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# Profile of Characteristic and Secondary Metabolite Content Alstonia Scholaris Medicinal Plants

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ABSTRACT

Pulai plant (Alstonia scholaris) has been known as a medicinal plant that is used by the community. Utilization in the field by the community usually uses boiling or brewing from the leaves and bark. The purpose of this study was to determine the characteristics and secondary metabolite content of steeping and boiling pulai plants. The method used is brewing and infusion from the leaves and bark of the pulai. The results of the research for the characteristics of this plant that met the MMI criteria were water content and total ash content, while secondary metabolites contained alkaloids, flavonoids, steroids, and triterpenoids.



# Profil Karakteristik dan Kandungan Metabolit Sekunder Tanaman Obat Alstonia Sholaris

*Kata kunci:*

Alstonia scholaris
Karakteristik
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ABSTRAK

Tanaman pulai (*Alstonia scholaris*) memiliki dikenal sebagai tanaman obat yang dimanfaatkan oleh masyarakat. Pemanfaatan di lapangan oleh masyarakat biasanya menggunakan perebusan atau penyeduhan dari daun dan kulit batang. Tujuan dari penelitian ini adalah untuk mengetahui karaetistik dan kandungan metabolit sekunder dari seduhan dan rebusan tanaman pulai. Metode yang digunakan adalah dengan cara menyeduh dan infusa dari daun dan kulit batang pulai. Hasil penelitian untuk karakteristik tanaman ini yang memenuhi kriteria MMI yaitu kadar air dan kadar abu total sedangkan senyawa metabolit sekunder mengandung senyawa alkaloid, flavonoid, steroid, dan triterpenoid.



## INTRODUCTION

Herbal plants are plants that are usually used for traditional medicine (Suparmi, 2010) or better known as traditional medicine (Ministry of Health, 2017). Traditional medicine itself is an ingredient or ingredient derived from plants, animals, minerals, preparations (galenic) or mixtures that have been used for generations in medicine (Mandang, 2004) Empirically, people use traditional medicine by brewing, boiling, pounding, kneading. , smeared, heated or affixed and eaten directly (Farida, 2018). One of the herbal plants used for medicine is the Pulai plant (Alstonia scholaris). This plant, which is the Apocynaceae family, has good sap on the leaves, stems and flowers and is white on

the stems (Mandang, 2004). This plant is one of the threatened species with a rare status (Mandang, 2004). The benefits of the Pulai plant (Alstonia scholaris) can be used to treat various diseases. The leaves are usually used to treat beriberi, liver, analgesics, wound healing, antiasma, antimutagenic, hepatoprotective and gastric. The bark is used for acute diarrhea, dysentery, antiparasitic (worms), and anti-fertility (Meena A.K, 2011). At the root it is used as an anticancer and antimicrobial. In previous research, it was also known that this plant contains secondary metabolites in the form of alkaloids (Bhanu, 2013), flavonoids (Meena A.K, 2011), polyphenols, terpenoids (Elsye, 2016) and has antioxidant activity (Zuraida, 2015) from its thick extract. There are no studies that directly use dry extracts to test the content of secondary metabolites and antioxidants.

The use of this medicinal plant in the field, most people process it by boiling or brewing the bark or leaves and then filtering it and drinking it (Farida, 2018) without knowing which chemical compounds and antioxidants are better. So that the purpose of this study is to determine the effective method for empirical processing in the community by comparing the content of compounds in boiled and brewed preparations. Benefits in the near term after knowing the results of secondary metabolites/chemicals and antioxidants, people can choose the right method so that the properties of this medicinal plant can work properly. The importance of this research is so that people get the right information about the use of herbal medicine.

## METHOD

### Time and Place of Research

This research was started from January - August 2020. The place for this research was carried out at the Laboratory of Chemistry of Natural Materials, Mathematics and Natural Sciences, Pharmacy, University of Garut.

### Materials and tools

The materials and tools needed for this research are test tubes, beaker, erlenmeyer, tube rack, micro pipette, drop pipette, volumetric flask, crucible, glass funnel, spatula, pliers, mortar, stamper, stirring rod, free filter paper. ash, electric stoves, aluminum foil, analytical scales, reagent sprayers, furnaces, desiccators, refrigerators, ovens, steam dishes, rotary evaporators, KLT GF245 silica gel plate, capillary tubes, 365 nm UV lamps, glass vials, and UV-spectrophotometer Vis, aquadest

### Research procedure

*Herbal tea drying & making*

Drying is done by washing the bark thoroughly with running water. Furthermore, the logs are aerated after a bit dry and put in a drying cabinet (Depkes, 1985).

