**Original Research Report**

# ANTIBACTERIAL POTENTIAL OF ENDEMIC MEDICINAL PLANTS OF THE MALUKU ISLANDS, INDONESIA, AGAINST URINARY TRACT INFECTION PATHOGENS

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**ABSTRACT**

Medicinal plants possess antimicrobial properties that have been believed to be efficacious for treating various diseases. However, antimicrobial-based medicinal plants remain an untapped source of potential pharmaceuticals. This study aimed to evaluate the antibacterial properties of ethanolic extracts derived from several medicinal plants endemic to Maluku, Indonesia, specifically regarding their antibacterial activity against isolates of pathogenic bacteria found in the urine samples of urinary tract infection (UTI) patients. This was a true experimental research with a posttest-only control group design. The medicinal plant extraction was conducted using the maceration method. The pathogens were isolated using the dilution method. Following the pathogen isolation, the isolates were identified according to the colony morphology, Gram staining, and biochemical test results. The antibacterial activity was determined through the Kirby-Bauer disc diffusion test. The results indicated that five selected isolates (i.e., 0A1, 0A2, 0A3, 0A4, and 0A5) had a round shape with flat edges. Isolates 0A2 and 0A4 exhibited a solid white color, whereas isolates 0A1 and 0A3 displayed an off-white color, and isolate 0A5 showed a yellow color. Isolates 0A1, 0A2, and 0A4 were Gram-positive bacteria, while isolates 0A3 and 0A5 demonstrated Gram-negative characteristics. The biochemical testing revealed that the five selected isolates were identified as *Staphylococcus warneri*, *Staphylococcus haemolyticus*, *Enterobacter aerogenes*, *Enterococcus faecium*, and *Escherichia coli*. The ethanol extracts of clove (*Syzygium aromaticum*) leaves, cinnamon (*Cinnamomum burmannii*) bark, and eucalyptus (*Melaleuca leucadendra*) leaves demonstrated the ability to inhibit pathogenic bacteria responsible for UTI. Clove leaf ethanol extract exhibited the highest average clear zone compared to the other two ethanol extracts. This study concluded that medicinal plants from Maluku, Indonesia, the clove, cinnamon, and eucalyptus, have antibacterial potential that may be effective in treating UTI. Further investigation is necessary to fully comprehend the mechanisms of action, detection, and description of bioactive compounds, particularly against human pathogens.

**Keywords:** Antibacterial; human and medicine; infectious disease; medicinal plant; urinary tract infection (UTI)

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**Highlights:**
1. Clove, cinnamon, and eucalyptus have found to be able to providing antibacterial agents for urinary tract infection (UTI).
2. Ethanol extracts derived from clove leaves, cinnamon bark, and eucalyptus leaves can inhibit pathogenic bacteria responsible for UTI, indicating their potential as sources of bioactive compounds for UTI medication development.

## INTRODUCTION

The prevalence of infectious diseases is among the primary contributors to global mortality. Infectious diseases result from infections by bacteria, viruses, fungi, and parasites that multiply in the body (Pangaribuan 2013, Saudi 2018). The World Health Organization (2020) has reported that out of 25 million fatalities worldwide, approximately one-third are attributed to infectious diseases. One of the most common infectious diseases is urinary tract infection (UTI), which is characterized by the presence of around 100,000 bacteria per milliliter of urine. UTI is an infectious disease with an increasing prevalence (Maryani et al. 2021).

It is estimated that there are 222 million UTI sufferers in Indonesia. The number predominantly consists of women, although there is also a notable UTI prevalence in men. The number of patients affected by UTI in Indonesia is 100,000 per year (World Health Organization 2020). According to Rachmawati et al. (2021), UTI is a common medical condition that causes significant morbidity and mortality. UTIs are caused by microorganisms in the urinary tract, including the bladder, prostate, kidneys, and collecting ducts.

