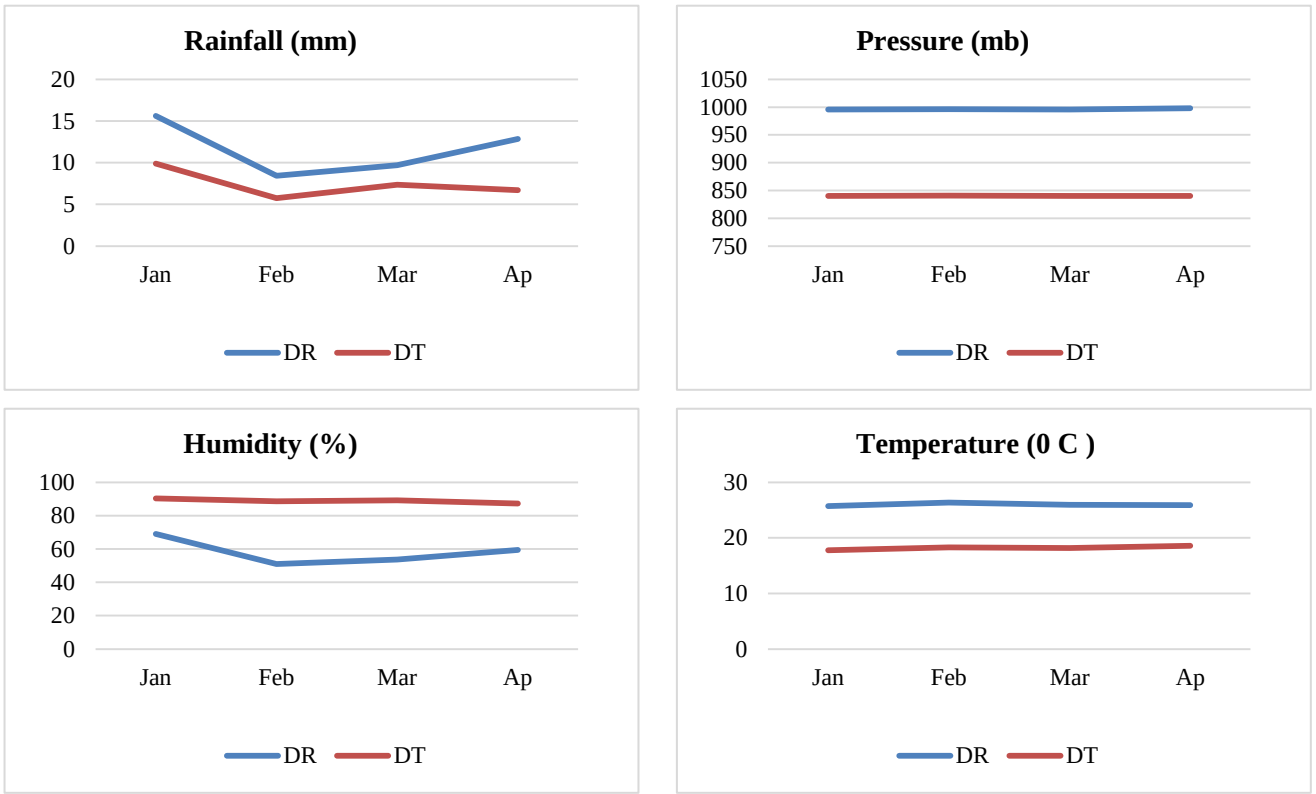

Figure 2. Climate Data of the West Sumatra BMKG Station. Type: Lowland (DR), Highland (DT)

Tali Putri (a type of vine) grown in the highlands of Nagari Alahan Panjang, at an altitude of >1,000 meters above sea level, has a better performance than Tali Putri (a type of vine) grown in other areas. This result is due to the lower air pressure in the highlands, which stimulates secondary metabolite content through ROS (Reactive Oxygen Species) in Tali Putri, thereby enhancing its antioxidant activity. This statement is supported by data from the West Sumatra Meteorology, Climatology, and Geophysics Agency (BMKG), which indicates that the air pressure during the Tali Putri growth period begins in March. In the long alahan area, the rainfall until April is 840.5 mm, which is lower than in the lowlands, at 99.3 mm.

