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Pharmacognostic and Phytochemical Characterization of *Colocasia esculenta* var. antiquorum Corm (Araceae) and Preliminary Evaluation of Its Antimicrobial Activity

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ABSTRACT: *Colocasia esculenta* has been reported in reputable journals to exhibit various bioactivities, including antioxidant, anti-inflammatory, and antidiabetic properties. This study aimed to investigate the pharmacognostic characteristics and phytochemical profile of *Colocasia esculenta* var. antiquorum corm (Araceae). The corms were collected, authenticated, oven-dried, and subjected to preliminary pharmacognostic and phytochemical analyses. Pharmacognostic evaluations included organoleptic, microscopic, and physicochemical examinations. The dried corm powder was extracted with distilled water, followed by qualitative phytochemical screening. The extract was found to contain saponins, flavonoids, tannins, and triterpenoids. Antimicrobial activity was further evaluated against *Staphylococcus aureus*, *Escherichia coli*, and *Candida albicans*, which were selected as representative pathogenic microorganisms for Gram-positive bacteria, Gram-negative bacteria, and fungi, respectively. These microorganisms were chosen based on previous reports demonstrating that the ethanolic leaf extract of taro exhibited antifungal activity against *Candida albicans* and antibacterial activity against *Pseudomonas aeruginosa*, *Staphylococcus aureus*, *Klebsiella*, and *Escherichia coli*. However, no inhibitory effect was observed in this study. All analyses were performed in accordance with the guidelines of the Indonesian Herbal Pharmacopeia. The findings provide reference data for the pharmacognostic identification of *Colocasia esculenta* corm and highlight its phytochemical constituents. Further investigations on other biological activities are required to validate its therapeutic potential.

KEYWORDS: *Colocasia esculenta*; Taro Corm; Aqueous Extract; Pharmacognostic Analysis; Phytochemical Screening.

1. INTRODUCTION

Tuber plants are an important staple food source for carbohydrates. They are generally processed in various forms before consumption. *Colocasia esculenta*. var. antiquorum (taro) belongs to the Araceae family (aroid family) and is a tuber plant that is also used in traditional medicine [1]. This plant originates from humid tropical rainforest regions of Southeast Asia, including Indonesia. Taro is a monocot herbaceous plant that grows to a height of 1-2 m. It has green or purple heart-shaped leaves along with long petioles and fibrous roots as nutrient storage (corm). As a root vegetable, taro is grown primarily for its edible corms [2,3]. The ethanolic leaf extract of taro showed antifungal activity against *C. albicans*, and antibacterial activity against *P. aeruginosa*, *S. aureus*, *Klebsiella*, and *E. coli* [3,4]. According to several studies, there are bioactive components in taro with antioxidant, anticancer, antimetastatic, wound healing, antihyperlipidemic, anti-inflammatory, antimelanogenic, probiotic, antidiabetic, antihypertensive, and antimicrobial properties to control plant disease. Taro leaves contain phytochemical compounds such as anthraquinones, vitexin and isovitexin, catechins, apigenin, cinnamic acid derivatives, saponins, flavonoids, tannins, alkaloids, terpenoids, and

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steroid hormones participating in the healing process. Taro fruit has potential as an anticolytic agent against TNBS-induced ulcerative colitis, which is related to its antioxidant and anti-inflammatory potential. Nutritionally, taro corm contains anthocyanin, cyanide 3-glucoside, 11% protein, 85-87% starch, and other nutrients such as minerals, Vitamin C, thiamin, riboflavin, and niacin, which are better than other cereals. Taro corm also contains more protein than other tubers [4–11].

In Indonesia, there are several herbal products called jamu, which contain taro tubers/corms in both single and polyherbal forms. Jamu is an Indonesian traditional herbal medicine indicated by empirical evidence [12]. These herbal products can help relieve joint pain and improve urination. In recent decades, medicinal plants have been the subject of pharmacognostic and phytochemical studies. This will help us to identify the right plants for further study and development to standardize the manufacturing of herbal products. The origin of the plant material, the method of extraction, and the method of drying were adjusted to the procedures normally carried out in herbal medicine factories. Therefore, in this study, we investigated the pharmacognostic characteristics and phytochemical profile of *Colocasia esculenta* var. antiquorum corm (Araceae).

