Antibacterial Activity of Ethanol Extract and Ethyl Acetate of Ginseng Bugis (Talinum Paniculatum Gaertn.) Leaves Against Staphylococcus Aureus and Escherichia coli Bacteria

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Abstract. Ginseng bugis leaf (Talinum paniculatum Gaertn.) contains flavonoids which are potential as antibacterial compounds. The aims of the research are to obtain the antibacterial activity of ethanol and ethyl acetate extracts from ginseng bugis leaves against Staphylococcus aureus and Escherichia coli. This study used ethanol and ethyl acetate extracts divided in 8 concentration series, namely 0.1%, 0.5%, 1%, 2%, 4%, 8%, 16%, and 32%. Testing antibacterial activity using the agar diffusion. The results of the screening test showed that ethanol extract and ethyl acetate of ginseng bugis leaf have inhibitory concentration at 0.5% against the test bacteria. The results of testing the antibacterial activity of ethanol extract and ethyl acetate have MIC value at a concentration of 2% and MBC value at a concentration of 8%.

Keywords: Talinum Paniculatum Gaertn, Staphylococcus aureus, Escherichia coli, Extract ethanol, Extract ethyl acetate

1. Introduction

Infectious disease is one of the problems in the health sector, which gradually continues to develop. One of the factors is bacteria [1]. Pathogenic bacteria are more dangerous and cause infections, both sporadically and endemically. It includes Staphylococcus aureus (SA), Escherichia coli (EC), and Pseudomonas aeruginosa (PA) [2]. Staphylococcus aureus is gram-positive bacteria found on the skin of normal human flora. [3] Staphylococcus aureus can cause infection in hair follicles and sweat glands, ulcers, and infection in wounds. These bacteria have low invasion ability and cause a variety of skin infections [4]. Escherichia coli is gram-negative bacteria of Enterobacteriaceae family. Escherichia coli bacterial infection in humans can lead to bloody or non-bloody diarrhea [5].

People in South Sulawesi are familiar with Ginseng Bugis (Talinum Paniculatum Gaertn.) as traditional medicinal plants. The use of Talinum paniculatum Gaertn leaves can treat various ulcers, coughs with phlegm and blood, pneumonia, cold sweat, diarrhea, lots of urine, irregular menstruation, vaginal discharge, immunomodulators, e.t.c. Therefore, they have been proven to be a nutritious, medicinal plant empirically [6,7]. In general, ginseng bugis contains chemical substances including saponins, flavonoids, steroids, polyphenols and tannins [8].
2. Research methods

2.1. Extraction
Ethanol extract of ginseng bugis was made by maceration. The simplicia powder of Ginseng Bugis leaves (Talinum paniculatum Gaertn) was put into the maceration vessel and added with 70% ethanol. Then, it resulted in the liquid extract at the rotary evaporator. Next, it went for freeze-drying until dry extract was obtained [9]. The same method was applied to ethyl acetate extract.

2.2. Preparation of test microbes
2.2.1. Rejuvenation of bacteria. Rejuvenation bacteria tested (Staphylococcus aureus and Escherichia coli) were taken from pure colonies of each and inoculated by scratching on an oblique Nutrient Agar (NA) medium. Afterwards, the incubation process took for 24 hours at 37ºc [10].

2.2.2. Bacterial Suspension. Bacterial suspension preparation test (Staphylococcus aureus, Escherichia coli) and the rejuvenation result were suspended by sterile physiological NaCl solution. The transmittance was evaluated at 25% using a spectrophotometer which used 0.9% physiological NaCl solution at a wavelength of 580 nm [11].

2.3. Antibacterial screening test.
Ethanol and ethyl acetate extract was weighed as much as 10 mg, 50 mg, and 100 mg respectively and put into a sterile vial. Then the extract was dissolved with 0.2 mL of DMSO. After dissolving, added 9.8 mL Nutrient Agar (NA). The mixture was homogenised and poured into a petri dish and allowed to solidify. The suspended bacteria were taken as much as 20 µL and rubbed over the solidified medium. Next, it went into incubation for 24 hours. Then, it observed antibacterial activity indicated by bacterial growth [12].

2.4. Testing for antibacterial activity
2.4.1. Determination of Minimum Inhibitory Content. The ethanol extract and ethyl acetate extract obtained were evaluated for Minimum Inhibitory Concentrations (MICs) against Staphylococcus aureus and Escherichia coli bacteria at different concentrations of 0.1%, 0.5%, 1%, 2%, 4%, 8%, 16% and 32%. It used agar diffusion method. Nutrient Agar Medium were put into a sterile vial and added with bacterial suspension. Next, it was homogenised and poured into a sterile petri dish. Then it was allowed to solidify. The blank disc was inserted into vial containing ethanol extract and ethyl acetate extract with various concentrations (0.1%, 0.5%, 1%, 2%, 4%, 8%, 16%, 32%) and placed on the medium containing the bacterial suspension. Subsequently, it was incubated at 37 ºC for 24 hours to see the clear zone around the blank disc [13].

