



Standardization And Phytochemical Testing Of Purple Leaf Simplicia (*Graptophyllum Pictum*)

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ABSTRACT

Abstract: Background: Purple leaf (*Graptophyllum pictum*) is a medicinal plant widely used in traditional medicine due to its bioactive compounds, including flavonoids, alkaloids, and steroids. To ensure its safety, quality, and efficacy as a herbal raw material, standardization and phytochemical evaluation are essential. Objective: This study aimed to determine the standardization parameters and phytochemical characteristics of purple leaf simplicia. Methods: The study employed qualitative and quantitative approaches. Purple leaves were cultivated, harvested, processed into simplicia powder, and extracted using the maceration and Soxhlet methods with 96% ethanol. Standardization included moisture content, drying loss, total ash, water-soluble extractive value, ethanol-soluble extractive value, and thin-layer chromatography (TLC) analysis. Data were analyzed descriptively. Results: The cultivation process produced 9 g of simplicia powder. The moisture content, drying loss, total ash, water-soluble extractive value, and ethanol-soluble extractive value were 47.3%, 27.6%, 21.7%, 0.227 g, and 10 g, respectively. Soxhlet extraction yielded 20 mL of concentrated extract. Thin-layer chromatography analysis under UV 254 nm produced Rf values of 0.625, 0.750, and 0.8125, indicating the presence of phytochemical constituents in the extract. Conclusion: Purple leaf simplicia can be successfully standardized using physicochemical parameters and TLC analysis. The findings provide baseline information for quality control and support the utilization of *Graptophyllum pictum* as a potential raw material for herbal medicine development

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1. INTRODUCTION

The history of traditional medicine in the world includes the use of a variety of natural ingredients, such as plants, minerals, and animals, for medicine and health care. This practice has been around since prehistoric times and has continued to evolve over time. In prehistoric times, humans have relied on natural ingredients as medicines, including plants, fruits, roots, and leaves. The use of natural remedies used by prehistoric humans can be found in archaeological records, such as the remains of plants at prehistoric sites, such as the discovery of archaeological sites that people used in Iraq mallow (*small mallow*) and yarrow (*Achillea millefolium*) about 60,000 years ago to treat wounds, respiratory infections, digestive problems, skin diseases, and liver diseases. (Nanda Setyani. 2022.)

In the Mesopotamian civilization (3500-300 BC) the Sumerians in Mesopotamia used plants and minerals for medical purposes. Prehistoric records on clay prisms provide information on the use of traditional medicine at the time, an example of which is one well-documented nail-shaped medical source is the *Uruanna Lexicon: maštaka*). It contains about 1300 terms for drugs of plant origin that, taking into account variants and synonyms, describe about



340 different plants. Plants (plants, trees and shrubs, spices, grasses, algae, aromatic plants and fungi). (Yuli Widiastuti. 2017.)

Traditional medicine continues to develop in modern civilization as it is today. Over time, traditional medicine has developed and adapted to social, cultural, and scientific changes, and traditional medicine is still used today.

Traditional *Chinese Medicine (TCM)* which uses a variety of herbal plants, such as ginseng, ginkgo biloba, and echinacea, to treat a wide variety of diseases, including heart disease, stroke, and cancer, in India *Ayurveda* is a traditional Indian medicine system that has been used for more than 5,000 years. *Ayurveda* uses a wide variety of herbal plants, such as turmeric, tamarind, and ginger, to treat a wide variety of ailments, including digestive diseases, skin diseases, and heart disease, in Japan *Kampo* is a traditional Japanese medicine system that has been used for over 1,000 years. *Kampo* uses a wide variety of herbal plants, such as ginseng, ginkgo biloba, and tamarind, to treat a wide variety of ailments, including digestive diseases, skin diseases, and heart disease. At this time traditional medicine in the world is often used as an alternative or complementary medicine to modern medicine. Traditional medicine can be a good option for people who have allergies to modern medicines or who want to avoid the side effects of modern medicines. (Ministry of Health of the Republic of Indonesia. 2000.)

Traditional medicine has been used by the Indonesian people since ancient times. Evidence of the use of traditional medicine has been found at various archaeological sites in Indonesia, including in the Leang-leang Cave in South Sulawesi, which is estimated to be more than 40,000 years old. In ancient times, traditional medicine was often used as the only treatment available. Modern medicine only began to develop in the 19th century. However, traditional medicine remains popular in Indonesia, even today. Types of Traditional Medicines in Indonesia.