*Characterization*

Characterization to determine the quality of simplicia to be tested, namely water content, total ash content, acid insoluble ash content, water insoluble ash content, total extract content, acid insoluble extract content, water insoluble extract content, and drying loss.

*Organoleptic & microscopic tests*

The organoleptic test that will be carried out is by testing the smell, taste, shape and size. Microscopic test yourself by taking a little herbal tea powder then dropping anhydrous chlorine and distilled water. Next observe under a microscope.

*Brewing*

Brewing is done by pouring boiling water into herbal tea/simplicia, let stand for 15 minutes then filtered.

*Infussion*

Boiling is done by infusion, which is boiling a portion of the herbal tea for 15 minutes at 90 ° C. Made by using 10% of the total simplicia.

*Alkaloid Testing*

A total of 2 grams of simplicia powder moistened with 5 ml of 30% ammonia added 20 ml of chloroform. From this mixture, 10 ml of filtrate (A) was extracted with HCL (1:10) then shaken and the top part was taken. Solution A was dropped a few drops on filter paper and sprayed or dropped with Dragendorff reagent. Solution B was added by Dragendorff and Mayer reagent. The presence of brick red sediment (dragendroff reagent) and white precipitate (Mayer reagent) indicates the presence of alkaloid compounds.

*Flavonoid Testing*

Two grams of simplicia are boiled in 100 ml of water for 5 minutes, then 5 ml of the filtrate is taken. The filtrate is added with mg powder and a solution of alcohol: HCl (1: 1) and 5 ml of amyl alcohol. then shaken vigorously and allowed to separate. The formation of a yellow, red or orange color on the amyl alcohol layer indicates the presence of flavonoid compounds.

*Saponin Testing*

Two grams of simplicia powder are boiled in 100 ml of water for 5 minutes then the filtrate is taken. From the filtrate, 1m ml was taken and shaken for 1m minutes. To form, add 100 ml of hot water and boil for 10 seconds, let it stand for 10 minutes, then add 1% HCL.

*Tannin & Quinone Testing*

Two grams of simplicia powder are boiled with 100 ml of water for 5 minutes then the filtrate is taken and divided by 3. The first filtrate is added with 1% FeCl while the second is given *Steasny* reagent and heated in a heat, filtered and saturated with sodium acetate.

The formation of a dark blue or blackish green color indicates the presence of a tannin group compound in the first filtrate. For the second filtrate, if a pink precipitate is formed, it indicates the presence of catekat tannins. Then the precipitate was filtered, the filtrate was saturated with sodium acetate, and a few drops of 1% FeCI3 solution were added. The formation of an ink blue color indicates the presence of an error tannin. 5 ml of the test solution (Filtrate 3) is added a few drops of NaOH solution. If a red color is formed, it indicates the presence of quinones.

*Steroid and Triterpenoid Testing*

One gram of simplicia is macerated by ether as much as 20 ml for 2 hours and the filtrate is taken. The filtrate is evaporated and dripped with acetic acid and Lieberman-Bourchard reagent. The presence of a green or red color indicates the presence of a steroid / triterpenoid class compound (Djamil, 2009)

*Thin layer chromatography*

Test the content of secondary / chemical metabolites by using the phytocytocytic filtering test (Thin Layer

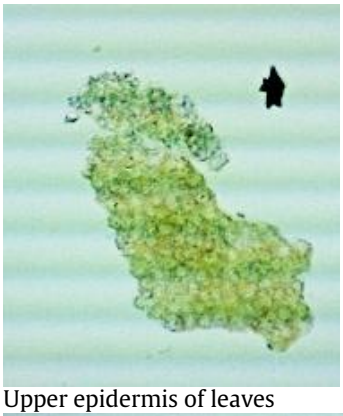
Chromatography) TLC test was carried out by bottling the liquid extract into TLC of silica gel and the mobile phase of chloroform: methanol: water (6: 3: 1).