The main causes of UTI are Gram-negative or Gram-positive bacteria, fungi, and viruses. A prior study conducted by Ritonga (2018) has identified the primary Gram-negative bacteria responsible for UTI as follows: *Escherichia coli* (48.44%), *Klebsiella pneumonia* (17.19%), *Acinetobacter baumannii* (14.07%), *Proteus mirabilis* (6.25%), *Strenotrophomonas maltophilia* (3.13%), and *Pseudomonas aeruginosa* (3.13%). Gram-positive bacteria *Enterococcus faecalis* (4.69%) and *Staphylococcus haemolyticus* (3.13%) have also been found to cause UTI. Nevertheless, *Escherichia coli* has been recognized as the most prevalent bacterium causing UTI (Rostinawati 2021).

Antibiotics are intended to treat diseases caused by bacterial infections. The action mechanism of antibiotics involves the production of negative effects on the normal flora in the human gut, which can lead to changes in the microflora and cause itching in the pubic area, skin rashes, and nausea. However, prolonged use of antibiotics is associated with the occurrence of antibiotic resistance (Kidane et al. 2019, Kaewkod et al. 2021).

Currently, bacteria responsible for UTI are highly resistant to antibiotics, resulting in a higher mortality rate. Changes in bacterial resistance pattern may lead to a more rapid development of UTI compared to other infections (Muhammad et al. 2018). According to Rohadi et al. (2021), bacteria exhibiting antibiotic resistance are more difficult to treat. Consequently, alternative therapies are required to treat UTI through the use of natural ingredients derived from medicinal plants. Traditional medicinal plants may be a viable alternative to avoid antibiotic resistance, as they have been shown to be effective in treating bacterial infections with minimal side effects. The antimicrobial potential of plants is attributed to their phytochemical content and availability at an affordable price. Previous studies have found that medicinal plants contain secondary metabolites, such as alkaloids, flavonoids, terpenoids, steroids, carotenoids, and other phenolic compounds (Kidane et al. 2019, Mayekar et al. 2021). The 2018 data reported by the World Health Organization (2019) showed that 170 countries, representing 88% of the total members, acknowledge medicinal plants as alternative therapies. Indonesia is a developing country abundant in diverse natural resources, which provides a source of various medicinal plants utilized as raw materials for traditional medicines. There are 40,000 species of plants worldwide, whereas Indonesia is home to 28,000 species of medicinal plants (Daro et al. 2020).

Maluku is a group of islands located in the eastern part of Indonesia. It is known for its rich history as a spice-producing region. The island is renowned for its production of various spices, including nutmeg, cloves, cinnamon, and eucalyptus, which have been traded and used for centuries, not only as a food seasoning but also as a traditional medicine. It has been found that spices contain bioactive compounds. A prior study showed that fourteen secondary metabolites were successfully isolated from clove (*Syzygium aromaticum*) flower bud extract (Kiran et al. 2023). Extracts derived from *Cinnamomum zeylanicum* and *Cinnamomum cassia* (L.) J. Presl barks have been found to contain biologically active compounds, such as eugenol, trans-cinnamaldehyde, and linalool (Błaszczyk et al. 2021). In addition, different phenolic compounds have been identified from the extractions of five *Eucalyptus* species, i.e., *E. maidenii*, *E. robusta*, *E. citriodora*, *E. tereticornis*, and *E. camaldulensis*. The phenolic compounds include hydroquinone, hesperitin, pyrogallol, resorcinol, protocatechuic acid, naringenin, chlorogenic acid, and catechin (Nasr et al. 2019). The local people of Maluku have long recognized the medicinal properties of these spices and incorporated them into their traditional healing practices. They use cloves to relieve pain, nutmegs to treat digestive problems, and maces to remedy headaches. These traditional uses of spices reflect the ingenuity and resourcefulness of Maluku and showcase the importance of plants and natural remedies in traditional medicine. This study aimed to evaluate the antibacterial properties of several medicinal plant ethanol extracts against pathogenic bacterial isolates from urine samples of UTI patients.

## MATERIALS AND METHODS

This research was conducted at the Microbiology Laboratory of the Faculty of Medicine, Universitas Pattimura, Ambon, Indonesia, and the extraction process was carried out at the Basic Chemistry Laboratory, Department of Chemistry of the Faculty of Mathematics and Natural Sciences, Universitas Pattimura, Ambon, Indonesia. This was a true experimental laboratory study with a posttest-only controlled group design (Ma'rufi et al. 2020).

