

Cinnamend: Evaluating the Use of Kaningag (*Cinnamomum Mindanaense* Elm.) Leaf Extract as an Active Ingredient in Antibacterial Ointment against *Escherichia Coli*

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DOI: <https://doi.org/10.51244/IJRSI.2025.12040136>

Received: 08 April 2025; Accepted: 15 April 2025; Published: 21 May 2025

ABSTRACT

Advancements in wound care offer practical solutions, but the rise of antibiotic-resistant bacteria like *Escherichia coli* poses a growing threat, demanding urgent alternative approaches to infection management. This study aimed to evaluate the antibacterial effect of kaningag (*Cinnamomum mindanaense* Elm.) leaf extract as an ointment against *E. coli* and to compare its effectiveness across different concentrations (2.5%, 5%, 7.5%, and 10%) with a control treatment. A posttest-only experimental design was employed, where treatments were applied to Petri dishes inoculated with *E. coli*. Results showed that the 10% concentration (T4) exhibited significantly greater antibacterial activity (31 mm) compared to the 2.5% concentration (T1) and negative control (petroleum jelly). T4's effectiveness was comparable to the positive control (terramycin) at 20 mm. Statistical analysis revealed no significant differences among the other concentrations (T1, T2, T3), suggesting similar antibacterial effects. These findings indicate that kaningag leaf extract, particularly at higher concentrations, shows promise as a natural antibacterial agent. Given the increasing resistance of *E. coli* to conventional antibiotics, kaningag represents a potential alternative treatment, warranting further investigation for broader therapeutic applications.

Keywords: *Cinnamomum mindanaense*, antibacterial effectiveness, antibacterial ointment, antimicrobial resistance, *Escherichia coli*

INTRODUCTION

Advancements in wound care have led to more convenient, cost-effective, and practical prevention strategies. At the same time, the rise of antibiotic resistance is increasingly undermining effective infection management, creating new challenges in treatment. Bacteria like *Escherichia coli* are increasingly developing antibiotic resistance, causing both common and severe infections. This escalating resistance is a critical threat to healthcare, demanding urgent action and alternative approaches to wound infection management.

One promising alternative approach involves using medicinal plants such as kaningag (*Cinnamomum mindanaense* Elm.), an endemic tree species in the Philippines (Magleo et al., 2017). Traditionally utilized by Indigenous communities for its therapeutic properties, kaningag contains bioactive compounds like cinnamaldehyde and eugenol, known for their antimicrobial, anti-inflammatory, and antioxidant effects (Sharma et al., 2016). While specific studies on *C. mindanaense* are limited, research on related species within the *Cinnamomum* genus has demonstrated significant antibacterial activity against various pathogens, including *E. coli*. For instance, essential oils from *Cinnamomum zeylanicum* have inhibitory effects on colistin-resistant gram-negative bacteria, with cinnamaldehyde being the major active compound (Selma et al., 2024).

The prevalence of antimicrobial-resistant *E. coli* and other bacterial infections significantly threatens healthcare systems and public safety worldwide. In 2019, infections from 33 different types of bacteria, including *E. coli*, caused 7.7 million deaths globally, accounting for 1 in 8 deaths (Nuffield Department of Medicine, 2022). While *E. coli* remains a leading cause of multidrug-resistant (MDR) infections, other

resistant bacteria, such as *Staphylococcus aureus* and *Pseudomonas aeruginosa*, also contribute to the growing antimicrobial resistance (AMR) crisis (WHO, 2018). Infections in developed and developing countries are becoming harder to treat, leading to prolonged hospital stays, increased healthcare costs, and higher mortality rates (Galindo-Mendez, 2020; Mueller & Tainter, 2023).

The growing threat of AMR, particularly concerning *E. coli*, highlights the need for innovative antibacterial agents. *C. mindanaense* has shown promise in laboratory studies as an antimicrobial agent, inhibiting Gram-positive and Gram-negative bacteria, including *E. coli* (Selma et al., 2024). In one study, *C. mindanaense* leaf extract exhibited significant antibacterial activity, with an effective inhibition zone that rivals synthetic antibiotics (Magleo et al., 2017). Its phytochemicals, such as alkaloids and saponins, are believed to disrupt bacterial cell membrane integrity, an essential mechanism in combating resistant bacterial strains (Alexander et al., 2017).

