# Phytochemical Screening And Antibacterial Activity Ethanolic Extract Of *Solanum Mauritianum* Scop Leaves Against *Staphylococcus Aureus* And *Pseudomonas Aeruginosa*

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***Abstract***
*According to the Karo people, Solanum mauritianum Scop leaves are widely used as traditional medicine, which is used topically to treat sprains, wounds, and bruises because of their high phenol content. The purpose of this study was to identify the content of secondary metabolites (alkaloids, flavonoids, saponins, tannins, steroids, triterpenoids, and glycosides) that inhibit the antibacterial activity of Staphylococcus aureus and Pseudomonas aeruginosa by the diffusion method using paper discs from the ethanolic extract of Solanum mauritianum Scop leaves. The results of phytochemical screening showed that Solanum mauritianum Scop leaves were positive for alkaloids, flavonoids, glycosides, saponins, tannins, and steroids. Results The antibacterial activity test of the ethanolic extract of Solanum mauritianum Scop leaves showed effective results at a concentration of 80 mg/mL on Staphylococcus aureus with an inhibition zone diameter of 14.54 ± 0.13 mm. Meanwhile, Pseudomonas aeruginosa showed effective results at a concentration of 300 mg/mL with an inhibition zone diameter of 15.70 ± 0.09 mm. Both bacteria showed activity in the strong category. Conclusion The ethanolic extract of Solanum mauritianum Scop leaves has antibacterial activity against both test bacteria with a strong category.*

***Keywords:*** *Solanum mauritianum Scop leaves, Staphylococcus aureus, Pseudomonas aeruginosa, and antibacterial.*

## I. INTRODUCTION

Microbes that cause infection are extremely easy to assault in people, especially if their immune systems are already compromised. Bacteria, viruses, and fungi are all capable of causing infections [1]. *Pseudomonas aeruginosa, Klebsiella pneumoniae,* and *Escherichia coli* are a few infectious bacteria that can cause skin infections, upper respiratory tract infections, and lower respiratory tract infections. Gynecological infections are also brought on by *E. coli*. Skin infections, lower respiratory tract infections, bone and joint infections, and intraabdominal infections are all brought on by *Staphylococcus aureus*. An opportunistic bacteria called *Proteus vulgaris* is in charge of producing urinary tract infections and wound infections [2]. A form of bacteria called *Staphylococcus aureus* causes pneumonia, skin infections, skin tissue infections, and upper respiratory tract infections. Erythromycin is one of the most popular antibiotics used to treat infections brought on by *Staphylococcus aureus* [3]. *Pseudomonas aeruginosa* is another bacterium that is similarly dangerous and pathogenic due to its amazing capacity to acquire new drug resistance as well as its virulence, or the ability of pathogenic bacteria to cause damage [4].An efficient and secure antibacterial agent that needs more research is needed to handle issues relating to bacterial infection.

Compared to synthetic medications, the development of natural medicine currently offers a huge possibility in the treatment of infectious diseases. In order to address the issue of antimicrobial resistance against the careless use of antibiotics, research and development into the discovery of antibacterial compounds derived from natural ingredients continues. Secondary metabolites found in abundance in plants, including tannins, lignins, carotenoids, flavonoids, and alkaloids, are thought to have antibacterial properties that prevent the growth of dangerous microorganisms [5], [6].According to the Karo Tribe in North Sumatra,

*Solanum mauritianum* Scop leaves are used as medicine for sprains and to treat wounds on the skin by applying the leaves to the injured part, using boiled water to treat colds. Based on the foregoing, the researchers will study the *Solanum mauritianum* Scop species by evaluating the ethanol extract of the plant's leaves' ability to inhibit the growth of *Staphylococcus aureus* and *Pseudomonas aeruginosa* using the agar diffusion method.

## II. METHODS

### 2.1 Apparatus

The Apparatus used in this study were glassware (petri dish, beaker glass, erlenmeyer, measuring cup), autoclave (Tomy), incubator (Memmert), caliper (sigmat vernier), Laminar Air Flow Cabinet (BioBase).

### 2.2 Material

The materials used in this study were *Solanum mauritianum* Scop leaves, *Staphylococcus aureus* (ATCC 25923) and *Pseudomonas aeruginosa* (ATCC 8027) bacterial cultures. The media used were Muller Hinton Agar / MHA (Oxoid) and Muller Hinton Broth / MHB (Oxoid), and dimethylsulfoxide / DMSO (SmartLab).