Traditional medicine plays an important role in the health system in Indonesia. Traditional medicine is used by most Indonesian people, especially people in rural areas. Traditional medicine also plays a role in economic development in Indonesia. The traditional medicine industry in Indonesia has grown rapidly in recent years. (Chemical Test. (2017).

The sample used in the manufacture of simplicia is purple leaves (*Graptophyllum Pictum*) is a herbal plant that has long been used in traditional Indonesian medicine. Purple leaves have a wide range of active compound content, including flavonoids, alkaloids, and steroids. These compounds have a variety of health benefits, including:

Purple leaves have been researched by many researchers, one of which is conducted by Fitri Makkiyah, Nurul Hidayah, and Sri Haryani from the University of Lampung, with the title Antioxidant and Anti-Inflammatory Activity in Purple Leaves (*Graptophyllum pictum*) Year 2021. This study aims to test the antioxidant and anti-inflammatory activity of purple leaves. The results of the study show that purple leaves have strong antioxidant activity. Purple leaves also have strong anti-inflammatory activity, characterized by decreased levels *malondialdehyd (MDA)* and increased rates *glutathione (GSH)*. The results of this study also show that the administration of purple leaf extract can significantly reduce pain, swelling, and bleeding in hemorrhoid patients. Purple leaf extract can also improve the elasticity of the rectal veins and facilitate bowel movements. (Fitri Makkiyah et al, 2021)

another study entitled Development of Purple Leaf Potential Booklet (*Graptophyllum pictum L.*) Study *Antagonism Staphylococcus aureus* and *Candida albicans TE* In 2020, it was carried out by Yunia Dwi Friska from IAIN Palangkaraya. This study aims to determine the effect of purple leaf ethanol extract in inhibiting growth *Staphylococcus aureus* and *Candida albicans*. The results of the study showed that the optimal ethanol extract of purple leaves in inhibiting growth *Staphylococcus aureus* and *Candida albicans* is 60% and 80%, that's because purple leaves contain flavonoids that can function as antibacterial and antifungal. These studies show that purple leaves have a variety of potential benefits, including as an antioxidant, anti-inflammatory, antibacterial, and antifungal. (June 2020)

The purpose of this study is the standardization and phytochemical test of leaf simplification is to ensure the quality, safety, and consistency of plant or herbal products derived from the plant parts.

2. LITERATURE REVIEW

Farmakognosi

Pharmacognosy was created through the combination of two words in Greek. *Pharmacogon* (medicine) and *Gnosis* (knowledge), i.e. knowledge of medicine. The nomenclature of Pharmacognosis was first and most often used by C.A. Seydler, a medical student in Halle/Saale, Germany, who expressly used *Analetica Pharmacognostica* as the main title of his thesis in 1815. In addition, further research has revealed that *Schmidt* had first used the term Pharmacognosis in his monograph entitled *Lehrbuch der Materia Medica* (i.e. notes on lectures on Medical Materia) in 1811 in Vienna. This compilation specifically discusses medicinal plants and their corresponding characteristics.

Simplisia

In the book *Materia Medika Indonesia*, it is defined that simplicia is a natural ingredient that is used as a medicine that has not undergone any processing and unless otherwise stated, in the form of dried ingredients. Simplicia

are differentiated into plant-based simplicia, animal simplicia and pelican (mineral) simplicia. Plant simplicia is a simplicia that is in the form of a whole plant, part of a plant or *plant exudate*. *Plant exudate* is the cell content that spontaneously comes out of the plant or the cell content that is in a certain way removed from the cell or other plant compounds that are in a certain way separated from the plant and are not yet pure chemical compounds. Plant simplicia often originate and are in the form of entire parts of plants, but often in the form of plant parts or organs such as roots, root bark, stems, bark, wood, flower parts and so on. In addition, there are *exudates* such as *gum*, *latex*, *tragacanta*, *oleoresin*, and so on. (Ministry of Health of the Republic of Indonesia, 2000).

Standardization of Simplification

Standardization of simplicia is an effort to ensure the quality of traditional medicine raw materials derived from plants. Standardization is carried out to ensure that the simplicia used have the correct botanical identity, a guaranteed active compound content, and are free from contamination (Rumaseuw *et al.*, 2023). The standardization parameters of simplicia can be divided into two, namely specific parameters and non-specific parameters.

