



# Screening Phytochemical and Antimicrobial Activity of *Balanophora elongata* Extract

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DOI: 10.47760/ijpsm.2022.v07i08.003

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## Abstract

*Balanophora elongata* is categorized as *Balanophoraceae* which is the most found in West Sumatra on their family. The plants in similar family shown the potential as significant anti radical, cytotoxic in cancer and also have a role in toxification. The aim of this study is to know the antibacterial activity of this plant toward *Pseudomonas aeruginosa*. The method that we use is extraction and antimicrobial test with diffusion disc. The compound in extract consists of terpenoids / steroids, phenolics, flavonoids and negative alkaloid. The antibacterial activity was assayed with diffusion disc method. This study showed that *Balanophora elongata* is effective as anti-*Pseudomonas aeruginosa*. The isolates from this plant were obtained and need more characterization.

**Keywords:** *Balanophora elongata*, *Pseudomonas aeruginosa*

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## INTRODUCTION

*Balanophora* is a parasitic plant that grow on the roots of pine and evergreen trees, especially in the family of Leguminosae, Ericaceae, Urticaceae, and Fagaceae<sup>1, 2</sup>. It grows at an altitude of 900-2800 meters above sea level<sup>3</sup>.

*Balanophora elongata* is one of the species from *Balanophoraceae* families that most commonly found in the area of West Sumatra compared to another species of *Balanophora* (*Balanophora dioica*, *Balanophora fungosa* ssp. *Indica* var. *Indica*) and plants are scattered in the area where the rainfall is about 2500-5000 mm/year. Host of *Balanophora* reported from *Endospermum malacense*, *Villebrunea rubescens*, *Vasculosa ficus*, *Ficus vulva*, *Ficus* sp., *Laportea* sp., *Syzigium cumini*, and *Macaranga triloba*<sup>4</sup>.

Until today, the research about *Balanophora elongata* is only about the morphology differences with another plants in their family, and none of them about the bioactivity of this species. The plants in similar family shown the potential as significant anti radical, cytotoxic to cancer and also have a role in toxification. Based on preliminary test, the methanol extract of this species has anti-bacterial activities. The test was done to 12 bacteria and one of them to *Pseudomonas*



*aeruginosa*. Studies shown that the compounds on the genus *Balanophoraceae* are tannins or other phenolic compounds. Tannin that has phenylacrylic acid derivatives such as cafeoil, cumaroil, furoil or cinnamoyl. In addition, some galoil, cafeoil and esters hexahydroxyphenol, dihidrokalkon glucoside, phenylpropanoid, flavonoids, terpenoids and sterols are also contained in the genus of *Balanophoraceae*<sup>1</sup>.

Another species from *Balanophora* have other biological properties such as antipiretic and antidot, and neutralize the effects of alcohol beverages, and is used as a tonic for the treatment of hemorrhoids, abdominal pain, and hemoptysis by local people in China<sup>2,5</sup>.

The purpose of this study was to screening phytochemical and antimicrobial activity of *Balanophora elongata* Blume extract.

## METHODS

### Tool

The tools needed are maceration bottled, rotary evaporator, water bath, desiccator, vial volume of 20 ml, the plate drops, pipette, spatula, analytical balance, funnels, Erlenmeyer (capacity 250 ml, 500 ml and 2 L) , glass cup (100 ml capacity), funnel (capacity 1 L), measuring cups (capacity 10 ml, 100 ml and 250 ml), oven, aluminum foil, pipette capillary, plate TLC, column chromatography, UV lamp 254, UV-Vis, IR spectrometers, Fisher John melting point apparatus, chamber and silica gel PF 60.

The tools used for testing the antibacterial activity are test tubes, electric scales, micro pipette, pipette, petri dishes, glasses measuring, vials, needles ose, cotton bud, autoclave, a petri dish, bud swab, test tube, calipers, and tweezers.

### Material

The materials used for isolation are *Balanophora elongata* parts (flowers and tuber), distilled water, methanol, hexane, ethyl acetate, 70% ethanol, butanol, citric acid, boric acid, FeCl<sub>3</sub>, sulfuric vanillin.

Materials used in testing the bioactivity are the media Nutrient Agar (NA), distilled water, *Pseudomonas aeruginosa* culture, ciprofloxacin, dimethyl sulfoxide (DMSO), Mueller Hinton Agar (MHA) Media.

The sample used is a parasitic plant *Balanophora elongata* that took in Bancah Nagari Balingka, District IV Koto, Agam, and identified in Herbarium ANDA Universitas Andalas.

### Extraction, fractionation and Skrinning Phytochemicals

*Balanophora elongata* Blume. 880 grams of tubers and 172 grams of the flower and then macerated with methanol, and the solvent is evaporated to obtain extract. Then fractionated by non-polar solvent (hexane), semi-polar (ethyl acetate) and the last with a polar solvent (butanol). Then each of these extracts will be tested antibacterial activity that against *Pseudomonas aeruginosa*. The each extracts then monitored with thin layer chromatography (TLC) plate and the compound was analyzed using specific reagents.

