

# PHYTOCHEMICAL PROFILING AND ANTIOXIDANT PROPERTIES OF ETHANOL EXTRACT OF TAKOKAK EGGPLANT LEAVES (*Solanum torvum*)

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## Abstract

**Background:** Takokak eggplant (*Solanum torvum*) has been traditionally used in medicine, and its secondary metabolites possess antioxidant potential.

**Objective:** This study aimed to determine the phytochemical profiling and analyze the antioxidant properties of Takokak eggplant leaf extract (*Solanum torvum*) using ethanol solvent.

**Methods:** An experimental research design was used. Antioxidant activity was tested using the DPPH method.

**Results:** Phytochemical screening test of the 70% ethanol extract revealed the presence of flavonoids, terpenoids, saponins, and tannins, while the 96% ethanol extract of Takokak eggplant leaves (*Solanum torvum*) contained only flavonoids, terpenoids, and tannins. Antioxidant activity tests showed that the IC<sub>50</sub> (Inhibition Concentration) value of 70% ethanol extract was 49.9 ppm (very active), and 96% ethanol extract was 65.7 ppm (active). The AAI (Antioxidant Activity Index) value of the 70% ethanol extract was 0.80, while the 96% ethanol extract was 0.60.

**Conclusions:** Takokak eggplant leaf extract (*Solanum torvum*) with 70% ethanol had higher phytochemical content and antioxidant activity than 96% ethanol.

## Cite this Article

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## INTRODUCTION

Changes in modern lifestyles, including increased consumption of fast food, have contributed to elevated levels of free radicals in the body. The emergence of free radicals can also be caused by the body's natural processes, such as nutritional deficiencies, inflammatory processes, and normal cell metabolism. Damage caused by free radicals can trigger the emergence of degenerative diseases, such as cancer, atherosclerosis, diabetes, and high blood pressure (1). Antioxidants inhibit oxidation by reacting with free radicals to form stable radicals and are safe for human cells. Antioxidants neutralize free radicals in the body through natural metabolic processes. Over time, antioxidant production in the human body becomes ineffective (2).

The Takokak eggplant is a natural substance traditionally used as a medicine because of its antioxidant potential. The fruit of the Takokak eggplant can be cooked as a vegetable or consumed raw. The fruit and roots of Takokak eggplant contain secondary metabolite compounds that have traditionally been used as an antidote for stomach aches, heart disease, back pain, chronic coughs, hypertension, and scabies (3,4).

The selection of solvents is essential in the extraction process, and solvents must be selected according to their polarity and solubility to make it easier to separate active compounds in natural material samples. During the extraction process, the polarity level of the solvent used must be the same as that of the compound being identified, because differences in the type and concentration of solvents can affect the extraction process. Ethanol (EtOH) is a solvent that can extract compounds with polar properties (5). This explains why different ethanol concentrations change the solvent's polarity and affect bioactive compounds' solubility (6). This study provides further information on the effects of 70% and 96% EtOH solvents on the phytochemical profiling and antioxidant properties of Takokak eggplant leaf extract (*Solanum torvum*) as an alternative treatment for diseases caused by free radical factors.

## MATERIALS AND METHODS

This study used an experimental laboratory design with three repetitions. The sample was 4 kg of leaves obtained from Sayan Village, Ubud, Gianyar Regency, Bali. The study was conducted in the Basic Chemistry Laboratory and Applied Chemistry Laboratory of the Medical Laboratory Technology Department, Poltekkes Kemenkes Denpasar, between January and March 2024. The unit of analysis in this study was the *Takokak* eggplant leaf extract with extract solvent concentrations of 70% and 96% ethanol (EtOH). A UV-VIS Spectrophotometer was used for the antioxidant test using the DPPH method. The data were analyzed to obtain each extract sample's Antioxidant Activity Index (AAI) value.

The research stages conducted in this study involved the collection of 4 kg of *Takokak* eggplant leaves, sorting, washing, drying, powdering, and extraction with 70% and 96% EtOH solvents using the remaceration method for 7 days. The extraction process was carried out with two treatments: the first used 70% EtOH solvent, and the second used 96% EtOH

solvent, with the same ratio of powder amounts. The filtrate was evaporated using an evaporator until a thick extract was obtained.

The preparation of simplicia started with preparing a sample by weighing 10 g of thick extract of *Takokak* eggplant leaves, which was dissolved in 250 mL of EtOH. The EtOH concentration was adjusted according to the type of solvent used during the extraction process, followed by stirring and filtering. Filtrate from *Takokak* eggplant leaves, ready for qualitative phytochemical tests.

Qualitative phytochemical tests were performed to determine the Flavonoid, Alkaloid, Terpenoid, Tannin, and Saponin content.