To combat AMR, innovative antibacterial solutions are being explored alongside conventional antibiotics. Antimicrobial ointments containing silver sulfadiazine, mupirocin, or neomycin are commonly used for wound infections, particularly in regions with limited access to advanced medical care. However, the effectiveness of these treatments has diminished due to the rise of resistant bacterial strains (Aijaz et al., 2023). Global researchers are investigating alternative therapies such as bacteriophage treatments, which use viruses to target and destroy specific bacteria. Additionally, antimicrobial peptides (AMPs) derived from natural sources show promise in combating resistant bacteria by disrupting their membranes (Monk et al., 2024; Tacconell et al., 2017). These emerging therapies offer hope in the fight against resistant infections while reducing the reliance on traditional antibiotics.

In the search for alternatives, kaningag (*C. mindanaense* Elm.) has attracted attention for its antibacterial properties. Extracts of kaningag, particularly its essential oils, have shown efficacy against several strains of *E. coli*, including multidrug-resistant variants (Vasconcelos et al., 2020). Research indicates that kaningag essential oil, when applied topically, can inhibit bacterial growth by disrupting cell wall synthesis and membrane permeability, leading to bacterial cell death (Ju et al., 2023). Furthermore, its anti-inflammatory effects make it an ideal candidate for wound care formulations to treat infections caused by resistant bacteria, such as *E. coli* (He et al., 2022).

Global efforts to address AMR require coordinated actions across sectors. Enhanced surveillance systems, stricter regulations on antibiotic use in agriculture, and developing novel therapeutics are essential to slow the spread of resistance (Samreen et al., 2021; Pabst, 2023). Strengthening infection prevention measures, investing in research for new antibacterial solutions, and promoting global antimicrobial stewardship programs can mitigate the impact of resistant bacteria. In connection, a collaborative approach involving governments, healthcare providers, researchers, and the pharmaceutical industry is necessary to combat the escalating threat of AMR.

Wound infections occur when harmful microorganisms invade and proliferate at the wound site, which prevents routine healing. Common pathogens include *S. aureus*, particularly the methicillin-resistant variety, or MRSA (Maheswary et al., 2021). Correspondingly, ASEAN countries are experiencing a rise in AMR and the emergence of multidrug-resistant *E. coli* strains. According to San et al. (2021), the high risk of AMR in the region is driven by factors such as high population density, disease burden, and increased antibiotic use. Kaningag (*C. mindanaense* Elm.) presents a natural alternative to combat the challenges AMR poses in the Philippines (Picardal & Agoo, 2019; Cruz, 2017). In addition to its antibacterial properties, it has shown promise as a low-cost treatment option, especially in rural communities with limited access to healthcare. Studies conducted in various regions of the Philippines found that kaningag extracts not only inhibited the growth of *E. coli* but also demonstrated cytotoxic effects against biofilm-forming bacteria, which are often more resistant to conventional antibiotics (Wang et al., 2020; Didehdar et al., 2022). These properties make kaningag a potentially effective remedy in the fight against AMR, contributing to efforts to alleviate the strain on the healthcare system caused by resistant bacterial infections (Vicente & Castrejón, 2020). Among ASEAN nations, Myanmar has the highest prevalence of AMR, whereas Brunei has the lowest (Rosero et al., 2021). Furthermore, a study by Tauk et al. (2022) found a 35% (80 isolates) prevalence of bacterial infections in

Indonesian open fracture cases, with 62.5% (50 isolates) being gram-positive and 37.5% (30 isolates) gram-negative.

Recent studies highlighted the growing prevalence of multidrug-resistant (MDR) *E. coli* strains. A study analyzing 15,647 wound samples from five continents: Africa, Europe, the USA, Australia, and Asia found that 59% contained Gram-negative bacteria, with *E. coli* (17%), *P. aeruginosa* (11%), and *Klebsiella pneumoniae* (11%), all exhibiting high rates of multidrug resistance (Chelkeba et al., 2021). Similarly, two studies reported that among 120 samples, 19 (27.94%) *E. coli* isolates demonstrated 100% resistance to vancomycin and tetracycline. In contrast, an analysis of 350 samples identified 65 *E. coli* isolates with 90.91% resistance to amikacin, 81.82% to imipenem, and 45.45% to colistin (Alhlale et al., 2020; Banik & Shamsuzzaman, 2021).