The sample preparation began with obtaining clove (*Syzygium aromaticum*) leaves, cinnamon (*Cinnamomum burmannii*) barks, and eucalyptus

(*Melaleuca leucadendra*) leaves from traditional markets in Ambon, Indonesia. The collected samples were washed, cut into small pieces, and dried at room temperature. They were then dehydrated at 60°C for 24 hours (Jafarzadeh et al. 2020). The samples were then ground using a blender, filtered, and dried.

The extraction was performed using the maceration method. Each sample, weighing 200 mg, was soaked at room temperature for 3 × 24 hours in a 96% ethanol solvent (1:10 w/v) with continuous stirring using a whisk and agitator (Figure 1). After the third day, the maceration results were filtered using filter paper to separate the extract from the residue. The extraction results were then condensed with a rotary evaporator to produce a concentrated extract. The extracts were prepared at different concentrations of 1%, 5%, 25%, 50%, 75%, and 100% through dilution using distilled water (Abubakar & Haque 2020).

Figure 1. Samples soaked in 96% ethanol solvent.

Urine samples were obtained from UTI patients at Dr. Johannes Leimena Central General Hospital, Ambon, Indonesia. The middle portion of the urine was used, yielding the most optimal sample for UTI examination due to it being free of contamination. The samples were collected under aseptic conditions using a sterile syringe. After acquiring the samples, they were subsequently transferred to a sterile urine container and immediately transported to the laboratory (Oros et al. 2020). The isolation was carried out using the dilution method. The colonies were spread onto a petri dish containing nutrient agar (NA) (Oxoid, UK) as the culture medium and then incubated for 24 hours. The growing colonies were differentiated according to their morphological characteristics, including shape, color, and edge. The selected colonies were purified using the streak method on MacConkey Agar (MCA) (HiMedia Laboratories, Mumbai, India) as the culture medium. The isolates were characterized by colony morphology identification, Gram staining, and biochemical testing using the VITEK 2 Compact system (Biomerieux, USA).

Figure 2. Isolation of pathogens from UTI patients' urine samples.

The antibacterial activity was determined using the Kirby-Bauer disk diffusion susceptibility test on Mueller-Hinton agar (MHA) (HiMedia Laboratories, Mumbai, India). Blank paper discs (Blank Disc Macherey Nagel 484000) were soaked in extracts at different concentrations for 15 minutes before being drained until little extract remained on the paper discs. Subsequently, the paper discs were placed on agar media inoculated with pathogenic bacterial isolates and incubated for 24 hours. Clear zones formed following the incubation, and their diameter was measured. This study used distilled water (ONELAB) as a negative control as well as 5 µg of Cefixime (Oxoid, CT06553B) as a positive control (Ebani et al. 2018, Mickymaray & Al Aboody 2019, Ahmed et al. 2021).

## RESULTS

In this study, bacteria were isolated from the urine samples of patients diagnosed with UTI. Five colonies (i.e., 0A1, 0A2, 0A3, 0A4, and 0A5) were selected according to their morphological differences, as shown in Figure 3. The five selected isolates from the urine samples had a round shape with flat edges. Isolates 0A2 and 0A4 exhibited a solid white color, while isolates 0A1 and 0A3 displayed an off-white color. Contrary to the other four isolates, isolate 0A5 showed a yellow color.

The bacterial colonies selected according to their morphology were then identified microscopically by Gram staining. The Gram staining results indicated that the selected isolates consisted of two Gram-negative bacteria and three Gram-positive bacteria. Isolates 0A1, 0A2, and 0A4 were classified as Gram-positive, whereas isolates 0A3 and 0A5 were identified as Gram-negative. The cells of the five selected isolates exhibited coccus and bacillus morphology. As detailed in Table 1, isolates 0A1 and 0A2 had a coccus shape, whereas 0A3, 0A4, and 0A5 presented a bacillus shape.