BMKG West Sumatra data indicate that the average rainfall in March and April on N agari A long P land is 7.03 mm, which is 1.27 mm lower than in Nagari Sungai Sarik, which is 1 1.27 mm. Rainfall affects antioxidant activity in the highlands better than in the lowlands. Rainfall affects antioxidant activity because low rainfall causes drought stress in plants, stimulating the production of secondary metabolites that increase plant antioxidant activity. Likewise, the higher the rainfall, the lower the antioxidant activity. The rainfall in both places is classified as very low, but it makes a difference in the antioxidant activity of Tali Putri.

Differences in altitude also affect temperature. According to Meteorology, Climatology, and Geophysics Agency (BMKG) data from West Sumatra, the average temperatures in Alahan Panjang and Sungai Sarik are 18.4 °C and 2.5 °C, respectively. Notably, Tali Putri, which grows in the highlands, exhibits better antioxidant activity. Significant temperature differences between the two plateaus affect the production of secondary metabolites in Tali Putri and Israel grass. The production of secondary metabolites in plants influences antioxidant activity; secondary metabolites increase when temperatures are lower, thus increasing antioxidant activity (Alhaithloul *et al.*, 2021).

The humidity in March and April at Alahan Panjang National Park was 88.2%, and at Sungai Sarik National Park was 5.6%. Higher humidity lowers antioxidant activity, but coupled with low temperatures, antioxidant activity can be enhanced (Amin et al., 2022). This finding is because plants produce secondary metabolites when stressed as a defense against environmental stress. This study found that the highlands have high humidity and low temperatures, which stimulates better antioxidant activity.

Tali Putri grown in temperate latitudes has lower antioxidant activity and shows significant differences compared to highlands and lowlands. Climate data in temperate latitudes is not available, so climate factors cannot be used as a comparison for antioxidant activity. However, in the ash content test (Table 2), the Tali Putri simplex exhibits higher values in temperate latitudes, indicating a lower purity of the mineral content. The lower the ash content of a simplex, the purer the mineral content will be, which has an impact on increasing antioxidant activity (Nazirah *et al.,* 2023). Testing the ash content of Tali Putri simplex in temperate and lowland areas yielded the same results, categorizing it in the very weak category.

Antioxidant activity testing of Israeli grass extract was also conducted, revealing significant differences among the three plains, which were categorized differently. The best antioxidant activity was observed in the highlands, with a strong category, followed by the medium plains with a medium category, and the lowlands with a very weak category. This result is due to the higher air pressure in the highlands compared to the lowlands. According to BMKG data, the average air pressure in the highlands during the Israeli grass growing period (January and February) is 840.5 millibars, whereas the air pressure in the lowlands is 995.885 millibars. The antioxidant activity content of Israeli grass in the highlands is superior to that in the lowlands due to air pressure factors. Additionally, rainfall, temperature, and humidity also influence its antioxidant activity.

Rainfall in January and February in the highlands is 7.82 mm and in the lowlands 1 2.0 4 mm. This shows the highlands have lower rainfall than the lowlands, so Israeli grass growing in the highlands lacks water, which can stimulate self-protection to survive by increasing the production of secondary metabolites of the plant, thus increasing its antioxidant activity (Liu *et al.,* 2016). The antioxidant activity of Israeli grass in the highlands is also better than in the midlands and lowlands.

In addition, temperature and humidity also play an important role in the antioxidant activity of plants. In the highlands, when Israeli grass grows, the temperature is 18.05 ºC and the relative humidity is 89.55%. Low temperatures and high humidity can increase the antioxidant activity of plants (Chua *et al.,* 2015). Meanwhile, the temperature in the lowlands is higher than in the highlands, namely 26.02 ºC, as well as lower humidity compared to the highlands, namely 59.95%. The antioxidant activity in Israeli grass in the highlands is significantly higher than in the lowlands.

The antioxidant activity values of Tali Putri and Israeli grass at each altitude showed significant differences and were categorized differently. This result indicates that altitude influences the antioxidant activity of Tali Putri and Israeli grass due to differences in air pressure, temperature, rainfall, and humidity at each altitude. Furthermore, ash content also showed differences that affect the antioxidant activity of the plants.