- (a) The flavonoid test was carried out with 1 mL of the *Takokak* eggplant leaf extract sample, which was then heated in a bath. The filtrate obtained was added with 0.1 g Mg, then 0.4 ml of amyl alcohol and 4 ml of EtOH were added and shaken until homogeneous. Positive results were indicated by forming a yellow-orange precipitate (7).
- (b) Alkaloid tests were carried out on samples of *Takokak* eggplant leaf extract, and 3 mL of the extract was added to 5 drops of HCL. Test tube I was tested with two drops of Meyer's reagent, and test tube II was tested with two drops of Dragendorff's reagent. Positive tests for alkaloid compounds are characterized by orange deposits on Meyer's reagents and white deposits on Dragendorff's reagents (8).
- (c) For the terpenoid tests, 1 mL is added with 2 mL of chloroform, 10 drops of acetic anhydride, and three drops of concentrated sulfuric acid. A positive reaction to the presence of steroids is indicated by the formation of a red color, which then changes to blue-green (9).
- (d) Tannin tests were carried out by adding 1 mL of extract with 2-3 drops of 1% FeCl<sub>3</sub> solution and observing the color change. Blue color indicates the presence of three hydroxyl clusters in the tannin aromatic core. The green color indicates the presence of two hydroxyl clusters in the tannin aromatic core (10).
- (e) The saponin test was carried out by adding 1 mL of the sample to 10 mL of hot water, then shaking and allowing it to stand for a while. The formation of stable foam indicates the presence of saponins (8).

Antioxidant activity tests were conducted on 70% and 96% EtOH extract samples of *Takokak* eggplant leaves using the same treatment and procedure.

- (a) Preparation of DPPH solution.

Weigh 4 mg) was weighed and dissolved in methanol p.a and placed in a 10 mL measuring flask (DPPH 0.1 M). Then, 200  $\mu$ L of 0.1 M DPPH solution was pipetted into a 200 mL measuring flask, and methanol was added to the limit mark (11).

- (b) Measurement of the extract samples.

10 mg of thick *Takokak* eggplant leaf extract was weighed and dissolved in methanol p.a. in a 10 mL measuring flask. Stock solutions at each concentration were prepared by adding each solution to methanol to the limit mark (10 mL). A total of 2 mL of each solution was placed in a test tube, and 2 mL of DPPH solution was added. The solution was then incubated for 30 min in the dark. The absorbance of the sample was measured at the maximum wavelength with three repetitions at each concentration. The blank solution was calculated using the same procedure and treatment, and the sample solution was replaced with DPPH solution (12).

- (c) Determination of inhibition percentage and Inhibition Concentration (IC<sub>50</sub>) value. The Percentage of inhibition against DPPH radicals from each concentration can be calculated using the formula:

$$\% \text{ Inhibition} = \frac{\text{control absorbance} - \text{sample absorbance}}{\text{control absorbance}} \times 100\%$$

- (d) Determination of IC<sub>50</sub> and AAI values. The inhibition percentage and concentration were then plotted on the y and x axes in linear regression until the equation  $y = a + bx$  was obtained. This equation was used to determine the IC<sub>50</sub> value of each sample by substituting the “y” value of 50 and the “x” value obtained as the IC<sub>50</sub> value (11). The AAI value is obtained using the following formula:

$$\text{AAI} = \frac{\text{DPPH concentration}}{\text{IC}_{50} \text{ value}}$$

## RESULTS AND DISCUSSIONS

This study aimed to assess the phytochemical composition and antioxidant activity of *Takokak* eggplant leaf extracts. The results of the qualitative phytochemical tests of 70% and 96% EtOH extracts of *Takokak* eggplant leaves are as follows:

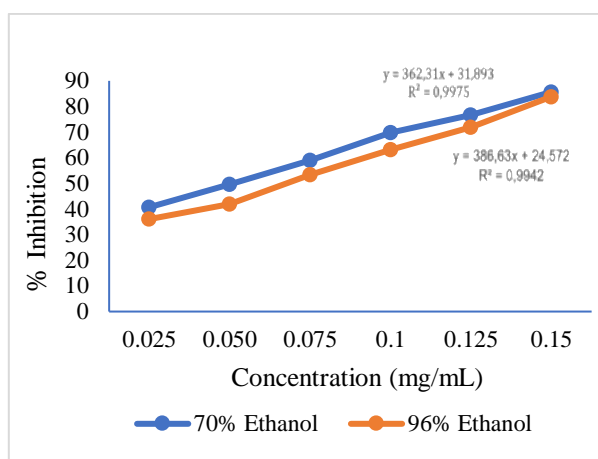
**Table 1.** Phytochemical Profiling of Extracted *Takokak* Eggplant Leaves

No	Compounds	70% EtOH Extracts	96% EtOH Extracts
1	Flavonoid	Detected	Detected
2	Alkaloid	Not detected	Not detected
3	Terpenoid	Detected	Detected
4	Tannin	Detected	Detected
5	Saponin	Detected	Not detected

Based on the qualitative phytochemical tests of the Takokak eggplant leaves extract, which have been carried out, it indicates that the 70% EtOH extract of Takokak eggplant leaves positively contains flavonoids, terpenoids, saponins, and tannins. The 96% EtOH extract positively contains flavonoids, terpenoids, and tannins only. This result is based on the results of previous studies using Takokak eggplant fruit, which contains flavonoid, saponin, and steroid compounds, and the root part contains alkaloid, flavonoid, triterpenoid, and tannin compounds (4).