Additionally, the increase in antibiotic-resistant infections presented a significant public health challenge. Child mortality in India occurs approximately every nine minutes due to antibiotic-resistant bacterial infections, with over 50,000 infants at risk of dying from sepsis as common treatments become ineffective (Gandra et al., 2018; McDonnell & Klemperer, 2022; Subramaniam & Girish, 2020). Biotechnology advancements have developed plant-based creams and topical agents with antibacterial, anti-inflammatory, and antioxidant properties as viable alternatives that can disrupt biofilms and promote faster wound healing (Proshina et al., 2020).

The *Cinnamomum* genus, including *C. mindanaense*, has been widely studied for its antimicrobial potential. For example, *Cinnamomum zeylanicum* essential oil demonstrated vigorous antibacterial activity against multidrug-resistant *E. coli*, particularly due to cinnamaldehyde, a compound also found in other *Cinnamomum* species like *mindanaense*. These essential oils damage bacterial cell membranes and interfere with energy metabolism, making them effective against antibiotic-resistant strains (Selma et al., 2024).

In the national context, the cases of antibiotic resistance in the Philippines have led to severe public health challenges, such as an increase in mortality rates. In 2019, AMR contributed to 56,700 fatalities and directly caused 15,700 deaths (Institute for Health Metrics and Evaluation, 2022). The Global Research on Antimicrobial Resistance (GRAM) study ranked the Philippines 77th out of 204 countries regarding age-standardized mortality rates per 100,000 population associated with AMR (Calpito, 2022). AMR is a significant health threat in the Philippines, causing more deaths than chronic respiratory, digestive, maternal, and neonatal disorders. It also surpassed fatalities from self-harm, interpersonal violence, and other non-communicable diseases (Global Health Data Exchange [GHDEx], 2024). The study by Abdon et al. (2024) on developing two ointments from corn silk and rice hull conferred an answer to the search for alternative medicine against bacteria, including *E. coli*. The results showed that the positive control, amoxicillin, has a larger mean zone of inhibition than the ointments. This alarming trend highlighted the urgent need for stronger antibiotic stewardship and public health interventions to combat antimicrobial resistance.

In Davao City, Palicte (2021) noted that the issue of *E. coli* contamination is a serious public issue. The recent diarrhea outbreaks have been associated with food and water sources contaminated with bacteria, specifically *E. coli*, that have caused countless food-related illnesses (Monteverde, 2021). The bacteria were said to be identified as one of the pathogens determined in a significant diarrhea outbreak in 2022 associated with ingesting contaminated street food (Cantal-Albasin, 2022). Chronic diarrhea can lead to the development of anal fissures, which are susceptible to infection (NHS, 2024). This issue poses a significant risk to public health, emphasizing the need for better monitoring and management of food and water safety to prevent contamination. Antibiotics are used in wound infections to deal with bacteria. Their use prevents the spread and reduces risks like sepsis, delayed healing, or chronic infection. However, broad-spectrum antibiotics have increased antibiotic resistance. Finding alternative antibacterial agents, like kaningag leaf extract, is crucial. Kaningag (*C. mindanaense* Elm.) leaf extract has been tested as an alternative antibacterial agent. A study on antimicrobial activity revealed that the extract inhibited *S. aureus* but showed no effect against *E. coli* (Agton et al., 2019). This highlights the importance of further research to optimize the extract's concentration, explore its effects on other microorganisms, and identify the responsible antimicrobial compounds. In the context of rising antibiotic resistance and *E. coli* contamination, investigating kaningag as an alternative offers potential for addressing these public health concerns. This comes when antibiotic resistance increases worldwide, and rural people lack access to cheap antibacterial products.