### 2.3 Plant Identification

Plant identification was carried out in the plant laboratory "Herbarium Medanense" Faculty of Mathematics and Natural Sciences Department of Biology, Universitas Sumatera Utara.

### 2.4 Preparation of Dried Powder of *Solanum mauritianum* Scop leaves

*Mauritian Solanum* scop leaves were collected and thoroughly cleaned, then drained and weighed as wet weight. It is then dried at 40$^{o}$C in a drying chamber. The dried substance is then ground into powder using a blender after being weighed as dry weight. [7].

### 2.5 Characterization of Dried Powder

Examination of the characteristics of dried powder includes determination of water content, determination of water soluble content, determination of ethanol-soluble essence, determination of total ash content, determination of acid insoluble ash content. [8], [9].

### 2.6 Preparation of Ethanol Extract *Solanum mauritianum* Scop leaves

Using a maceration ratio of 1:10, the ethanol extract of *Solanum mauritianum* Scop leaves was created. 10 parts of 96% (pa) ethanol solvent and 1 part of *Solanum mauritianum* Scop leaves dried powder were combined in a container. Soaked for six hours, occasionally stirred, and then left to stand for eighteen hours. Next filtered once more, the filtering procedure was used, after which all of the macerate was collected and evaporated with a rotary evaporator to produce a thick extract [4].

### 2.7 Phytochemical Screening

*Solanum mauritianum* Scop leaves dried powder and an ethanol extract were subjected to phytochemical screening, which included determine for alkaloids, glycosides, saponins, flavonoids, tannins, and steroids or triterpenoids [10], [11].

### 2.8 Sterilization of Apparatus and Materials

Glass were used in this study were sterilized by the dry heat method using an oven at 170$^{o}$C for 1 hour. The media used in this study were sterilized using the wet heat method by autoclaving at 121$^{o}$C for 15 minutes [5], [12].

### 2.9 Preparation of Test Solution of Various Concentration

The ethanol extract was weighed up to 5 g, then dissolved in dimethylsulfoxide (DMSO) to 10 mL (concentration 500 mg/mL), then diluted with 400, 300, 200, 100, 80, 60, 40 (mg/mL) [12], [13].

### 2.10 Antibacterial Activity Test

A total of 0.1 mL of the test bacterial suspension was mixed with 15 mL of MHA media in sterile petri dishes, then allowed to solidify. On the solidified media, discs that had been soaked in the test solution were placed, then incubated at 37oC for 18–24 hours [14], [15].

## III. RESULT AND DISCUSSION

### 3.1 Plant Identification Result of *Solanum mauritianum* Scop leaves

Based on the results of identification carried out in the plant laboratory "Herbarium Medanense" Department of Biology, Faculty of Mathematics and Natural Sciences, Universitas Sumatera Utara with the number 6708/MEDA/2021 is a plant species *Solanum mauritianum* Scop leaves with complete classification as follows :

Kingdom : Plantae
Division : Spermatophyta
Class : Dicotyledoneae
Ordo : Solanales
Family : Solanaceae
Genus : Solanum
Spesies : *Solanum mauritianum* Scop.
Local name : Daun Lancing

Pictures of *Solanum mauritianum* Scop leaves can be seen in Figure 1.

**Fig 1.** *Solanum mauritianum* Scop leaves

### 3.2 Examination of Dried Powder Characteristics

In order for dried powder to be used as a medical ingredient, certain conditions must be met, and this examination of dried powder's quality attributes serves as the means of determining values for various product parameters. The *Solanum mauritianum* Scop leaves plant is still not classified in Materia Medika Indonesia, therefore it lacks a standard parameter reference that can be utilized. Dried powder parameters used as medicinal raw materials must match the conditions given in the official monograph of Materia Medika Indonesia. Table 1 displays the findings of the characterisation analysis of *Solanum mauritianum* Scop leaves dried powder.

**Table 1.** Characterization of *Solanum mauritianum* Scop leaves dried powder

| No | Parameter | Result (%) |
|---|---|---|
| 1 | Water content | 5,94 |
| 2 | Water-solube content | 16,67 |
| 3 | Ethanol-solube content | 14,73 |
| 4 | Total ash content | 1,69 |
| 5 | Acid insoluble ash content | 0,69 |

The water content based on the requirements is generally not more than 10% where the results of characterization of the moisture content of *Solanum mauritianum* Scop leaves get a value of 5.94% [16]. If the water content obtained in this examination exceeds 10%, it will be a good medium for the growth of microorganisms, especially fungi so that the shelf life and quality of the dried powder will be reduced and even damaged [17]. Determination of water soluble extract content aims to determine the levels of chemical compounds from dried powder that are polar such as flavonoids, glycosides, and saponins [18]. About

16.67% of the extracted water-soluble material was found. While the ethanol soluble extract content seeks to identify the polar, semipolar, and nonpolar chemical components. The extracted soluble ethanol concentration was 14.73%. Differences in the soluble molecules in each solvent are possible due to the different levels in the two studies; polar chemicals are therefore more soluble than non-polar ones [11].