## Test of antibacterial activity against

### *Pseudomonas aeruginosa*

#### a. Paper disc diffusion methods

All the tools for testing are sterilized by autoclave. Media NA 25 ml which had been sterilized was poured into each petri dish and left for some time to solidify. In dense media spread the bacterial suspension by swap previously been adjusted by 0.5 Mc Farland standard (McF) using a sterile spreader rod until the bacterial suspension uniformly across the surface of the media NA 25 ml. Discs with a diameter of 6 mm embedded on the media and was given 10  $\mu$ L of the *Balanophora elongata* extract (10 mg / ml). Ciprofloxacin as positive control and dimethyl sulfoxide (DMSO) as negative control each 10  $\mu$ L with concentration (10 mg / ml). Testing with three repetitions, then incubated at 37°C for 18-23 hours. Inhibition zone was measured and recorded and bioactivity was assessed by a scoring system is very active (> 19 mm), active (13-19 mm), the active half (10-12 mm), inactive (<10 mm).

#### b. Antimicrobial Test

Some extracts from hexane fraction, ethyl acetate, buthanol, and subfraction *Balanophora elongata* created a solution with a concentration of 20%, 10%, 5%, and 2.5% in DMSO. Testing with three repetitions and then incubation at 37°C for 18-24 hours and zones of inhibition were measured and recorded.

## RESULTS AND DISCUSSION

### Sampling

The sample is *Balanophora elongata* Blume., which was found in Bancah Nagari Balingka, District IV Koto, Agam. It was identified on Herbarium Anda Universitas Andalas.

### Extraction

A total of 880 grams of tuber and 172 grams of flower on solvent extraction using methanol for 3 days in 3 repetitions. Thick tuber extract obtained 75 grams with 8.52% yield and 23 grams of flowers viscous extract with 13.37% yield.

### Phytochemicals Test to Extract

The extract was tested by phytochemical reagent for early identification the classes of chemical compounds contained in the extract *Balanophora elongata*. Phytochemicals test using a mobile phase Silica Gel with eluent hexane-ethyl acetate-methanol (1:4:1). With Dragendorff reagent to detect alkaloid; Sulfuric acid-vanilin to detect terpenoids and steroids; FeCl<sub>3</sub> solution 5%o for detect phenolic and Sitroborat to detect flavonoid. The test result shows the extract contained terpenoid/ steroid, phenolic and flavonoid compounds.

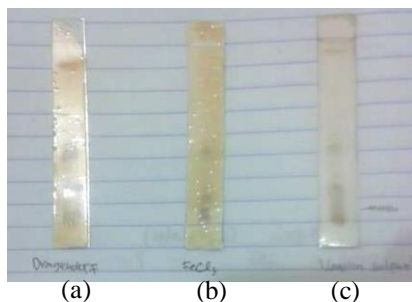


Figure 1. The Phytochemicals results of Methanol Extracts *Balanophora elongata* (a. Dragendorff, b. FeCl<sub>3</sub>, c. Vanillin-sulphate).

Description:

Alkaloids : Negative  
Phenolics : Positive  
Terpenoids/steroid : Positive

**Preliminary Extracts Test against *Pseudomonas aeruginosa***

Preliminary test results showed that the methanol extract of *Balanophora elongata* has activity to against *Pseudomonas aeruginosa* with a diameter of inhibitory 1.9 cm for flower extracts categorized as very active in inhibiting the growth of bacteria and diameter of inhibitory 1.3 cm for extract tuber *Balanophora elongata* categorized as active in inhibiting the growth of bacteria, with a diameter of inhibitory of control 3cm.

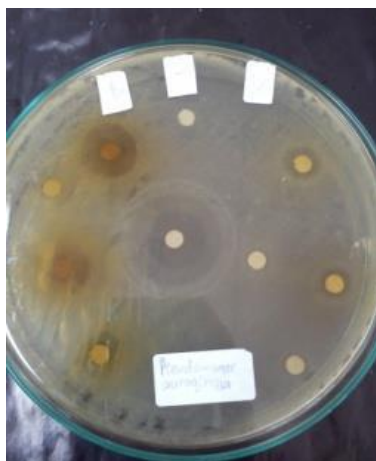


Figure 2. Results of Testing Activities *Balanophora elongata* Methanol Extracts to against *Pseudomonas aeruginosa* bacteria.



Goldha Faroliu, International Journal of Pharmaceutical Sciences & Medicine (IJPSM),  
Vol.7 Issue. 8, August- 2022, pg. 20-24

ISSN: 2519-9889  
Impact Factor: 5.721

## CONCLUSION

The study about screening phytochemical and antimicrobial test of *Balanophora elongata* Blume. which is active against clinical isolates of *Pseudomonas aeruginosa* and the results obtained extract of the flower of *Balanophora elongata* Blume active against *Pseudomonas aeruginosa* and the classes of compounds that have been identified are phenolics, flavonoids and terpenoids/steroids.

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