Moreover, further investigation into the properties of plants such as kaningag is necessary to explore alternatives for addressing this issue. Numerous studies have utilized various types of this plant to treat infectious diseases caused by pathogenic microorganisms, which anchors the established concepts of Germ Theory (1861) by Louis Pasteur and Robert Koch. This theory provides a foundational understanding of how kaningag leaf extract functions as an antibacterial agent against *E. coli*. Furthermore, this investigation aligns with Moerman's Non-Random Medicinal Plant Selection Theory (1979), which posits that medicinal plants are chosen based on their therapeutic efficacy, rather than randomly (Kutal et al., 2021). This theory highlights indigenous communities' systematic, knowledge-based approach in selecting kaningag for its antibacterial properties. Based on its observed therapeutic effects, this knowledge, passed down through generations, has guided the use of kaningag leaf extract in developing antibacterial ointments, reflecting a non-random, culturally informed plant selection. Thus, this study aimed to evaluate the use of kaningag (*C. mindanaense* Elm.) leaf extract as an active ingredient in antibacterial ointment for combating *E. coli* and its antibiotic resistance.

Statement of the Problem

This study aimed to test the antibacterial effect of the kaningag (*C. mindanaense* Elm.) leaf extract against *E. coli* as an ointment.

Specifically, the study aimed to answer the following questions:

- 1.) What is the antibacterial effectiveness in terms of zone of inhibition (ZOI) of the different concentrations of kaningag (*C. mindanaense* Elm.) leaf extract against *E. coli* in the following treatments:
 - 1.1 2.5 % concentration of kaningag leaf extract;
 - 1.2 5.0 % concentration of kaningag leaf extract;
 - 1.3 7.5 % concentration of kaningag leaf extract; and
 - 1.4 10 % concentration of kaningag leaf extract?
- 2.) What is the antibacterial effectiveness in terms of ZOI of the following control treatments against *E. coli*:
 - 2.1 commercial topical antimicrobial ointment (positive control); and
 - 2.2 ointment base (negative control)?
- 3.) Are there significant differences in the antibacterial effectiveness of *E. coli* under varied concentrations of kaningag (*C. mindanaense* Elm.) leaf extract and the control groups?

Hypothesis

To objectively address the problem listed in the preceding section, the given null hypothesis was formulated:

H₀: There is no significant difference in the antibacterial effect against *E. coli* at different concentrations of kaningag (*C. mindanaense* Elm.) leaf extract and the control groups.

Significance of the Study

Researchers have found that kaningag (*C. mindanaense* Elm.) contains bioactive phytochemicals that exhibit antibacterial activity. The significant antibacterial and wound-healing properties may enhance healing and prevent infections. This unique approach allows the researchers to investigate natural remedies as alternatives for treating antibacterial infections, which may differ from previous studies that focused on different formulations or ingredients. Hence, this study would benefit the following:

Department of Health Officials. This research could provide the Department of Health (DOH) with essential benefits by thoroughly investigating the use of kaningag leaf extract as an antibacterial medication. By gaining insights, healthcare professionals can improve wound treatment methods, making them more effective, enhancing patient recovery, and contributing to better treatment strategies.

Department of Science and Technology. This research could allow the Department of Science and Technology (DOST) to explore kaningag (*C. mindanaense* Elm.) as a natural antibacterial agent. By examining its bioactive properties, the DOST could support the development of cost-effective, sustainable alternatives to synthetic medicines, aligning with its goal of advancing local, eco-friendly healthcare solutions.

Pharmaceutical Companies. Companies could investigate this study commercially by developing a thorough range of antibacterial products using this natural leaf extract. By utilizing the antibacterial properties of these extracts, companies could innovate and create an eco-friendly alternative to the traditional manufactured chemicals.

Healthcare Professionals. Medical practitioners and pharmacists could discover new solutions for treating antibacterial infections, particularly in regions where traditional medicine is preferred. By cultivating these natural remedies, they could provide affordable healthcare options and promote a more holistic approach by formulating this plant-based product.

Local Communities. Developing this extract into a viable product could help local areas where kaningag is abundant at a low cost, making it a viable source of income for the locals. It is also a form of supporting traditional and local herbal medicine practices, where it is valued and integrated as a potential cure.

Patients. Treatments developed from the natural leaf extract could provide alternative remedies for patients with antibiotic-resistant bacterial strains. The results of this study could provide baseline data for the development of treatments for the said patients.