2. MATERIALS AND METHODS

2.1. Materials

C. esculenta corm was collected from Purwakarta, West Java, Indonesia. These corms were harvested when they were four months old. The plants are sent for authentication to authorized institutions.

2.2. Methods

2.2.1. Macroscopical Evaluation

Fresh corms from the base of the main stem were peeled with a knife. Observations and measurements of the habit and plant parts were performed using an eye lens, thread, and ruler. Photographs of the habit and plant parts were then captured using a digital camera. The characters were then interpreted.

2.2.2. Microscopical Evaluation

Taro corms were subjected to microscopic evaluation by obtaining transverse sections using standard procedures and then subjecting them to microscopic examination [13]. The specimen was observed under an Olympus light microscope at magnifications of 10X and 40X. They were studied, and photomicrographs were taken with a digital camera (Sony DSC-W230, China).

2.2.3. Extraction and Phytochemical Screening

Taro corms were dried in an oven at a temperature of 35-45 ^{0}C and ground to powder with a mesh of 60. Taro corm was extracted using aquadest as a solvent at a temperature of 40-50 ^{0}C. The aqueous extract was filtered and then concentrated at a temperature of 45-50 ^{0}C to obtain the viscous extract. The viscous extract was dried in an oven at a temperature–30-45 ^{0}C and then ground to powder using an 80-100 mesh to obtain the dry extract. Phytochemical screening was performed to determine the secondary metabolites of taro, such as alkaloids, flavonoids, phenols, tannins, saponins, quinones, and terpenoids/steroids, using standard procedures [14–16].

2.2.4. Heavy Metal Contamination Test

Pb contamination in the extract was measured using the method described previously [17] with an Atomic Absorption Spectrophotometer (AAS) AA7000 Shimadzu.

Preparation of Pb Standard Solution

1 mL of 1000 ppm Pb standard solution was taken into a 10 mL volumetric flask, diluted to volume with 0.125 N HCl to obtain a Pb standard solution in a concentration of 100 ppm. 0.2 mL; 0.4mL; 0.6mL; 0.8mL; 1.0 mL of Pb standard solution were taken into a 10 mL volumetric flask, and the volume of a volumetric disk with 0.125 N HCl was added to obtain concentrations of 2.0, 4.0, 6.0, 8.0, and 10.0 ppm, respectively.

Preparation of test solution

2-3 g of the sample were dried in a porcelain cup by heating until no more smoke was formed. Do ashing in the furnace at a temperature of 500 ^{0}C until the ashes turn white. If the ash is still gray (not carbon-free), add 0.5 mL - 3 mL HNO_3 (P), and continue ashing until the ashes turn white. Add 10 mL of HNO_3 6 N, and heat for 30 min (until dry). 10 mL of 0.1 N HNO_3 was added to a 25 mL volumetric flask.

A blank solution was prepared using the same treatment as the sample. The standard solution, test solution, and blank solution with AAS were measured. The metal content of the sample was calculated using the following formula:

$$\text{Metal content } (\mu g/mL) = C \times VW$$

Where:

C = Sample concentration from the calibration curve (µg/mL)

V = Final solution volume (mL)

W = Sample weight (g)

2.2.5. Microbial contamination test

The microbial contamination test determined the Total Plate Count and the Yeast and Mold Count. Ten grams of extract was added to 90 mL of Tryptic Soy Broth (TSB) medium in an Erlenmeyer flask to obtain a concentration of 10^{-1}. One milliliter of the 10^{-1} solution was placed into a test tube containing 9 mL of TSB (10^{-2}) and diluted to a concentration of 10^{-6}. One milliliter of each solution (10^{-1} - 10^{-6}) was diluted into petri dishes for each dilution. Total yeast and mold count using SDA (Sabouraud Dextrose Agar) as medium and incubated at a temperature of 2025°CC, incubated for 5-7 days (Observe on the 3rd day, if no colonies have grown, wait until the 5th day). Total plate count using Tryptic Soy Agar (TSA) as the medium and incubation at a temperature of 3035°CC for 3-5 days. Observe on the 5th day. If no colonies had grown, wait until the 7th day. The number of colonies was counted in each plate, with the highest number of colonies being less than 250. The number of colonies was counted in each plate, with the highest number of colonies being less than 50. The number of yeasts in each 1 g of the sample was calculated by multiplying the counted colony average by the dilution factor.