Moisture Content Test

A moisture test is a method to determine the amount of water contained in a material. It is important to test the moisture content to determine the quality and safety of a material (Prasetyo *et al.*, 2019). The *gravimetric* method is based on the principle of evaporation of water present in the material by heating path, then weighed until a constant weight. The weight reduction that occurs is the water content contained in the material. The formula is as follows:

$$\text{Kadar air (\%)} = (W1 - W2) / W1 \times 100$$

Description:

W1 = Initial sample weight (grams)

W2 = Weight of the sample after drying (grams)

The advantage of this method is that the method is simple and easy to do but the level of accuracy is high, the disadvantage of this method is that it takes a long time to get the results of the analyzed sample must be stable to heating. (Ibrahim, M., & Yuliana, W. (2016).

Drying Shrinkage Test

Drying shrinkage test is a method to determine the amount of water lost from a material during the drying process. This test is important to determine the quality and safety of a material, especially materials that will be stored for a long time. The principle of the drying shrinkage test is to measure the residual substance after drying at a certain temperature over a certain period of time. The higher the temperature and drying time, the more water is lost from the material. The formula is as follows:

$$\text{Drying shrinkage (\%)} = (W1 - W2) / W1 \times 100$$

Description:

W1 = Initial sample weight (grams)

W2 = Weight of the sample after drying (grams)

Ash Level Test

Ash content testing is a method to determine the amount of inorganic substances contained in a material. This test is important to determine the quality and safety of a material, especially the material that will be consumed by humans. The principle of the ash content test is to burn the sample at a high temperature until all the organic substances are burned out, then weigh the remaining inorganic substances. The formula is as follows:

$$\text{Drying shrinkage (\%)} = (W1 - W2) / W1 \times 100$$

Description:

W1 = Initial sample weight (grams)

W2 = Weight of the sample after drying (grams)

Water Soluble and Ethanol Soluble Juice Test

a. Water Soluble Juice Test

The water-soluble essence test refers to the measurement of the amount of substances or compounds that can be soluble in water in a sample. Water-soluble cider levels are often required in fields such as the food, pharmaceutical, and chemical industries. In the pharmaceutical industry, it is used to evaluate the extraction of medicinal ingredients from simplicia (the part of the plant that has medicinal properties) and determine the potential activity of the medicinal ingredients. The formula is as follows:

Water soluble cadence:

$$= \frac{\text{Berat sari larut air}}{\text{berat sampel (g)}} \times \frac{\text{jumlah pelarut}}{\text{larutan yang diambil}} \times 100\%$$

b. Ethanol Soluble Test

Ethanol soluble test is an analytical method to determine the percentage of compound content in a material that can be extracted (soluble) in ethanol. This test is very important in the pharmaceutical industry because it is used to



evaluate the extraction of medicinal ingredients from simplicia (the part of the plant that has medicinal properties) and determine the potential activity of the medicinal substance. The formula is as follows:

Soluble Ethanol Characterization:

$$\frac{\text{berat sari larut etanol}}{\text{berat sampel (g)}} \times \frac{\text{jumlah pelarut}}{\text{larutan yang diambil}} \times 100\%$$

Ethnopharmacy

Ethnopharmacy is the study of how the people of an ethnic or region use a medicinal plant or multi-disciplinary science that studies the use of medicines, especially traditional medicines, by a local community. *Ethnopharmacists* are people who explore how a plant is used as a medicine. *Ethnopharmaasis* comes from the words ethno and pharmaceutical. *Ethnophobia* is a tribe or group, while *pharmacy is the science that studies about drugs* (Attamimi, 2003).

Medicinal Plants

Medicinal plants are plants that have been identified and known to have compounds that are beneficial for health. These compounds can be used to prevent and cure diseases, perform certain biological functions, and prevent insect and fungal attacks. Medicinal plants have been used by humans since ancient times and are still widely used today.

Extract

According to the book "*Extraction Techniques*" by Ir. Suparmin, M.Sc., extracts are substances obtained from raw materials by extracting them. Extracts can be a single substance or a mixture of several substances. An extract is a concentrated preparation obtained by extracting the active substance from a vegetable simplicia or animal simplicia using a suitable solvent, then all or almost all of the solvent is evaporated and the remaining mass or powder is treated in such a way that it meets the established standards.