Future Researchers. The antibacterial properties of kaningag leaf extract could provide researchers with new approaches and ideas, leading to future research and development. Moreover, this natural resource could inspire future researchers to delve into more local herbal medicine and progress toward a potential treatment.

Scope and Limitations

This study mainly evaluated the antibacterial potential of kaningag (*C. mindanaense* Elm.) leaf extract as the active ingredient in antibacterial ointment at different concentrations against *E. coli*. The dried kaningag leaves were sourced within the Davao Region. The research was conducted during the first and second semesters of the academic year 2024-2025 in laboratories within Davao del Sur. The antibacterial efficacy of the kaningag leaves was evaluated using a standardized agar diffusion assay, wherein *E. coli* was cultured on Mueller-Hinton (MH) agar plates at 37 °C. After 22 hours, the resulting zone of inhibition was measured in millimeters.

Nonetheless, this study had some limitations. First, it only employed the extracts of kaningag (*C. mindanaense* Elm.) leaves and their antibacterial effect against *E. coli*. No other extraction methods or solvents were utilized. Second, the study was limited to *E. coli*, and thus, other forms of bacterial species were not used. Methodologically, utilizing laboratory materials and equipment was also a constraint since MH agar plates were explicitly used in the study. Additionally, the geographical area was restricted to the Davao region for the procurement of kaningag leaves, which may impact the generalizability of the results to samples from other areas.

Definition of Terms

The terms below are defined conceptually and operationally as applied in this study.

Active Ingredient. This refers to a combination of various ingredients used to diagnose, cure, alleviate, and treat diseases. These can be natural or synthetic chemical compounds typically found in therapeutic and veterinary medications (Kumar et al., 2022). In the context of this study, the active ingredient is kaningag leaf extract, which possesses properties expected to exhibit an antibacterial effect against *E. coli* that make the antibacterial ointment work.

Antibacterial Ointment. This is used to prevent or treat infections and contains medication that kills bacteria or prevents their growth. This ointment is commonly applied to cuts, burns, or other minor wounds to aid

healing and reduce the risk of infection (Huber, 2024). In this study, the ointment contains kaningag leaf extract as the main active ingredient, combined with an ointment base. Its effectiveness was assessed based on the zone of inhibition against *E. coli* bacteria.

Escherichia coli. This refers to a species of bacteria commonly found in the intestines of humans and animals. While most strains of *E. coli* are harmless, certain types of *E. coli* are pathogenic and can cause infections, including urinary tract infections, gastroenteritis, and food poisoning. In this study, *E. coli* was used as the target bacteria to evaluate the antibacterial efficacy of kaningag (*C. mindanaense* Elm.) leaf extract in an ointment form.

Kaningag (*Cinnamomum mindanaense* Elm.) Leaf Extract. This refers to the leaf extracted from kaningag, which is widely used in food, spices, and herbal remedies. Its bioactive compounds offer various pharmacological benefits, such as antibacterial, anti-inflammatory, and antioxidant (Sharifi-Rad et al., 2021). In this study, the kaningag leaf extract was evaluated for its antibacterial activity against *E. coli* bacteria in the form of an ointment.

Zone of Inhibition. This refers to the clear area around an antibiotic or antimicrobial agent applied to an agar plate, where bacterial growth is inhibited. The size of the zone is measured to determine the effectiveness of the agent against the bacteria. In this study, the zone of inhibition was used to assess the antibacterial activity of kaningag leaf extract against *E. coli*.

METHODS

This chapter encompassed the methodologies employed in carrying out the study. It covered aspects such as research design, the subject of the study, sampling procedure, data gathering procedure, measures, analysis and interpretation, and ethical considerations.

Research Design

In this study, the researchers employed the quantitative true experimental method, specifically the posttest-only design. The outcome of interest was assessed only once, after the intervention, allowing for the evaluation of the effect of the treatment (Choueiry, 2021). This design involved manipulating an independent variable or applying a treatment and then measuring the dependent variable once the treatment had been administered (Krishnan, 2019). Consequently, establishing a cause-and-effect relationship among the different variables was considered a true-experimental design.