2.2.6. Antimicrobial activity test

The antimicrobial test was adapted from a previously described method 18–21 with modifications. Prepared microbial cultures (E. coli, S. aureus, and Candida albicans) were aged 24 h for bacteria and 48 h for fungi. Take 1 ose input in diluent (sterile TSB) and then homogenize. Measured %T by UV-Vis spectrophotometer at a wavelength of 580 nm to 25%T (108 CFU/mL for bacteria and 106 CFU/mL for fungi). Pour 20-25 mL of TSA media for bacteria and SDA for fungi into a petri dish and let it solidify. 0.2 mL of the microbial suspension was placed on the surface of solid TSA/SDA media. Spread the microbes over the entire surface of the media with a sterile swab and then wait until the agar is solid. The sample, positive control, and negative control were placed on the surface of the media in a paper disc with a diameter of 6 mm. Wait for 1 h, and let the test solution absorb. Aerobic incubation at a temperature of 30-35°C for 18-24 hours and bacteria at a temperature–20-25°C for 24-48 hours. A clear zone/inhibition zone was observed.

3. RESULTS

3.1. Authentication

The plant was authenticated by Mr. Heri Santoso S.Si., the head of the executive branch of the Indonesian Biology Generation Foundation, with Ref No 08.128/Genbinesia/IX/2022.

Classification	
Regnum	*Plantae*
Divisi	*Magnoliophyta*
Class	*Liliopsida*

Ordo	*Alismatales*
Family	*Araceae*
Genus	*Colocasia*
Species	*Colocasia esculenta*. var. *antiquorum.*

3.2. Description

It is a seasonal herbaceous plant. Rhizome branching, tube-to-circular, scaly, blackish-brown. Single leaf, light green, drooping root, petiole 14–45 cm long; shield leaf, heart; flat edge; lobe base; blunt end; soft texture.

Macroscopic evaluation is an important initial step in the identification of plant material. C. esculenta was evaluated for its basic botanical and organoleptic characteristics as shown in Figure 1. The corms have brown skin with a size ranging between 3-6 cm. In the horizontal section, it had a white inner part that contained amylum.

Figure 1. *Colocasia esculenta (L.) Schott* corms

3.3. Microscopical evaluation

The horizontal section of the taro corms shows several vascular bundles (A) (Figure 2). C, shows air vesicles. In the outermost layer, there is the cuticle, followed by the upper epidermis, and then the collenchyma. There was a long palisade layer. Epidermal cells on the upper side look tightly closed compared with those on the lower epidermis. The spongy parenchyma contained vacuoles. The microscopic features of *C. esculenta* are presented in Figure 2.

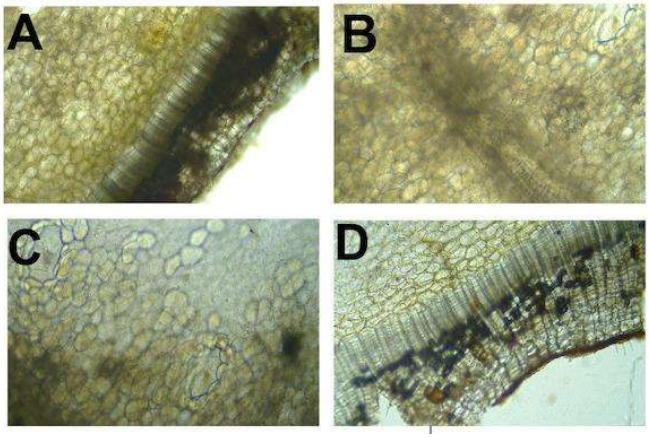

Figure 2. Microscopic characteristics of *C. esculenta.*

3.4. Phytochemical screening

Based on the results of the phytochemical screening test, C. *esculenta* corm contained flavonoids, saponins, tannins, triterpenoids, and quinones (Table 1).