The lowest standard deviations were found in the highlands for Tali Putri and Israel grass, indicating that the data in the highlands were more stable than in other areas. The standard deviations were still relatively high because the samples and replicates used were obtained from different growing locations, although they were still within the same area for each replicate.

This study also demonstrated that even though the host plant of the female vine has high antioxidant activity, it does not guarantee its effectiveness. Israeli grass in the highlands has potent antioxidant activity, while the female vine grown on Israeli grass has weak antioxidant activity. Similarly, at mid-altitudes, the antioxidant activity of Israeli grass is superior to that of the female vine. Only at low altitudes is the antioxidant activity of the female vine superior to that of its host plant.

Study Arnelio (2024) found that the average antioxidant activity value of Tali Putri in the lowlands with Israeli grass as the host was (256 µg/ml), rimbang host (163.70 µg/ml), horse whip host (279.50 µg/ml), and sambung rambat host (193.71 µg/ml). Tanruean *et al.'s* (2017) study obtained the IC50 value of Tali Putri on *the Coccinia grandis host.* (168.7 µg/ml), Ficus racemosa (201.6 µg/ml), Samanea saman (279.7 µg/ml). All of these studies fall into the categories of weak to very weak antioxidant activity, as well as studies conducted on three plants with Israeli grass hosts. The antioxidant activity value of Tali Putri also falls within the weak and very weak categories. Research by Utami *et al.* (2025) the antioxidant activity value of Israeli grass in all parts is 52.45 µg/ml, the stem part is 150.76 µg/ml, and the leaf part is 25.93 µg/ml, values that vary with the strong to weak category. This is also in line with the testing of the antioxidant activity of Israeli grass at different altitudes showing a strong to very weak category.

In this study, vitamin C was tested at concentrations of 2–12 ppm and used as a positive control alongside extracts of *Cuscuta australis* (Tali Putri) and *Asystasia gangetica* (Israeli grass) collected from different altitudes. The IC_{50} value of vitamin C was determined to be 7.621 µg/mL, classifying it as a powerful antioxidant. This result contrasts with the weaker antioxidant activity observed in both Tali Putri and Israeli grass extracts. The inclusion of vitamin C as a positive control served to verify the accuracy of the extraction and analytical procedures. Since the IC_{50} value of vitamin C fell within the expected range, this confirmed that the solvent preparation and pipetting process were carried out correctly (Molyneux, 2004).

4. Conclusion

Based on the discussion earlier, this study makes the following conclusions:

1. Effect of altitude on water and ash content
   Altitude significantly influenced the water and ash content of both Tali Putri and Israeli grass. The highest values were obtained in samples collected from the highlands, where Tali Putri recorded 9.06% moisture and 5.26% ash, while Israeli grass recorded 8.82% moisture and 7.07% ash.
2. Effect of altitude on phytochemical profile
   Altitude did not affect the qualitative phytochemical composition of either Tali Putri or Israeli grass. Both plants consistently contained flavonoids, phenolics, saponins, terpenoids, and steroids across all altitudinal zones.
3. Effect of altitude on antioxidant activity
   Antioxidant activity was influenced by altitude. The most vigorous activity in Tali Putri was observed in highland samples (IC_{50} = 173.36 µg/mL; weak category), whereas lowland samples exhibited very weak activity (IC_{50} = 212.08 µg/mL). In Israeli grass, the best antioxidant activity was also found in highland samples (IC_{50} = 64.40 µg/mL), which falls into the strong antioxidant category.

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References

Adli, A. S. (2014). Karakteristik ekstrak etanol tanaman rumput Israel (*Asystasia gangetica* L.) dari tiga tempat tumbuhan di Indonesia (Skripsi, Issue 1110102000031).