3. RESEARCH METHODS

Types and Material Collection Plans

The method used in this study is a combination of 2 methods, namely qualitative methods and quantitative methods. Quantitative research is research aimed at the processing of numbers. Each data collected is in the form of numbers and the results of the research refer to the data of these numbers. In quantitative research, the use of graphs can help with data measurement.

Meanwhile, qualitative research is not based on numerical data. Qualitative research generally collects data and information in the form of sentences. Although the results are not as accurate as quantitative research, qualitative research can be relied upon to make better conclusions due to the application of applicable theories.

Number of Plants and Number of Samples

The number of purple leaf plants planted was 4 main plants and 5 reserve plants, the 9 plants managed to survive until harvest in the 3rd month. The first weighing result when the plant is still intact is 72 grams, after the leaves are sorted and washed the weighing result is as much as 47 grams, after 5 days of drying the weighing results of the purple leaves are as much as 9 grams, so that the total dry plants that are ready to be used as simplicia only amount to 9 grams of whole plants planted for 4 months, which is very less if you look at the applicable provisions, namely the total plants that should be needed is as much as 250 grams, So to cover the shortage, 100 grams of purple leaf powder was purchased, and 250 grams of dried purple leaves were purchased on one of the online applications that provide herbal products. So the total purchase is 350gram of purple leaf products.

Number of Samples

1. Inclusion criteria and exclusion criteria

Crop inclusion and exclusion criteria refer to certain parameters or standards used to select or evaluate crops to be planted or maintained. Inclusion criteria in planted purple leaves include

- Climate and Environmental Suitability:

Purple leaf plants that have been planted can adjust to the climate and living environment with an average temperature of 27-30 degrees Celsius with a humidity of 91% and an average air pressure of 1,015 hPa-1,023 hPa with fertile loose soil conditions.

- Plant Health and Condition:

Purple leaf plants that have been planted grow well and are free from diseases and pests that can be detrimental to growth. The leaves, stems, and roots are absent showing signs of disease or nutrient deficiencies that are causing the plant to wilt, or the leaves are dull in color.

- Planting Objectives:

The purpose of growing purple leaves is for the manufacture of simplicia products, which will pass specific parameter tests and non-specific parameter tests, as well as phytochemical screening.

- Resistance to environmental stress:

Purple leaf plants that have been planted have a good level of resistance to environmental stressors such as drought or extreme weather.

Meanwhile, the exclusion criteria for planted purple leaves include:

- Severe Physical Damage

Purple leaf plants that have been planted do not have severe physical damage, such as broken stems or excessively damaged leaves.

- Incompatibility with Planting Zoning

The purple leaf plants planted have been adapted to the growth zone, and the plant environment such as weather, humidity temperature, and soil conditions.

- The Importance of High Maintenance

Purple leaf plants require intensive care and good maintenance.

2. Large Sample

The sample size on each plant $\pm$ 27 cm on each plant

3. Sampling Techniques

The technique used in sampling is treated for 3 months or 12 weeks.

Sample Collection Location and Time

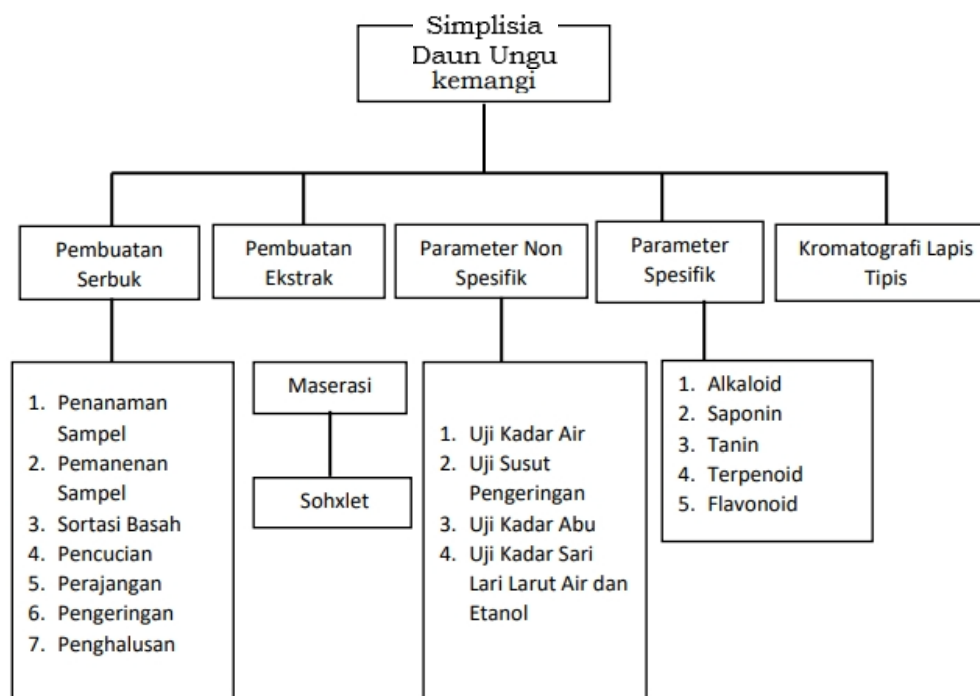
The sample collection location was carried out at the Batam University Laboratory on November 17, 2023, at 13:00 WIB. Where the duration of this sample collection lasts approximately 4 months.