Furthermore, the investigation aimed to determine whether *Cinnamomum mindanaense* can be used as an active ingredient in an antibacterial ointment that can work against *Escherichia coli*, a bacterium found in wounds. Hence, the study employed a posttest-only design. According to Krosel et al. (2022), subjects underwent a specific intervention or treatment in this research design, with their outcomes measured only after the intervention. This approach enabled researchers to assess alterations in bacterial growth after exposure to the plant without the need for a baseline measurement. Thus, the posttest-only design was chosen for this study to evaluate bacterial growth in samples exposed to different concentrations of the plant extract, making it the most suitable design for this investigation.

Subject of the Study

This study was focused on investigating the subject, a gram-negative bacterium, *E. coli*. They are predominantly benign, often aiding food digestion, synthesizing vitamins, and defending humans from harmful microorganisms. However, in some cases, it can cause diarrhea, infections, and other kinds of sickness (Centers for Disease Control and Prevention [CDC], 2022).

In this study, *E. coli* strains were procured in a saline solution from a private laboratory in Digos City, Philippines. *E. coli* bacteria are predominantly found in the intestines of humans and warm-blooded animals, where they are typically harmless and aid digestion (World Health Organization [WHO], 2018). However,

pathogenic strains can cause illnesses through contaminated food, water, or contact with animals or infected individuals. Common sources of infection include undercooked ground beef, raw milk, contaminated vegetables, and untreated water (WHO, 2018). Transmission occurs via the fecal-oral route, emphasizing the importance of proper hygiene and food safety practices (CDC, 2022).

Sampling Technique

This study utilized a completely random design for selecting subjects in the experiment. A Complete Random Design (CRD) involved choosing bacterial samples without systematic bias. By doing so, researchers were able to guarantee that the subset of bacteria used for examination or analysis was neutral and representative. According to DiCiaccio (2023), complete random design was frequently employed by researchers as a method for observation or data collection. The variables in the series exhibited equal distributions. Complete Random Design functioned as a method for drawing statistical inferences about a population, playing an essential role in upholding strong internal validity by using randomization to reduce the impact of potential confounding factors. This approach ensured that every strain would have an equal opportunity to be included in the sample, thereby decreasing bias and enhancing representativeness.

Furthermore, with an adequately large sample size, a random sample significantly enhanced external validity by reliably representing the target population's characteristics. To address the challenges related to the complete random design, researchers ensured a full list of all individuals within the population and the ability to contact or access each selected member. They allocated sufficient time and resources for collecting data from the required sample size (Thomas, 2023). This approach ensured the selection of a representative subset of *E. coli*, thereby enhancing the findings' applicability to the broader population of this bacterial species.

Data Gathering Procedure

Data collection procedures were crucial to the research process, as the researchers ensured the reliability and validity of the results. This section of the report outlined the methods used to gather data, providing a clear and detailed explanation of how the necessary information was obtained. By describing the process step by step, the report helped readers understand how the data were collected, reinforcing the trustworthiness of the findings and allowing for replication or evaluation of the study.

Pre-Experimental Protocol

1. A formal letter was submitted to the school principal to request approval for the conduct of the study. After reviewing the objectives and procedures, permission was granted.
2. A formal letter was submitted to Digos Doctor's Hospital for permission to use their laboratory facilities. Approval was given after evaluating the purpose and scope of the study.
3. A formal letter was passed to Davao del Sur State College (DSSC) to use specific equipment needed for the study. The institution approved the request following their assessment.
4. A formal letter was submitted to the school administration to obtain permission to use the available laboratories and equipment. Access was granted upon approval of the research plan.

Collection and Extraction of Plant Materials

The following procedures were adapted from the study of Mersil and Alifia (2023):

1. Dried leaves of *C. mindanaense* were collected for 2 weeks. The researchers used an electronic weighing scale for the leaves to ensure equal measurements. Then, the leaves were crushed into a coarse powder using an electric grinder.
2. The plant extract was prepared using the maceration method. Three hundred grams of the powdered material were evenly soaked in 1 liter of 70 % ethanol solution for 72 hours.
3. The resulting extracts were filtered with filter paper to separate the liquid extract from the solid residue.
4. The solvent was evaporated using a rotary evaporator for an hour at 100°C to obtain the extract with the active ingredients of the kaningag leaves.