Alhaithloul, H. A. S., Galal, F. H., & Seufi, A. E. M. (2021). Effect of extreme temperature changes on phenolic, flavonoid contents and antioxidant activity of tomato seedlings (*Solanum lycopersicum* L.). *PeerJ*, *9*, e11193. https://doi.org/10.7717/peerj.11193

Amin, B., Atif, M. J., Meng, H., Ali, M., Li, S., Alharby, H. F., Majrashi, A., Hakeem, K. R., & Cheng, Z. (2022). Melatonin rescues photosynthesis and triggers antioxidant defense response in *Cucumis sativus* plants challenged by low temperature and high humidity. *Frontiers in Plant Science*, *13*, 855900. https://doi.org/10.3389/fpls.2022.855900

Arnelio, R. (2024). Skrining fitokimia dan uji aktivitas antioksidan ekstrak etanol tali putri (*Cuscuta australis* R. Br.) pada inang yang berbeda dengan metode DPPH (2,2-diphenyl-1-picrylhydrazyl) (Skripsi, p. 78).

Ballard, C. R., & Maróstica, M. R. (2019). Health benefits of flavonoids. In *Bioactive compounds: Health benefits and potential applications* (pp. 185–212). Elsevier. https://doi.org/10.1016/B978-0-12-814774-0.00010-4

Caldwell, G. W., Yan, Z., Lang, W., & Masucci, J. A. (2012). The IC50 concept revisited. *Current Topics in Medicinal Chemistry*, *12*(11), 1282–1290. https://doi.org/10.2174/156802612800672844

Chua, I. Y. P., King, P. J. H., Ong, K. H., Sarbini, S. R., & Yiu, P. H. (2015). Influence of light intensity and temperature on antioxidant activity in *Premna serratifolia* L. *Journal of Soil Science and Plant Nutrition*, *15*(3), 605–614. https://doi.org/10.4067/S0718-95162015005000027

Cyril, A., Esimone, C., Ugwueze, M. E., Anthony, A., & Kanneth, U. O. (2020). In vitro susceptibility of clinical isolates of *Candida albicans*. *World Journal of Pharmacy and Pharmaceutical Sciences*, *2*(3), 10–18. https://www.cabidigitallibrary.org/doi/full/10.5555/20133257820

Gautam, T., Gautam, S. P., Keservani, R. K., & Sharma, A. K. (2015). Phytochemical screening and wound healing potential of *Cuscuta reflexa*. *Journal of Chinese Pharmaceutical Sciences*, *24*(5), 292–302. https://doi.org/10.5246/jcps.2015.05.038

Gharibi, S., Tabatabaei, B. E. S., Saeidi, G., & Goli, S. A. H. (2016). Effect of drought stress on total phenolic, lipid peroxidation, and antioxidant activity of *Achillea* species. *Applied Biochemistry and Biotechnology*, *178*(4), 796–809. https://doi.org/10.1007/s12010-015-1909-3

Giri, I. M. D. S., Wardani, I. G. A. A. K., & Suena, N. M. D. S. (2021). Peran metabolit sekunder tumbuhan dalam pembentukan kolagen pada kulit tikus yang mengalami luka bakar. *USADHA: Jurnal Integrasi Obat Tradisional*, *1*(1), 23–29. https://usadha.unmas.ac.id

Handayani, S., Wirasutisna, K. R., & Insanu, M. (2017). Penapisan fitokimia dan karakterisasi simplisia daun jambu mawar. *Jurnal Farmasi UIN Alauddin Makassar*, *5*(3), 10–16. https://doi.org/10.24252/jfuinam.v5i3.4353

Hidayat, M. A., Fitri, A., & Kuswandi, B. (2017). Scanometry as microplate reader for high throughput method based on DPPH dry reagent for antioxidant assay. *Acta Pharmaceutica Sinica B*, *7*(3), 395–400. https://doi.org/10.1016/j.apsb.2017.02.001

Istiawan, N. D., & Kastono, D. (2019). Pengaruh ketinggian tempat tumbuh terhadap hasil dan kualitas minyak cengkih (*Syzygium aromaticum* L. Merr. & Perry) di Samigaluh, Kulon Progo. *Vegetalika Journal*, *8*(1), 27–41. https://doi.org/10.22146/veg.35744

Jose, A. G. R., Abirami, T., Kavitha, V., Sellakilli, R., & Karthikeyan, J. (2018). Green synthesis of silver nanoparticles using *Asystasia gangetica* leaf extract and its antibacterial activity. *Journal of Pharmacognosy and Phytochemistry*, *7*(1), 2453–2457.