## RESULTS AND DISCUSSION

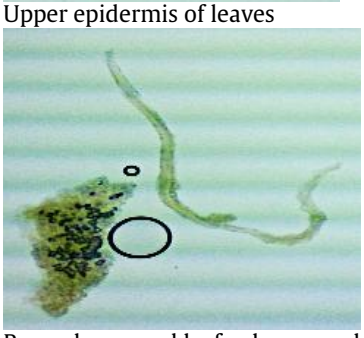
**Table 1**
**Organoleptic Test Results**

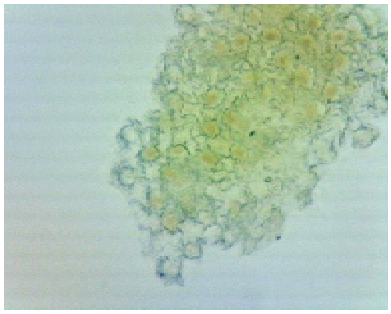
| No | Parameter | Simplicia Leaf | Simplisia Bark |
|---|---|---|---|
| 1 | Forms | Fine powder dry leaves | Fine powder |
| 2 | Colour | Green | Brown |
| 3 | Smell | typical leaves | Typical smell of stems |
| 4 | Taste | bitter | bitter |
| 5 | size | 6.5 cm long, 2.5 cm wide | 5cm long, 2cm wide |

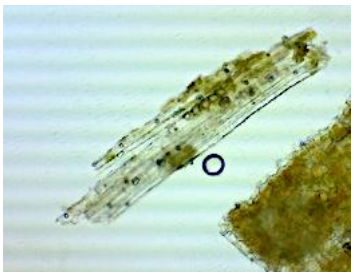
**Table 2**
**Microscopic Image of Leaves and Bark of Pulai (Alstonia Scholaris) (L.) R.Br.**

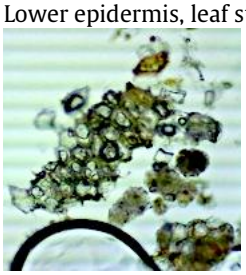

Upper epidermis of leaves


Sclerenchyma and stem bark xylem

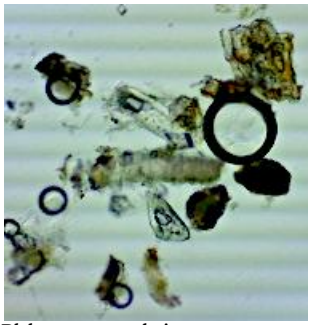

Parenchyma and leaf xylem vessels


Lower epidermis, leaf stomata


Vascular tissue


Parenchymal tissue of stem bark


Crystal


Phloem vessel tissue

## Pulai plant description

The pulai plant has the name ilmia Alstonia scholaris which comes from the phylymic Apocynaceae. This plant is known as the "devil tree". This tree is evenly distributed in the forests of India, the western Himalayas, and southern India. This tree has sizes ranging from medium to large. The largest has a height of about 40 m. the outer skin smells gray to white. The outer bark is cream to yellow and has abundant sap when the body is injured (a characteristic of the Apocynaceae family) (LIPI, 2019)

The leaves are 4-8 crusted, slightly whitish on the abaxial, with about 30-50 leaf bones with very tight spacing and almost straight to the mother leaf bone. Leaf shape oval with the number of leaves per stalk about 4-8. Dark green leaf surface and white green on the underside. The ends are rounded and tapered towards the base. (Meena, 2011)

Terminalis inflorescences, have inflorescences 7-13cm in size. white flower color to greenish white tubular. Flower crown measuring 7-9mm. fruit shaped like bumbung, pairs with a size of about 20-5- cm (LIPI, 2019).

## Organoleptic and microscopic tests

Organoleptic tests on plants include shape, color, smell, taste, and size. The following are the results of the organoleptic test on the simplicia of the leaves and bark of the pulai.

For microscopic examination of the simplicia of the leaves, this plant has an upper and lower epidermis. In the lower epidermis there are stomata of the parasitic type. In addition, it is clear that the xylem vessels are present.

The microscopic part of the stem bark has a characteristic prism-shaped crystal. In addition, another prominent characteristic is the presence of the sclereid of the brachyclerid type. The xylem and phloem vessels are very clearly visible. Another part that shows the presence of cortical parenchyma which is usually used as a place to store food reserves (Shypunov, 2018). The results of leaf microscopy observations and pulai bark can be seen in table 2.

## Leaf and Bark Characteristics Test of Pulai

The next test result is sympathetic characterization examination. From table 3, the results obtained from the water content parameter of 5%, which means that this parameter has met FHI standards below 10% and meets drug standardization (Saifudin, 2011) The second parameter is drying loss. This test aims to determine the maximum limit of compounds that are lost during drying shrinkage (Noviyanti, 2019). The drying shrinkage of pulai leaves has a higher value, which means that the compound content is lost more than the bark of the stem.

The next parameter is the ash content, for the ash content itself is divided into three parameters. For the ash content itself that has met the standards, namely the water soluble ash content <20%. (Ministry of Health, 1977).