The ash content examination aims to provide an overview of internal and external mineral content originating from the initial process of raw materials to become dried powder [19]. Determination of total ash content was carried out to determine the levels of inorganic compounds in dried powder, for example metals K, Ca, Na, Pb, and Hg [20]. The results of the examination of the ash content obtained a value of 1.69%. These results indicate a low ash content where the internal mineral content in *Solanum mauritianum* Scop leaves is quite low. Acid insoluble ash content indicates the presence of mineral or metal contamination that is not soluble in acid [20]. The yield of acid insoluble ash content in dried powder was 0.69%. Acid insoluble ash content indicates the presence of silicate content from soil or sand [21].

### 3.3 Phytochemical Screening of *Solanum mauritianum* Scop leaves Dried Powder

The results of phytochemical screening of dried powder and ethanol extract contained secondary metabolites of alkaloids, flavonoids, glycosides, saponins, tannins and steroids/triterpenoids. The results of phytochemical screening can be seen in Table 2.

**Table 2.** Secondary metabolite compounds found in dried powder and ethanol extract of *Solanum mauritianum* Scop leaves.

| Secondary Metabolites | Dried Powder | Extract |
|---|---|---|
| Alkaloid | + | + |
| Flavanoid | + | + |
| Glikosida | + | + |
| Saponin | + | + |
| Tannin | + | + |
| Steroid / Triterpenoid | + | + |

(+) = Positif

The results of the examination of secondary metabolites showed a positive alkaloid group. This was indicated by the formation of a white precipitate in the Meyer reagent and a reddish brown precipitate in the Wagner and Dragendorff reagents [22]. The three reagents showed positive results where the alkaloids were declared positive if 2 or 3 reagents showed positive results. Examination using $FeCl_3$ on dried powder and ethanol extract of *Solanum mauritianum* Scop leaves showed positive results, namely a color change to strong green, red, purple and black colors [23]. This shows that tannin components are present in both the dried powder and the ethanol extract of *Solanum mauritianum Scop* leaves. Dried powder and ethanol extract of *Solanum mauritianum* Scop leaves also contain saponin components, as demonstrated by the stable foam formation observed after shaking saponins with hot water and adding HCl 2N [24].Flavonoid examination also showed positive results with a marked change in color that occurred, namely reddish black, so both of them were declared positive for containing flavonoids [25]. The reaction using the Lieberman-Bouchard reagent gives a red-purple color (indicating a positive triterpenoid), whereas if it shows a green-blue color (positive triterpenoid) [11], [26]. Both showed positive results on steroids and triterpenoids.

### 3.4 Antibacterial Activity Test

The results of the antibacterial activity test against *Staphylococcus aureus* and *Pseudomonas aeruginosa* showed the formation of an inhibitory zone around the disc in the overall variation of the test concentration. The results of the antibacterial activity test can be seen in Table 3.

**Table 3.** Activity test results of Ethanol Extract *Solanum mauritianum* Scop leaves against *Staphylococcus aureus* and *Pseudomonas aeruginosa*

| Concentration (mg/mL) | Diameter Inhibitory Zones (mm) | | | |
|---|---|---|---|---|
| | *Staphylococcus aureus* | Category [27] | *Pseudomonas aureginosa* | Category [27] |
| 500 | 23,46 ± 0,28 | Very Strong | 19,43 ± 0,21 | Strong |

| | | | | |
|---|---|---|---|---|
| 400 | 21,54 ± 0,16 | Very Strong | 17,47 ± 0,15 | Strong |
| 300 | 19,36 ± 0,18 | Strong | 15,70 ± 0,09 | Strong |
| 200 | 17,68 ± 0,11 | Strong | 13,51 ± 0,20 | Strong |
| 100 | 15,52 ± 0,35 | Strong | 11,68 ± 0,23 | Strong |
| 80 | 14,54 ± 0,13 | Strong | 10,60 ± 0,32 | Strong |
| 60 | 12,56 ± 0,33 | Strong | 9,24 ± 0,14 | Weak |
| 40 | 11,21 ± 0,16 | Strong | 8,29 ± 0,17 | Weak |