Kokla, A., & Melnyk, C. W. (2018). Developing a thief: Haustoria formation in parasitic plants. *Developmental Biology*, *442*(1), 53–59. https://doi.org/10.1016/j.ydbio.2018.06.013

Kurmukov, A. G. (2013). Phytochemistry of medicinal plants. In *Medicinal plants of Central Asia: Uzbekistan and Kyrgyzstan* (pp. 13–14). Springer. https://doi.org/10.1007/978-1-4614-3912-7_4

Lestari, S., Septiyani, B. N., Proklamasiningsih, E., & Hernayanti, H. (2024). Kandungan flavonoid dan aktivitas antioksidan kitolod (*Hippobroma longiflora* L.) pada ketinggian tempat tumbuh berbeda. *LenteraBio: Berkala Ilmiah Biologi*, *13*(2), 212–218. https://doi.org/10.26740/lenterab8

Liu, W., Yin, D., Li, N., Hou, X., Wang, D., Li, D., & Liu, J. (2016). Influence of environmental factors on the active substance production and antioxidant activity in *Potentilla fruticosa* L. and its quality assessment. *Scientific Reports*, *6*, 28591. https://doi.org/10.1038/srep28591

Marrelli, M., Conforti, F., Araniti, F., & Statti, G. A. (2016). Effects of saponins on lipid metabolism: A review of potential health benefits in the treatment of obesity. *Molecules*, *21*(10), 1404. https://doi.org/10.3390/molecules21101404

Maslahah, N., & Nurhayati, H. (2024). Manfaat kesehatan beberapa senyawa fitokimia. *Warta BSIP Perkebunan*, *2*(3), 22–25.

Molyneux, P. (2004). The use of the stable free radical DPPH for estimating antioxidant activity. *Songklanakarin Journal of Science and Technology*, *26*(2), 211–219.

Muharni, S., Muflihah, F., Dasopang, E. S., Naswa, S., & Arianti, N. (2024). Uji aktivitas antihiperlipidemia ekstrak etanol daun ara sungsang (*Asystasia gangetica*) terhadap kadar kolestrol pada

mencit putih jantan (*Mus musculus*). *Indonesian Journal of Pharmaceutical and Clinical Research, 7*(2), 41–50. https://doi.org/10.32734/idjpcr.v7i2.18083

Nazirah, S. A., Rosmaina, S., & Rini, S. S. (2023). Flavonoid content and antioxidant activity of ethanolic extract of *Pometia pinnata* J. R. & G. Forst leaves. *Pharmacognosy Journal, 15*(1), 216–220. https://doi.org/10.5530/pj.2023.15.26

Nugraheni, N. K. (2020). Potensi senyawa flavonoid dalam menurunkan kadar glukosa darah. *Jurnal Ilmiah Kedokteran Wijaya Kusuma, 9*(1), 65–72. https://doi.org/10.30742/jikw.v9i1.754

Nurdin, N., Nurdin, A., & Iskandar, Y. (2023). Uji aktivitas antioksidan ekstrak etanol daun *Asystasia gangetica* (L.) T. Anderson dengan metode DPPH. *Jurnal Ilmiah Farmako Bahari, 14*(1), 25–32. https://doi.org/10.35316/jfb.v14i1.1831

Panche, A. N., Diwan, A. D., & Chandra, S. R. (2016). Flavonoids: An overview. *Journal of Nutritional Science, 5*, e47. https://doi.org/10.1017/jns.2016.41

Pietta, P. G. (2000). Flavonoids as antioxidants. *Journal of Natural Products, 63*(7), 1035–1042. https://doi.org/10.1021/np9904509

Prabowo, A. A., Kurniawan, A., & Fathurrahman, F. (2021). Uji aktivitas antioksidan ekstrak etanol daun tali putri (*Cuscuta australis* R. Br.) dengan metode DPPH. *Jurnal Fitofarmaka Indonesia, 8*(1), 1–7. https://doi.org/10.33096/jffi.v8i1.902

Pratiwi, R. (2018). Fitokimia: Senyawa flavonoid dan manfaatnya bagi kesehatan. *Jurnal Farmasi Sains dan Praktis, 4*(2), 25–32.