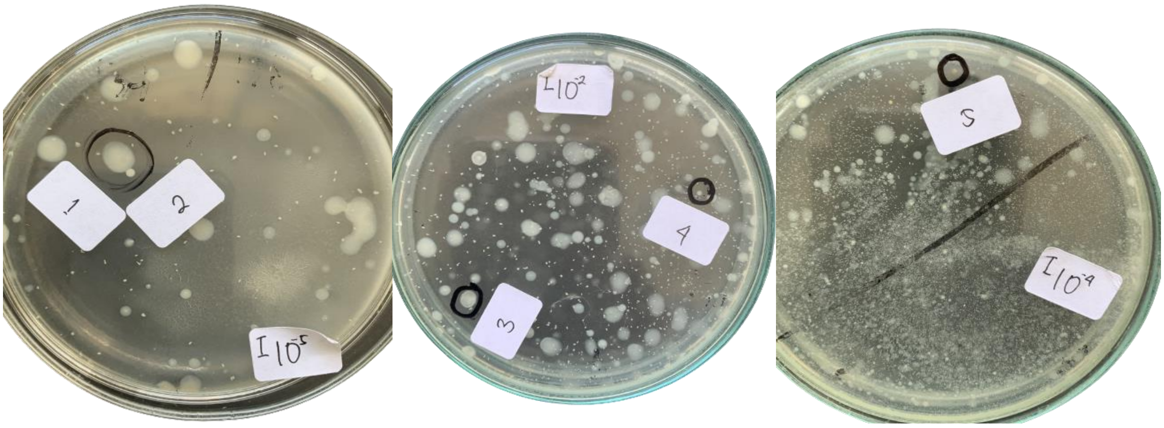


Figure 3. Bacterial colonies selected from the isolation of urine samples obtained from UTI patients.

Table 1. Macroscopic and microscopic identification of bacterial isolates from the urine samples.

| Isolates | Shape | Color | Edge | Cell shape | Gram |
|---|---|---|---|---|---|
| 0A1 | Round | Off-white | Flat | Coccus | Positive |
| 0A2 | Round | Solid White | Flat | Coccus | Positive |
| 0A3 | Round | Off-white | Flat | Basil | Negative |
| 0A4 | Round | Solid White | Flat | Coccus | Positive |
| 0A5 | Round | Yellow | Flat | Basil | Negative |

The identification process continued with the use of the VITEK 2 Compact system. The results indicated that the five selected isolates from the urine samples comprised *Staphylococcus warneri*, *Staphylococcus haemolyticus*, *Enterobacter aerogenes*, *Enterococcus faecium*, and *Escherichia coli* (Table 2).

Table 2. Results of the bacterial identification using the bioMérieux VITEK 2 Compact system.

| Isolates | Identification results | Level of confidence |
|---|---|---|
| 0A1 | *Staphylococcus warneri* | 94% |
| 0A2 | *Staphylococcus haemolyticus* | 99% |
| 0A3 | *Enterobacter aerogenes* | 99% |
| 0A4 | *Enterococcus faecium* | 86% |
| 0A5 | *Escherichia coli* | 98% |

Five species of isolated uropathogenic bacteria were tested to determine the antibacterial activity of the ethanol extracts of clove (*Syzygium aromaticum*) leaves, cinnamon (*Cinnamomum burmannii*) barks, and eucalyptus (*Melaleuca leucadendra*) leaves. The results indicated that the extracts had the ability to inhibit the growth of the test bacteria. This was evident by the formation of inhibition zones at all concentrations of the ethanol extracts used in this study.

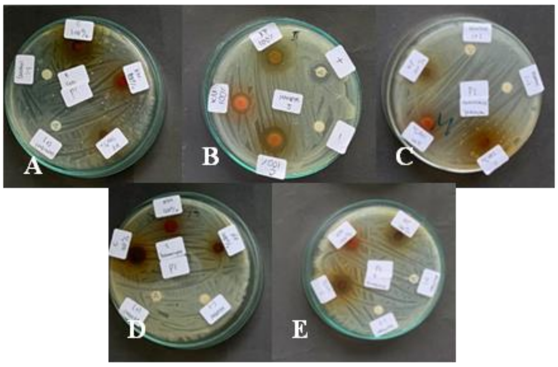


Figure 4. Inhibition zones of clove, cinnamon, and eucalyptus ethanol extracts against UTI bacterial isolates (100% concentration as the representative). Legends: (A) *E. coli*; (B) *S. warneri*; (C) *E. faecium*; (D) *S. haemolyticus*; (E) *E. aerogenes*.