2.4.2. Determination of the minimum kill levels. The minimum kill levels determination was observed on the clear zone of the minimum inhibitory level assay. If it found a clear zone, then it would repeat incubation for 24 hours. Afterwards, the clear zone on the petri was observed to know if the petri did not show bacteria growth. Then, the clear zone indicated the Minimum bactericidal concentration (MBC) [13].

3. Results and discussion
3.1. Antibacterial screening
The screening test was carried out using two types of bacteria, consisting of Staphylococcus aureus and Escherichia coli because these bacteria are gram-positive and gram-negative bacteria. In addition, it aims to observe the effectiveness of ethanol extract and ethyl acetate extract of ginseng bugis leaves against the tested bacteria. At this stage, the results obtained for the ethanol and ethyl acetate extract of ginseng bugis (Talinum paniculatum Gaertn) can inhibit bacteria at a concentration of 0.5%.
3.2. Testing for antibacterial activity

3.2.1. Determination of Minimum Inhibitory Concentrations (MICs) levels. The results of the MICs of ginseng bugis ethanol extract against *Staphylococcus aureus* and *Escherichia coli* can be seen in Table 1, Figure 1, and Figure 2.

Table 1. The results of the MICs of ginseng bugis leaves ethanol extract against *Staphylococcus aureus* and *Escherichia coli* bacteria.

| Bacteria | R | 0.1 % | 0.5 % | 1% | 2% | 4% | 8% | 16% | 32% |
|----------|---|-------|-------|----|----|----|----|-----|-----|
| SA       | 1 | 6.02  | 6.10  | 6.11| 7.07| 7.36| 8.02| 9.13 | 9.58 |
|          | 2 | 6.04  | 6.10  | 6.13| 7.07| 7.38| 8.11| 9.28 | 9.58 |
|          | 3 | 6.04  | 6.08  | 6.16| 7.07| 7.38| 8.11| 9.29 | 9.61 |
|          | average | 6.03 | 6.09  | 6.13| 7.07| 7.37| 8.08| 9.23 | 9.59 |
| EC       | 1 | 6.04  | 6.09  | 6.32| 7.24| 7.92| 8.18| 9.08 | 9.55 |
|          | 2 | 6.04  | 6.11  | 6.47| 7.27| 7.93| 8.24| 9.19 | 9.58 |
|          | 3 | 6.04  | 6.12  | 6.48| 7.27| 7.99| 8.28| 9.22 | 9.58 |
|          | average | 6.04 | 6.10  | 6.42| 7.26| 7.94| 8.23| 9.16 | 9.57 |

Based on the results obtained in testing of the minimum inhibition level of ethanol extract, ginseng bugis can inhibit *Staphylococcus aureus* bacteria at a concentration of 2% with a diameter of 7.07 mm. This concentration is the smallest concentration that has an inhibition zone around the paper disk. Meanwhile, *Escherichia coli* bacteria can be inhibited by ethanol extract at a concentration of 2% with a diameter of 7.26%.

![Figure 1](image1.png)

**Figure 1.** The results of MICs of ginseng bugis leaves ethanol extract against *Staphylococcus aureus* bacteria. A. Concentrations: 0.1%, 0.5%, and 1%. B. Concentrations: 2%, 4%, and 8%. C. Concentrations: 16% and 32%.
Figure 2. The results of the MICs of ginseng bugis leaves ethanol extract against *Escherichia coli* bacteria. A. Concentrations: 0.1 %, 0.5%, and 1%. B. Concentrations: 2 %, 4%, and 8%. C. Concentrations: 16 % and 32%.

The MIC test results of the ethyl acetate extract of ginseng bugis leaves against *Staphylococcus aureus* and *Escherichia coli* bacteria can be seen in Table 2, Figure 3, and Figure 4.

Table 2. The results of the MICs of ethyl acetate extract of ginseng bugis leaves against *Staphylococcus aureus* and *Escherichia coli* bacteria.

| Bacteria | R  | 0.1 % | 0.5 % | 1%  | 2%  | 4%  | 8%  | 16% | 32% |
|----------|----|-------|-------|-----|-----|-----|-----|-----|-----|
| SA       | 1  | 6.07  | 6.14  | 6.49| 7.70| 8.33| 9.09| 14.60| 17.17|
|          | 2  | 6.08  | 6.14  | 6.50| 7.69| 8.41| 9.10| 14.71| 17.15|
|          | 3  | 6.08  | 6.15  | 6.53| 7.70| 8.42| 9.10| 14.75| 17.17|
| average  |    | 6.07  | 6.14  | 6.50| 7.69| 8.38| 9.09| 14.68| 17.16|
| EC       | 1  | 6.07  | 6.17  | 6.41| 7.02| 7.80| 8.94| 14.73| 15.43|
|          | 2  | 6.07  | 6.17  | 6.42| 7.04| 7.82| 8.95| 14.71| 15.43|
|          | 3  | 6.07  | 6.17  | 6.42| 7.04| 7.82| 8.95| 14.73| 15.47|
| average  |    | 6.07  | 6.17  | 6.41| 7.03| 7.81| 8.94| 14.72| 15.44|

The ethyl acetate extract of ginseng bugis leaves can inhibit the bacterial growth of *Staphylococcus aureus* at a concentration of 2% at 7.69 mm. The same thing occurs on *Escherichia coli* bacteria, ethyl acetate extract can inhibit the growth at a concentration of 2% and with a diameter of 7.03 mm.