Flavonoids are polyphenolic compounds known to have properties as free radical scavengers, inhibitors of hydrolytic and oxidative enzymes, and anti-inflammatory effects (13). Flavonoids, which are included in the category of phenol compounds, have many hydroxyl groups (-OH) and have a high electronegativity difference, making flavonoids polar. Therefore, flavonoids can be easily extracted using polar ethanol solvents because their hydroxyl groups allow the formation of hydrogen bonds (14). The study's results with two different solvent concentrations showed positive terpenoid compounds. This is because the organic fat compounds in the extract are well hydrolyzed in ethanol (15). The tannin test yielded positive results for both extracts. In addition to flavonoids, tannins are secondary metabolites widely found in plants. Tannins are generally used to treat various conditions, such as antibacterial skin diseases, diarrhea, hemostasis, and hemorrhoids (16). The phytochemical test results showed differences in the phytochemical content of the two extracts, with saponin content found only in the 70% EtOH extract. Saponin is a glycoside widely found in plants. It is complex and has foam-like characteristics. The saponin test is based on the principle that saponins contain sugar groups as polar components and steroid and triterpenoid groups as nonpolar components (17). Saponins have polar properties and are soluble in water and ethanol. The 70% EtOH extract had a higher polarity level because the water content was higher than that of the 96% EtOH extract. This causes the saponin content to be only shown in the 70% EtOH extract (18).

The antioxidant activity in both samples was determined by measuring the sample absorbance at a wavelength of 517 nm. The sample absorbance was measured in triplicate. Furthermore, the % inhibition of the sample was obtained from the absorbance of the sample and the blank.



**Figure 1.** Linear Regression Curve EtOH Extract of Takokak Eggplant Leaves

Based on the results of the linear regression equation (Figure 1), the IC<sub>50</sub> value of the 70% EtOH extract was 49.9 ppm, and the AAI value was 0.80 ppm (intermediate antioxidant category). In comparison, the IC<sub>50</sub> value of the 96% EtOH extract was 65.7 ppm, and the AAI value was 0.60 ppm (intermediate antioxidant category).

The antioxidant activity test showed that the 70% and 96% EtOH extracts of Takokak eggplant leaves were included in the intermediate antioxidant category. Still, 70% of the EtOH extract had a higher antioxidant activity value. These results are consistent with previous studies, where the 70% EtOH extract of Parijoto fruit has more potent antioxidant activity than the 96% EtOH extract (19). The higher antioxidant activity of the 70% EtOH extract compared to the 96% EtOH extract is related to the polarity of the solvent and the content of secondary metabolites extracted during the extraction process. The 70% EtOH solvent can attract compounds better because of the similarity in polarity with the compounds contained in the Takokak eggplant leaves. Differences in EtOH concentration can affect the solubility of bioactive compounds; the higher the ethanol concentration, the lower the polarity of the solvent. Different EtOH concentrations can alter the solvent's polarity, thereby affecting bioactive compounds' solubility (6).

The secondary metabolite content was higher in the 70% EtOH extract, which contains flavonoids, terpenoids, saponins, and tannins. Flavonoids, saponins, and tannins are known to have antioxidant activity. Flavonoids are a group of polyphenolic compounds known to possess potent antioxidant properties that ward off free radicals (13). Saponins have antioxidant effects because they can withstand superoxide by forming hyperoxide as an intermediary, thereby preventing damage to biomolecules by free radicals (20). The presence of saponin compounds in the 70% ethanol extract contributed to the high AAI value. This also shows that 70% EtOH is the optimal solvent for extracting antioxidant compounds.

## CLINICAL IMPLICATION

The clinical implication is as a reference for using traditional plants with antioxidant activity to reduce oxidative stress and prevent or alleviate various diseases.

## LIMITATIONS

This study has two significant limitations. First, phytochemical profiling was performed qualitatively on five secondary metabolite compounds. Although this approach can provide an initial indication of the phytochemical content of the sample, the results need further verification, especially using a quantitative approach. Second, antioxidant properties are focused on AAI values without additional testing, as phytochemical compounds provide significant activity as antioxidants.

## CONCLUSIONS

Takokak eggplant leaves extract (*Solanum torvum*) with 70% ethanol solvent has more phytochemical content (flavonoids, terpenoids, saponins, and tannins) and higher antioxidant activity than 96% ethanol solvent. The limitation of this study is that qualitative phytochemical testing was not carried out. Recommendations for further research are to conduct antioxidant properties tests using different extraction solvents.

## CONFLICT OF INTEREST

The author(s) declared no potential conflicts of interest concerning this article's research, authorship, and/or publication.

## AUTOR CONTRIBUTIONS

Ni Wayan Mita Efianti prepared the materials and conducted the tests. Gusti Ayu Made Ratih designed the study, calculated the IC<sub>50</sub> and AAI data, and prepared the manuscript. Ni Nyoman Astika Dewi provided laboratory equipment and supervised the phytochemical profiling and antioxidant properties tests.

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