Clove extract showed inhibition zone diameters ranging from 2 to 14 mm. Its inhibitory activity started at a concentration of 1%, indicating that the antimicrobial action of clove extract began at low concentrations. The inhibition zone diameters for cinnamon extract ranged from 1.25 to 9.75 mm. Different from clove extract, cinnamon extract exhibited inhibitory activity that started at a concentration of 1% for *Enterococcus faecium* and

*Escherichia coli* and at 5% for *Enterobacter aerogenes, Staphylococcus warneri,* and *Staphylococcus haemolyticus*

Table 3. Zones of inhibition produced by the medicinal plant extracts.

| Isolates | Medicinal plant extracts | Diameters (mm) of inhibition zones at varying concentrations | | | | | |
|---|---|---|---|---|---|---|---|
| | | 1% | 5% | 25% | 50% | 75% | 100% |
| *E. aerogenes* | Clove | 2 | 4.5 | 6 | 7.75 | 9.25 | 10.5 |
| | Cinnamon | 0 | 1.5 | 4.25 | 5.5 | 6.25 | 6.75 |
| | Eucalyptus | 4 | 4.5 | 5.75 | 6.25 | 8 | 8.75 |
| | Control (+) | 0 | 0 | 0 | 0 | 0 | 0 |
| | Control (-) | 0 | 0 | 0 | 0 | 0 | 0 |
| *E. faecium* | Clove | 3.5 | 6.75 | 8.75 | 10.75 | 11.5 | 13 |
| | Cinnamon | 2.75 | 5.25 | 7 | 8.25 | 8.5 | 9.75 |
| | Eucalyptus | 3.25 | 4.75 | 6.25 | 6.75 | 7.75 | 8.75 |
| | Control (+) | 0 | 0 | 0 | 0 | 0 | 0 |
| | Control (-) | 0 | 0 | 0 | 0 | 0 | 0 |
| *S. warneri* | Clove | 6 | 7.75 | 14.25 | 19.75 | 12 | 12 |
| | Cinnamon | 0 | 5.25 | 8 | 9 | 8 | 8.75 |
| | Eucalyptus | 5.5 | 4 | 9.25 | 14.5 | 6.5 | 11.5 |
| | Control (+) | 9.75 | 10.75 | 11 | 9.75 | 7.5 | 8.5 |
| | Control (-) | 0 | 0 | 0 | 0 | 0 | 0 |
| *S. haemolyticus* | Clove | 4 | 5.75 | 6.75 | 7.5 | 8.5 | 9.5 |
| | Cinnamon | 0 | 3.25 | 4.75 | 6 | 7.25 | 8.25 |
| | Eucalyptus | 5.5 | 4.5 | 7 | 8 | 8.5 | 9.25 |
| | Control (+) | 0 | 0 | 0 | 0 | 0 | 0 |
| | Control (-) | 0 | 0 | 0 | 0 | 0 | 0 |
| *E. coli* | Clove | 2.75 | 4 | 7.75 | 11.5 | 13.5 | 14 |
| | Cinnamon | 1.25 | 2.5 | 5.5 | 6.75 | 8.5 | 8.75 |
| | Eucalyptus | 5.25 | 4 | 6.75 | 8.5 | 11.5 | 9.75 |
| | Control (+) | 0 | 0 | 0 | 0 | 0 | 0 |
| | Control (-) | 0 | 0 | 0 | 0 | 0 | 0 |

The inhibition zone diameters for eucalyptus extract varied between 3.25 and 11.5 mm. The inhibitory activity commenced at a concentration of 1%, indicating that the antimicrobial action also initiated at low concentrations. In comparison to the positive control, *Enterobacter aerogenes, Enterococcus faecium, Escherichia coli*, and *Staphylococcus haemolyticus* were resistant to cefixime, while only *Staphylococcus warneri* demonstrated sensitivity to the medication.