Table 1. Phytochemical screening of *C. esculenta* corm

Group	Aqueous Extract of *C. esculenta* corm	References [2,10,22]
Flavonoid	+	+
Saponin	+	+
Alkaloid	-	+
Tannin	+	+
Triterpenoid	+	+
Quinone	+	+

3.5. Heavy metal and aflatoxin contamination test

In this study, heavy metal contamination was measured using AAS with a regression equation of (y) = 0.0157x - 0.0024, with a correlation coefficient (R2) of 0.9996. The results (Table 2) showed that the taro corm samples met the requirements for contamination, namely, they did not contain Pb and aflatoxin.

Table 2. Analysis of aflatoxin and heavy metals

Analysis	Result	Requirements
Total aflatoxin	none	≤ 20 µg/kg
Metal contamination: Pb	none	≤ 10 mg/kg

3.6. Microbial contamination test

The total plate count and yeast and mold counts were determined using serial dilutions (10^{-1} – 10^{-7}). The purpose of dilution is to obtain several colonies that do not accumulate in each petri dish, so that it is not difficult to calculate the result. The number of colonies was counted at the lowest dilution [23].

Table 3. TPC and TYMC of C.esculenta corm

Analysis	Result	Requirements
Total Plate Count (TPC)	8,3 x 10^4 CFU/g	≤ 10^5 CFU/g
Total Yeast and Mold Count (TYMC)	1 x 10^1 CFU/g	≤ 10^3 CFU/g

3.7. Antimicrobial activity

The antimicrobial activity of taro corm was determined using the diffusion method. The antimicrobial activity of the aqueous extract was determined by measuring the zone of inhibition after incubation. The results are presented in Table 4.

Table 4. Antimicrobial activity

Concentration (%)	*E. coli* (mm)			*S. aureus* (mm)			*Candida albicans* (mm)		
	1	2	3	1	2	3	1	2	3
100	6	6	6	6	6	6	6	6	6
75	6	6	6	6	6	6	6	6	6
50	6	6	6	6	6	6	6	6	6

25	6	6	6	6	6	6	6	6	6
12,5	6	6	6	6	6	6	6	6	6
5	6	6	6	6	6	6	6	6	6
2,5	6	6	6	6	6	6	6	6	6
1,25	6	6	6	6	6	6	6	6	6
0,625	6	6	6	6	6	6	6	6	6
K- (Aqueous sterile)	6	6	6	6	6	6	6	6	6
K+ (Chloramphenicol)	23.1	23.3	23.4						
K+ (tetracycline)				35.1	35.5	35.9			
K+ (Nystatin)							25	25.5	25
Antibiotic Disk				6					

Note: K+ (positive control), K- (negative control)

The results showed that the sample had no antimicrobial activity. This can be observed in the inhibition zone (Table 4). The results of the sample and positive control were different; the positive control had antimicrobial activity with an inhibition zone of 23-25 mm.

4. DISCUSSION

Pharmacognostic investigation is a fundamental step in the standardization and quality assurance of medicinal plants. It provides essential diagnostic parameters, such as organoleptic, microscopic, and physicochemical characteristics, that ensure the accurate identification and authentication of plant materials. Such analyses are crucial for distinguishing genuine plant sources from possible adulterants or substitutes that may compromise safety and efficacy. Furthermore, pharmacognostic data form the basis for the establishment of monographs in pharmacopeias and for developing reproducible formulations for both traditional and modern herbal medicines. Without proper pharmacognostic evaluation, the reliability of subsequent pharmacological or toxicological assessments is significantly undermined.