How It Works

In this study, the procedures used are as follows:

1. Powder making consists of simplicia planting, simplicia harvesting, wet sorting, washing, drying, and powder making.
2. The manufacture of extracts consists of maceration and socleration.
3. Non-specific parameters consist of moisture content test, drying shrinkage test, ash content test, total, water soluble juice content test and ethanol soluble juice content test.

The working procedure is as shown in the following chart:





Data Collection

The sample collection technique used is observation (observation). This technique is applied by directly observing the object to be studied, and the *accidental sampling technique*. That is, the researcher first determines the number of samples needed. The advantage of using this technique in sampling is that it is practical because the needs of the research sample have been determined in advance.

Sample collection in this study was carried out with various research methods such as observation and documentation, requiring tools as instruments. The instruments in question are cameras, mobile phones for videos, pens and so on.

4. RESULTS AND DISCUSSION

Sample Collection and Testing Results

In the research on purple leaf plants starting from August 21, 2023 to November 6, 2023, the following results were obtained:

Simplicia Planting Results

On August 21, 2023 at 08.00 WIB, purple leaves were planted. Purple leaves are grown from good seeds on potting media. The planting location was carried out in the Hawaii Garden at a height of 22.1 m. The way simplicia planting works is to prepare the necessary tools and materials such as seeds, loose soil, and pots. Prepare the pot to be used, then sprinkle the seeds into the pot and then water. Basil simplicia planting is carried out for 3 months. Things that must be considered in planting simplicia are soil, fertilizers, seeds, essential factor groups (light, water, nutrients), climate factor groups (rain, air temperature, air humidity, wind and light), and disturbance factor groups (pests, diseases, and weeds). The planting process and planting results are found in appendix 1.

Table 1. Identity Check Results

Yes	Plant Identity	Examination Results
1.	Plant Name	Purple leaves
2.	Latin Names	<i>Graphotphyllum pictum</i>
3.	Plant Parts used	Leaves
4.	Name Indonesia	Purple Leaves

Material Collection Results

Purple leaf plants were obtained as much as 72 grams with a whole result. This collection is carried out using a crackle bag tool. At the time of permanence, purple leaf plants are separated from the soil and then cleaned. Then the wet sorting stage is continued by separating the parts used for making samples and the dirt that is still attached to the plants. Next, washing with PAM water is carried out with the aim of cleaning the dirt that is still attached to the plant.

Next, the display was carried out using scissors. The size of the display is 1-2 cm. This size is taken because the contact between the air surface and the leaf surface is more if it is blended. After that, it is dried by aerating and avoiding exposure to sunlight. The time required for this drying is 5 days. This time is considered sufficient because of the condition of simplicia that has dried completely. After that, the dry sorting process is carried out. Next, the plants are blended into purple leaf powder.

The results of making simplicia powder are obtained as much as 9 grams while 250 grams must be needed (50 grams for testing, 100 grams for products, and 100 grams for maceration). The shortage can be overcome by buying at an online store to meet the number of simplicia needed. The final stage is packing that is packaged in glass bottles and avoided from the sun.