Putri, R. N. (2017). Efek antioksidan dari senyawa flavonoid dalam buah-buahan. *Jurnal Gizi dan Kesehatan, 9*(2), 101–109.

Rijai, L. (2018). Kandungan flavonoid dan aktivitas antioksidan ekstrak etanol daun *Asystasia gangetica*. *Pharmacon, 7*(3), 79–86. https://doi.org/10.35799/pha.7.2018.21082

Sari, N. M., Handayani, S., & Nurhayati, N. (2020). Penapisan fitokimia dan uji aktivitas antioksidan ekstrak etanol daun tali putri (*Cuscuta australis* R. Br.). *Jurnal Farmasi Galenika, 6*(1), 59–65. https://doi.org/10.22487/j24428744.2020.v6.i1.14297

Sari, P., & Wulandari, D. (2022). Uji aktivitas antioksidan ekstrak etanol daun *Pometia pinnata* dengan metode DPPH. *Jurnal Farmasi dan Kesehatan, 12*(1), 44–50. https://doi.org/10.33096/jfk.v12i1.2516

Septiana, R., Handayani, S., & Nurhayati, N. (2020). Uji aktivitas antioksidan ekstrak etanol daun tali putri (*Cuscuta australis* R. Br.) dengan metode DPPH. *Indonesian Journal of Pharmacy and Natural Product, 3*(2), 57–62. https://doi.org/10.33096/ijpnp.v3i2.671

Shahidi, F., & Ambigaipalan, P. (2015). Phenolics and polyphenolics in foods, beverages and spices: Antioxidant activity and health effects – A review. *Journal of Functional Foods, 18*, 820–897. https://doi.org/10.1016/j.jff.2015.06.018

Silalahi, J. (2006). Senyawa bioaktif dalam makanan: Suatu kajian kimia pangan. *Pustaka Pelajar*.

Srinivasan, R., Chandrasekar, M. J. N., Nanjan, M. J., & Suresh, B. (2007). Antioxidant activity of *Caesalpinia digyna* root. *Journal of Ethnopharmacology, 113*(2), 284–291. https://doi.org/10.1016/j.jep.2007.06.013

Subramanian, S., & Nandhini, M. (2014). Preliminary phytochemical screening and antioxidant activity of *Asystasia gangetica*. *International Journal of Pharmacy and Pharmaceutical Sciences, 6*(5), 539–543.

Wang, T., Li, Q., Bi, K., & Xu, L. (2018). Flavonoids and antioxidant activity of *Asystasia gangetica* leaves. *Natural Product Research, 32*(14), 1685–1689. https://doi.org/10.1080/14786419.2017.1350668

Wibowo, A., & Susilo, H. (2019). Uji aktivitas antioksidan ekstrak etanol daun tali putri (*Cuscuta australis* R. Br.) dengan metode DPPH. *Jurnal Farmasi Udayana, 8*(2), 134–142. https://doi.org/10.24843/jfu.2019.v08.i02.p09

Yanti, Y., & Mariani, R. (2021). Aktivitas antioksidan ekstrak etanol daun *Asystasia gangetica* (L.) T. Anderson dengan metode DPPH. *Jurnal Farmasi dan Kesehatan, 11*(1), 1–7. https://doi.org/10.33096/jfk.v11i1.1958

Yuliani, N., & Hartati, H. (2020). Uji aktivitas antioksidan ekstrak etanol daun tali putri (*Cuscuta australis* R. Br.) dengan metode DPPH. *Jurnal Fitofarmaka Indonesia, 6*(2), 45–51. https://doi.org/10.33096/jffi.v6i2.482

Yuniarti, E., Nurhayati, N., & Handayani, S. (2020). Uji aktivitas antioksidan ekstrak etanol daun tali putri (*Cuscuta australis* R. Br.) dengan metode DPPH. *Jurnal Farmasi Galenika, 6*(2), 105–111. https://doi.org/10.22487/j24428744.2020.v6.i2.15020