The next test is the content of the extract, the content of the extract from table 3 shows that the amount of water solvent extracted on the leaves is 7% and the bark is 3.2%. While the ethanol solvent extracted on the leaves was 3% and the bark was 5.4%.

*Table 3*
*Result of Simplicia Characterization Examination of Leaf of Pulai Alstonia scholaris (L.) R.Br*

| No | Parameter | Leaves | Bark | Standard MMI |
|---|---|---|---|---|
| 1 | Water content | 5% ± 0,01 | 5% ± 0,01 | <10% |
| 2 | Shrink drying | 16% ± 0,01 | 4% ± 0,01 | - |
| 3 | Total ash content | 7 % ± 0,01 | 7 % ± 0,01 | - |
| 4 | Water soluble ash content | 1 % ± 0,01 | 3,5 % ± 0,01 | <20 % |
| 5 | Acid insoluble ash content | 1 % ± 0,01 | 1 % ± 0,01 | - |
| 6 | Water soluble content | 7 % ± 0,001 | 3,2% ± 0,001 | - |
| 7 | Ethanol soluble content | 3 % ± 0,01 | 5,4% ± 0,01 | - |

## Identification of Secondary Metabolite Compounds

Phytochemical identification in simplicia is carried out to determine secondary metabolite compounds contained in simplicia. Based on the phytochemical results of secondary metabolites found in simplicia and extracts, there are alkaloid, flavonoid, saponin, phenol, and stroid / triterpenoid compounds (Tables 4 and 5). These secondary metabolites have health properties between doctors, pharmacists and nurses in the form of herbal medicines and are used by the community in promoting, preventive, curative, and rehabilitative efforts. (MOH, 2016).

**Table 4.**
**Results of Simplicia Phytochemical Dissipation of Pulai (Alstonia scholaris) Leaf Extract (L.) R.Br.**

| No | Compound | Screening results | | |
|---|---|---|---|---|
| | | Simplicia | Extract | |
| | | | Brew | Infussion |
| 1 | Alkaloid | + | + | + |
| 2 | Flavonoid | + | + | + |
| 3 | Saponin | + | + | + |
| 4 | Tanin | - | - | - |
| 5 | Kuinon | - | - | - |
| 6 | Steroid/Triterpenoid | + | + | + |

Note : (+) = detected
(-) = Not detected

**Table 5**
**Phytochemical Screening Result of Simplicia Stem Bark Extract of Pulai (Alstonia Scholaris) (L.) R.Br.**

| No | Compound | Screening results | | |
|---|---|---|---|---|
| | | Simplicia | Extract | |
| | | | Brew | Infussion |
| 1 | Alkaloid | + | + | + |
| 2 | Flavonoid | + | + | + |
| 3 | Saponin | + | + | + |
| 4 | Tanin | - | - | - |
| 5 | Kuinon | - | - | - |
| 6 | Steroid/Triterpenoid | + | + | + |

Note: (+) = detected
(-) = Not detected

**Table 6.**
**Results of TLC of Pulai (Alstonia Scholaris) Bark Extract (L.) R.Br.**

| Sample | UV254 | Appearance of Spotting | Spotting | Compound |
|---|---|---|---|---|
| **Steeping Leaves** | Yellow | $H_2SO_4$ | Green- yellowish | Flavonoid |
| | | | purple | Triterpenoid |
| | | DPPH | Yellow | Flavonoid |
| **Leather steeping** | Yellow | $H_2SO_4$ | Yellow | Flavonoid |
| **Leaf Stew** | Orange | Dragendroff | Orange | Alkaloid |
| | | $H_2SO_4$ | Purple | Triterpenoid |
| **Skin Decoction** | Purple | $H_2SO_4$ | Purple | Triterpenoid |

The results of TLC in (table 6) the leaf and skin steeping contains flavonoids which are indicated by the presence of a yellow color after being sprayed by the appearance of the spots. From (table 5) pulai leaf infusion contains alkaloid compounds in the presence of orange spots sprayed by dragendroff spots.

In addition to testing for flavonoids and alkaloids, triterpenoid compounds were tested which showed a purple color when sprayed by 5% H2SO4.

## CONCLUSIONS AND RECOMMENDATIONS

From the results of the research, this plant from the organoleptic test has similarities such as simplicia, in general, it only differs from size. The microscopic test has characteristic crystals on the bark whereas for the characteristics of this plant that meets the MMI criteria, namely water content and total ash content, while secondary metabolites contain alkaloids, flavonoids, steroids, and triterpenoids.

## Declaration of Conflicting Interests

The authors declared that no potential conflicts of interests with respect to the authorship and publication of this article.

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