Figure 3. The results of the MICs of ethyl acetate extract of ginseng bugis leaves against *Staphylococcus aureus* bacteria. A. Concentrations: 0.1 %, 0.5%, and 1%. B. Concentrations: 2 %, 4%, and 8%. C. Concentrations: 16 % and 32%.
3.2.2. Determination of the minimum kill rate. The results of Minimum Kill Concentrations (MBCs) assay of ginseng bugis leaves ethanol extract against *Staphylococcus aureus* and *Escherichia coli* can be seen in Table 3, Figure 5, and Figure 6

**Table 3.** The results of the MBCs of ginseng bugis leaves ethanol extract against *Staphylococcus aureus* and *Escherichia coli* bacteria

| Bacteria | R | Diameter of Inhibitory Zona (mm) |
|----------|---|----------------------------------|
|          | 8% | 16% | 32% |
| SA       | 1  | 7,01 | 7,48 | 8,02 |
|          | 2  | 7,10 | 7,52 | 8,07 |
|          | 3  | 7,10 | 7,52 | 8,07 |
|          | average | 7,07 | 7,50 | 8,05 |
| EC       | 1  | 6,38 | 7,21 | 7,50 |
|          | 2  | 6,50 | 7,27 | 7,50 |
|          | 3  | 6,50 | 7,28 | 7,52 |
|          | average | 6,46 | 7,25 | 7,50 |

Based on the evaluation, the MBCs value of ginseng bugis leaves ethanol extract can hinder the growth of *Staphylococcus aureus* bacteria at a concentration of 8% and diameter of 7.07 mm. Meanwhile, ethanol extract can result in inhibition of *Escherichia coli* bacteria at a concentration of 8% with a diameter of 6.46%.
Figure 5. The results of the MBCs of ginseng bugis leaves ethanol extract against *Staphylococcus aureus* bacteria. A. Concentrations: 2%, 4%, and 8%. B. Concentrations: 16% and 32%.

Figure 6. The results of the MBCs of ginseng bugis leaves ethanol extract against *Escherichia coli* bacteria. A. Concentrations: 2%, 4%, and 8%. B. Concentrations: 16% and 32%.

The MBCs results of the ethyl acetate extract of ginseng bugis leaves (*Talinum paniculatum* Gaertn) against *Staphylococcus aureus* and *Escherichia coli* bacteria can be seen in Table 4, Figure 7, and Figure 8.

| Bacteria |
|----------|
| SA       |
| 1        | 9.06  | 10.07 | 11.23 |
| 2        | 9.05  | 10.07 | 11.38 |
| 3        | 9.05  | 10.15 | 11.63 |
| average  | 9.05  | 10.09 | 11.41 |
| EC       |
| 1        | 9.03  | 10.78 | 11.02 |
| 2        | 9.04  | 10.79 | 11.04 |
| 3        | 9.04  | 10.79 | 11.04 |
| average  | 9.03  | 10.78 | 11.03 |

Table 4. The results of the MBCs of ginseng bugis leaves ethyl acetate extract against *Staphylococcus aureus* and *Escherichia coli* bacteria.
Based on the study results, the MBCs value of ginseng bugis leaves ethyl acetate extract indicate the inhibition activity of *Staphylococcus aureus* bacteria at a concentration of 8% and diameter of 9.05 mm. Meanwhile, the bacterial growth of *Escherichia coli* can be inhibited by ethyl acetate extract with a concentration of 8% with diameter of 9.03 mm.

![Figure 7](image)

**Figure 7.** The results of the MBCs of ginseng bugis leaves ethyl acetate extract against *Staphylococcus aureus* bacteria. A. Concentrations: 2%, 4%, and 8%. B. Concentrations: 16% and 32%.

![Figure 8](image)

**Figure 8.** The results of the MBCs of ginseng bugis leaves ethyl acetate extract against *Escherichia coli* bacteria. A. Concentration: 2%, 4%, and 8%. B. Concentration: 16% and 32%.

4. **Conclusion**
Based on the research results, it can be concluded that the ethanol extract and ethyl acetate extract of ginseng bugis leaves (*Talinum paniculatum* Gaertn) have antibacterial activity which can inhibit *Staphylococcus aureus* and *Escherichia coli* bacteria at a concentration of 2%. In addition, ethanol extract and ethyl acetate extract of the ginseng bugis leaves have a minimum kill rate of 8% in *Staphylococcus aureus* and *Escherichia coli* bacteria.

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