The results of the antibacterial test in table 3 show that there are differences in the diameter of the inhibition zone for each concentration of bacteria [2], [28]. It can also be seen in Figure 2. At the smallest concentration of 40 mg/mL, antibacterial activity was still visible against the two test bacteria. In table 3 and figure 2, it can be seen that the inhibition zone formed is larger in gram-positive bacteria than in gram-negative bacteria. This is because the lipopolysaccharide constituents are thicker in gram-negative bacteria compared to gram-positive bacteria [3], [29], [30]. Based on the Farmakope Indonesia VI edition, the diameter of the effective inhibition area was between 14 mm to 16 mm. Where the test results against *Staphylococcus aureus* showed an effective concentration of 80 mg/mL with an inhibition zone diameter of 14.54 ± 0.13 mm.

While the gram negative bacteria showed an effective concentration at 300 mg/mL with an inhibition zone diameter of 15.70 ± 0.09 mm. Inhibition zone categories based on Davis and Stoout (1971) classified 4 categories of inhibition zone diameter, namely no respons (diameter below 5 mm), weak category (5-10 mm), strong category (10-20 mm) and very strong category (above 20 mm) [27]. The result of the phytochemical screening revealed that all investigated classes of secondary metabolites produced favorable outcomes. One of the processes by which saponins operate as antibacterials is by lowering surface tension, which produces an increase in cell permeability and leakage and causes intracellular bacterial components to escape the cell [31]. These metabolites also serve as antibacterials. The capacity of tannins to deactivate microbial cell adhesion, inactivate enzymes, and obstruct protein transport in the inner layer of extracellular protein cells allows them to harm bacterial cell membranes as well as having antibacterial activity [32].

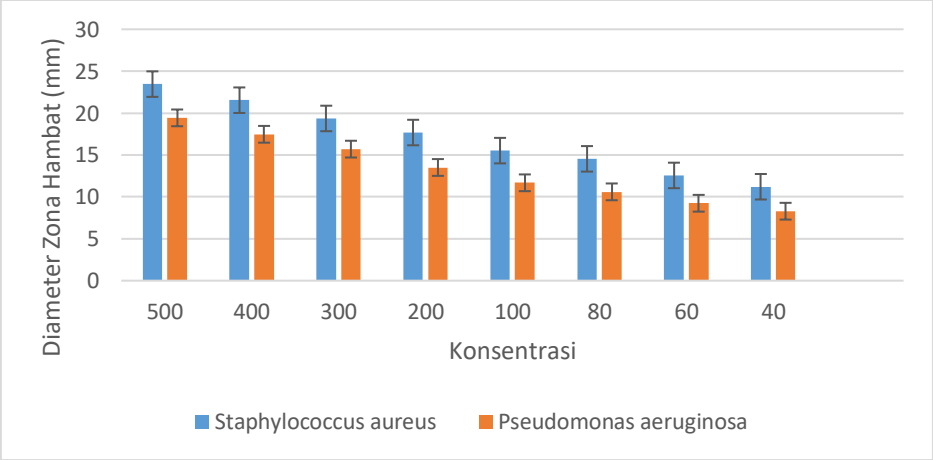


**Fig 2.** Graph of the diameter of the inhibition zone of the ethanolic extract of *Solanum mauritianum* Scop leaves against *Staphylococcus aureus* and *Pseudomonas aeruginosa*

Alkaloids also exert their effects by preventing the formation of bacterial cell walls, which results in cell lysis and death [6]. There is also speculation that other secondary metabolite substances, such flavonoids, have antibacterial properties. In order to damage bacterial cell membranes and cause bacterial cell lysis, which results in the release of intracellular fluid from cells, flavonoids generate complex compounds between their active ingredients and extracellular and dissolved proteins [33]. In accordance with the way that steroids work, which involves harming the cell membrane and letting the cytoplasm escape the cell [34]. This is consistent with Sbhatu's (2020) research, which used an ethanol extract of a plant related to *Solanum mauritianum* Scop called *Solanum incanum* which contains secondary metabolites of saponins, tannins, alkaloids, flavonoids, steroids and also flavonoids against gram-positive and gram-negative bacteria [35].

## IV. RESULT

The dried powder and ethanolic extract of *Solanum mauritianum* Scop leaves were subjected to phytochemical screening, and the results revealed the presence of secondary metabolites of alkaloids, flavonoids, tannins, saponins, glycosides, and steroids/triterpenoids with antibacterial activity against both gram-positive and gram-negative bacteria, including *Staphylococcus aureus* and *Pseudomonas aeruginosa*.

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