## DISCUSSION

Bacteria are the source of over 85% of infections, although viruses and yeast can also induce UTIs. In this study, the isolated colonies were round, white, and had flat edges. Some of the cells were in the shape of cocci and rods, with certain cells classified as Gram-positive bacteria and others as Gram-negative bacteria. The biochemical identification results indicated that the isolated bacterial colonies were *Staphylococcus wareri, Staphylococcus haemolyticus, Enterobacter aerogenes, Enterococcus faecium*, and *Escherichia coli*. It has been recognized that *Escherichia coli, Staphylococcus aureus, Enterococcus faecalis, Klebsiella pneumoniae, Pseudomonas aeruginosa, Proteus mirabilis, Acinetobacter baumannii*, and *Staphylococcus saprophyticus* are among the bacteria most frequently linked to UTI. A UTI is deemed established when the pathogen enters the urinary tract and multiplies to exceed 105 colonies/mL in the urine (Karam et al. 2019). The two most common bacteria found in UTIs are *Escherichia coli* and *Enterococcus* species. Due to the presence of both bacteria in the gastrointestinal environment, resistance genes can be acquired from other commensal species more easily (Noreen et al. 2018).

Prakash & Saxena (2013) revealed their findings on the prevalence of Gram-negative and Gram-positive bacteria in UTIs. It was determined that Gram-negative bacteria (90.32%) have a greater prevalence than Gram-positive bacteria (9.68%). Mickymaray & Al Aboody (2019) conducted a study on the isolation and identification of pathogenic bacteria from urine samples. Their analysis revealed that among 11 confirmed pathogenic bacteria, there were two Gram-positive and nine Gram-negative bacteria. In previous research carried out by Odoki et al. (2019), most cases of UTI were caused by bacteria. *Escherichia coli* and *Enterococcus* spp. were the two predominant bacteria attributable to these cases. Another study conducted by Thiyagarajan & John (2020) on UTI cases indicated that the infections were predominantly caused by bacteria and fungi. The three primary bacteria responsible for the disease were *Staphylococcus* sp., *Enterobacter* sp., and *Escherichia coli*. This is also in line with a prior study using urine culture, which identified *Staphylococcus haemolyticus* and *Escherichia coli* as the most common bacteria associated with UTI (Malau & Adipireno 2019).

The use of plants with medicinal properties for the treatment and prevention of various illnesses, including UTIs, is an old tradition. Nevertheless, herbal remedies have recently gained popularity and reliability due to their easy availability, minimal reported side effects, cost-effectiveness, compatibility with UTI patients, and lack of bacterial resistance (Shaheen et al. 2019). This study demonstrated that the lack of bacterial growth around the disks might be an indirect indicator of the compounds' ability to inhibit the pathogen. The ethanol extracts of clove leaves, cinnamon barks, and eucalyptus leaves could inhibit pathogen growth due to their phytochemical content, which plants mainly produce to protect themselves from pathogens. Previous research on the antimicrobial effect of medicinal spice plants identified the presence of secondary metabolites, such as alkaloids, flavonoids, tannin saponins, polyphenols, alpha-terpineol, cineol, and steroids, according to the phytochemical screening results (Mayekar et al. 2021, Khatri et al. 2023). This study revealed variations in the antibacterial activity of different plant extracts. This might result from differences in their chemical composition and the mechanisms of action of their bioactive constituents. Saponins exhibit antimicrobial activity that can cause the leakage of proteins and enzymes from the cells. Alkaloids have an antibacterial effect due to their ability to interact with bacterial deoxyribonucleic acid and interfere with cell division, whereas flavonoids and steroids can cause cell membrane leakage.

Although the precise way in which herbal remedies are used to treat UTIs is still not completely understood, it has been established that plant constituents and secondary metabolites can serve as diuretics, antioxidants, immunomodulators, and antimicrobials. As antimicrobials, plant extracts may prevent pathogens from attaching to the urinary tract and hinder the growth of microorganisms (Shaheen et al. 2019). The effects of medicinal plants result from the various phytochemical constituents of their secondary metabolites. The bacteriostatic and bactericidal effects of plant extracts are attributed to phytochemicals that function in one of two ways. They either function in the same way as conventional antibiotics by blocking the synthesis of the bacterial wall, affecting membrane cells, inhibiting nucleic acid synthesis, protein synthesis, or folate metabolism, or by inhibiting efflux pumps (Khosravani et al. 2020).