The corm is a tuber, which is a modified stem with a surface covered with leaf scars, forming a whorl-like ring around the tuber. Lateral shoots then develop into secondary cormels or corms (Figure 1). The outer side of the tuber was covered by a thick periderm consisting mostly of radially arranged phlem (cork cells) and compact radial rows (Figure 2A). The three regions of the felem can be distinguished as follows: an inner region of 10-15 cell layers, a middle region of 5-10 cell layers, and an outer region of 2-3 cell layers, which is brown in color. The fellogen is one to two cells wide, and there appears to be no clear demarcation between the phelloderm and the underlying outer cortex. This observation was similar to that of other studies [26].

Phytochemical profiling, on the other hand, offers valuable insights into the chemical constituents responsible for a plant's biological activities. The identification of secondary metabolites, such as alkaloids, flavonoids, tannins, saponins, and terpenoids, provides a scientific foundation for understanding potential therapeutic effects and mechanisms of action. Variations in phytochemical composition may occur because of factors such as plant part, extraction solvent, geographical origin, and environmental conditions, making systematic phytochemical screening an indispensable component of plant-based research. Comprehensive phytochemical characterization not only supports the rational use of herbal materials but also guides further bioactivity-directed studies for the discovery of novel lead compounds.

Phytochemical screening was carried out to determine the content of secondary metabolites in the taro corm dry extract (Table 1). Compared with other research reports, it shows that there are differences in the results, namely that no alkaloids were detected. This may be due to differences in growing locations, which cause low levels of alkaloids that cannot be detected by this phytochemical screening method [2,10,22].

Traditional medicine in Indonesia must meet quality and safety requirements according to the regulations of the Drug and Food Control in Indonesia: Farmakope Herbal Indonesia. Traditional medicine is safe when

it meets the specified requirements of microbial, aflatoxin, and metal contamination. The results of the analysis and requirements are presented in Tables 2 and 3. High heavy metal content in plants can cause poisoning and plant death. Heavy metals can contaminate plants as a result of human activity. Heavy metals are present in the air and contaminated soil. Pb is one of the most dangerous heavy metals. Pb can enter the body through the digestive tract (from food, such as plants contaminated with Pb) and the respiratory tract (through polluted air). Pb that accumulates in the blood can cause damage to organs and body systems such as the kidneys, liver, reproductive system, nervous system, urinary system, immune system, basic cell physiological processes, and gene expression [24].

Aflatoxin is a mycotoxin produced by fungi as a secondary metabolite. Plants can be contaminated by aflatoxin during harvesting and storage. Mycotoxins are produced by some fungi, such as *Aspergillus flavus* and *Aspergillus parasiticus*. Aflatoxin contamination can be a threat to plants, especially those used as food [25].

In the antimicrobial test, the taro corm water extract sample showed no activity, whereas the results of other studies showed that the methanol extract of *C. esculenta* corm provided antimicrobial activity with an inhibition zone of 22 mm [6,10]. The different results were caused by differences in the solvents used for extraction, thus causing differences in secondary metabolite content.

Future research should focus on comprehensive phytochemical characterization using advanced analytical techniques such as HPLC, LC–MS/MS, or GC–MS to quantify and identify active constituents. Comparative studies employing different solvents, extraction methods, and plant parts are necessary to optimize the recovery of bioactive compounds. Bioassay-guided fractionation would further aid in isolating compounds responsible for specific biological activities. Additionally, evaluating antioxidant, anti-inflammatory, cytoprotective, and antidiabetic potentials, supported by in vitro and in vivo models, could expand the understanding of this plant's therapeutic relevance. Integrating metabolomic profiling with molecular docking or network pharmacology approaches may also provide mechanistic insights and facilitate the development of *C. esculenta* var. antiquorum as a scientifically validated herbal candidate.

5. CONCLUSION

This study demonstrated the presence of macroscopic and microscopic structures. Microscopic structures, such as the epidermis, modified parenchyma, and sclerenchyma, were identified. In the phytochemical study, various phytoconstituents, such as saponins, flavonoids, tannins, and triterpenoids, were present in the aqueous extract of taro corm. From our library search and research, we conclude that this plant has various phytochemical-containing properties and may be useful for treating various diseases. Further studies are needed to determine the exact mechanism of the plant against various diseases.

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