Maceration Extraction Results

The way it works is to prepare tools and materials. The required simplicia weight is weighed as much as 100 grams. The sample is put in a dark bottle. Then the sample is measured in height. Pour 96% ethanol into a dark bottle 2 cm above the sample. Cover the dark bottle with aluminum foil and glue it with rubber band along with glue and insulation. Maceration is carried out for 3 days. The maceration results before screening are measured first after which the filter

The purpose of maceration is to extract active substances that are easily soluble in the extractor liquid, do not expand in the extractor, and do not contain benzoin while the purpose of extraction is to attract all the chemical components contained in the simplicia. The purpose of maceration for 3 days is to maximize the process of taking chemical compounds contained in the sample.

The solvent used in this maceration process is 96% ethanol, because ethanol is a universal solvent that can attract the desired compounds. Ethanol solvents are safer to use because they are neutral compared to other solvents.



The maceration process with 96% ethanol solvent is carried out for 3 days. This aims to maximize the chemical compounds contained in the sample. The result obtained from the maceration that has been filtered is 340 ml.

Extraction Results of Chocolate

The way it works is to prepare tools and materials. Prepare the boiling stones and put them in the base pumpkin. Prepare the filtered solution then put the boiling stone and solution in a 150 ml base flask then turn on the heater. Allow the solvent to evaporate to the top. If there is a solution in the sokhlet tube, turn off the heater if it has almost passed the predetermined limit. Do this repeatedly when the maceration solution has been exhausted and the solution in the sokhlet tube has almost crossed the predetermined limit. Put the solution in the sokhlet tube into the container. The existing solution is boiled and condensed to 20 ml and after 20 ml is put into a vial bottle. The socleration procedure reaches 5 cycles, as shown in the table below.

Table 2. Results of Chocolate Extraction

Cycles	Start Hours	Temperature	Clock Completion	Ethanol Yield
1	05:49 PM	75°C	06.05 PM	41 ml
2	06.09 PM	75°C	06.20 PM	40 ml
3	09.38 WIB	75°C	09.52 AM local time	43 ml
4	10.21 AM	75°C	10.38 a.m.	43 ml
5	10.40 a.m.	75°C	10.54 AM local time	33 ml
Amount of ethanol obtained: 200 ml				

Test Results of Water Soluble Juice and Ethanol Soluble Content

In this practicum, 10 grams of simplicia are needed, each test requires 5 grams. In the water-soluble cider content test, 100 ml of saturated solution derived from a mixture of aquadest and chloroform was given, while in the ethanol soluble content test, 100 ml of 96% ethanol solution was given. The 2 liquids that have been mixed with powder as much as 5 grams, then shake for 6 hours, by stirring every 15 minutes every hour.

After being shaken, the liquid is then filtered and the filtrate is taken as much as 20 ml, to be evaporated with a vaporizer cup that has been tempered at a temperature of 250°C. The evaporation will produce a scale on the cup which is then weighed, and redissolved with 10 drops of ethanol. The result of the vaporizer cup scales along with the scale that has been produced at the evaporation of the water-soluble juice content is 78,790 grams, while the ethanol soluble content is 49,764 grams. The results of the water soluble juice and ethanol test are found in appendices 3 and 4.

The water soluble juice test rate and ethanol soluble test rate can be calculated using the following calculation formula:

➤ Penimbangan

- Weight of water-soluble juice in solvent =
= 78.790 gr – 78.563 gr
= 0.227 g
- Soluble weight of ethanol in solvent =
= 49.764 gr – 39.764 gr
= 10 gr

➤ Percent of water-soluble juice content

$$= \frac{\text{Berat sari larut air}}{\text{berat sampel (gr)}} \times \frac{\text{jumlah pelarut}}{\text{larutan yang diambil}} \times 100\%$$

$$= \frac{0.227}{5} \times \frac{100}{20} \times 100\%$$

$$= 0.0424 \times 5 \times 100\%$$

$$= 0.227 \text{ gr}$$

So the percentage of water-soluble juice obtained is 0.227 grams. On the determination of the herbal pharmacopoeia edition II
In 2017, the content of water-soluble juice was not < 22.6%

➤ Percent ethanol content

$$= \frac{\text{Berat sari larut etanol}}{\text{berat sampel (gr)}} \times \frac{\text{jumlah pelarut}}{\text{larutan yang diambil}} \times 100\%$$

$$= \frac{10}{5} \times \frac{100}{20} \times 100\%$$

$$= 2 \times 5 \times 100\%$$



6. REKOMENDATION

To improve the quality of purple leaf simplicia, further research is needed to determine the content of chemical compounds. For people who want to use purple leaf simplicia as a raw material for traditional medicine, they should first consult with a competent health professional.

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