In previous research undertaken by Loose et al. (2020), it was found that the essential oils of cajuput, lemongrass, tea tree, and thyme can exert an antibacterial activity against all tested uropathogenic bacterial strains, with differing levels of susceptibility among individual species. Thyme and tea tree essential oils show a higher effectivity against the tested bacteria compared to lemongrass and cajuput oils. It can be inferred that linalool and terpinen-4-ol, the main components present in thyme and tea trees, are more potent against uropathogenic bacteria than 1,8-cineole and neral/geranial found in cajuput and lemongrass, respectively. Cinnamyl acetate, coumarin, eugenol, eucalyptol, trans-cinnamaldehyde, L-borneol, caryophyllene oxide, benzoic acid, linalool, caffeic acid, and camphor are bioactive compounds discovered in the fruits, leaves, and barks of cinnamon trees (Rao & Gan 2014, Vasconcelos et al. 2018, Błaszczyk et al. 2021). Moreover, cinnamon (*Cinnamomum zeylanicum*) bark oil has demonstrated both inhibitory and bactericidal properties against *Pseudomonas aeruginosa*, including multidrug-resistant strains associated with UTIs. An ethanolic extract derived from *Cinnamomum zeylanicum* has been found to potentially limit the growth of bacterial colonies extracted from urine samples of UTI patients. The bacterial colonies include *E. coli*, *P. aeruginosa*, and *K. pneumoniae* (Utchariyakiat et al. 2016, Dhore & Jha 2019). Additionally, Narayanan et al. (2017) demonstrated that an aqueous extract of *Cinnamomum zeylanicum*, at a concentration of 0.1 g/mL, has antimicrobial activity against pathogens responsible for UTIs. *E. coli*, *K. pneumoniae*, *S. aureus*, *Enterobacter* spp., P. *aeruginosa*, *S. typhi*, and *S. flexneri* were among the affected pathogens.

Previous studies, such as those conducted by Băicuș et al. (2022), Nuñez & D'Aquino (2012), and Mytle et al. (2006), showed that clove has been widely utilized for the treatment of UTIs because of its broad-spectrum antimicrobial effects against both Gram-positive and Gram-negative bacteria. Clove oil has demonstrated great efficacy in combating bacteria responsible for UTIs. It was reported that *E. coli* and *S. aureus* populations can be reduced by 99.999% and 99.9999%, respectively, after merely 8 hours of treatment using clove oil (Cui et al. 2015). Swamy et al. (2016) carried out a study to compare clove oil with cinnamon, bell pepper, thyme, oregano, and rosemary oils. Their analysis revealed that clove oil is the most effective in inhibiting the growth of *S. typhi*, *S. aureus*, and *P. aeruginosa*.

**Strength and limitations**

This research is an initial screening in the development of bioactive compounds derived from endemic medicinal plants of the Maluku Islands. Specifically, these medicinal plants are anticipated to be used as antibacterials against pathogenic bacteria responsible for urinary tract infections, many of which exhibit antibiotic resistance. However, no further tests were carried out in this study to determine the types of bioactive compounds contained in the medicinal plant samples.

## CONCLUSION

Ethanol extracts derived from clove (*Syzygium aromaticum*) leaves, cinnamon (*Cinnamomum burmannii*) barks, and eucalyptus (*Melaleuca leucadendra*) leaves can inhibit pathogenic bacteria responsible for urinary tract infections, including *Staphylococcus warneri*, *Staphylococcus haemolyticus*, *Enterobacter aerogenes*, *Enterococcus faecium*, and *Escherichia coli*. Clove leaf ethanol extract has the highest inhibitory activity compared to cinnamon bark and eucalyptus leaf extracts, as evident by its average clear zone. Thus, it can be concluded that cloves are the most effective at inhibiting bacterial growth. Further investigation is necessary to fully comprehend the mechanisms of action, detection, and characterization of bioactive compounds, particularly their antibacterial efficacy against human pathogens.

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**Conflict of interest**

None.

**Ethical consideration**

The Ethics Committee of the Faculty of Medicine, Universitas Pattimura, Ambon, Indonesia, issued the ethical approval for this study under recommendation No. 034/FK-KOM.ETIK/VIII/ 2022 dated 15/3/2022.

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**Author contribution**

EA contributed to the conception and design, analysis and interpretation of the data, drafting of the article, and final approval. VDL contributed to the drafting of the article, the provision of administrative, technical, and logistic support, and the collection and assembly of the data. SCA drafted the article, critically revised the article for important intellectual content, as well collected and assembled the data.

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