5. The extract was stored in a glass jar at 4°C in the refrigerator for future antibacterial testing and chemical analysis to protect it from light and degradation.
6. All materials used in the extraction process were sterilized to prevent bacterial contamination. Each item was first washed with dish soap, dried with a clean tissue, and then sterilized by pouring a 70% ethanol solution over the surfaces. This method ensured effective disinfection while preserving the integrity of the materials used.

Preparation of *E. coli* and Culture Media

The following procedures were adapted from the study of Son and Taylor (2022):

1. *E. coli* was purchased from a private laboratory in Digos City.
2. Before actuating the inoculation for the bacterial cultures, the researchers wore complete personal protective equipment (PPE), which included a laboratory gown, gloves, and a face mask to maintain and ensure proper handling and safety.
3. *E. coli* bacteria were obtained from a laboratory in a particular hospital.
4. An 18MH-agar plates were prepared and used as a medium for storage and culture for the bacteria set-up.
5. A culture tube containing a normal saline solution was prepared and inoculated with cultures of *E. coli*.
6. Each MH agar plate was evenly streaked on its surface using a sterile swab dipped into the inoculum.
7. The culture medium was isolated and placed in a controlled environment to maintain its bacterial growth and culture.

Formulation of Ointment Base

The following procedures were adapted from the study of Popa et al. (2020):

1. The ointment base was prepared with the help of a qualified pharmacist by mixing 250 grams of petroleum jelly and 250 grams of mineral oil in a water bath at 40°C to obtain a homogenous base.
2. The 500 grams of the ointment base were divided into five sets to obtain the needed measurements for each concentration. The mass of each solution was 100 grams. Four solutions were mixed with the leaf extracts, and one was not mixed with any extract for the negative treatment group.
3. To ensure equal measurements, an electronic weighing scale was used for each ointment base measurement for each concentration, which is the following: 97.5 grams, 95 grams, 92.5 grams, and 90 grams.

Formulation of Concentrations

The following procedures were adapted from the study of Magleo et al. (2017):

1. The kaningag (*C. mindanaense* Elm.) leaf extract was gradually added to the ointment and stirred thoroughly to ensure the even distribution of the compounds.
2. For Test 1, a concentration of 2.5 % of kaningag leaf extract was prepared by dissolving 2.5 g of extract in 97.5 g of pure ointment base. The concentration was computed by solving for the percentage mass of the solute which is to divide the mass of the solute (2.5 g) and mass of solution (100 g) and multiplied by 100 to achieve the desired concentration of 2.5 %. Furthermore, the mass of the pure ointment base was calculated by subtracting the mass of the solution (100 g) and the mass of the solute (2.5 g) to obtain 97.5 g.
3. Second, for Test 2, a concentration of 5.0% of kaningag (*C. mindanaense* Elm.) leaf extract was prepared by dissolving 5.0 g of extract in 95.0 g of pure ointment base. The concentration was computed by solving for the percentage mass of the solute which is to divide the mass of the solute (5 g) and mass of solution (100 g) and multiplied by 100 to achieve the desired concentration of 5 %. Furthermore, the mass of the pure ointment base was calculated by subtracting the mass of the solution (100 g) and the mass of the solute (5 g) to obtain 95 g.

4. Third, for Test 3, a concentration of 7.5 % of kaningag (*C. mindanaense* Elm.) leaf extract was prepared by dissolving 7.5 g of extract in 92.5 G of pure ointment base. The concentration was computed by solving for the percentage mass of the solute which is to divide the mass of the solute (7.5 g) and mass of solution (100 g) and multiplied by 100 to achieve the desired concentration of 7.5 %. Furthermore, the mass of the pure ointment base was calculated by subtracting the mass of the solution (100 g) and the mass of the solute (7.5 g) to obtain 92.5 g.
5. Lastly, for Test 4, a concentration of 10% of kaningag (*C. mindanaense* Elm.) leaf extract was prepared by dissolving 10 g of extract in 90 g of pure ointment base. The concentration was computed by solving for the percentage mass of the solute which is to divide the mass of the solute (10 g) and mass of solution (100 g) and multiplied by 100 to achieve the desired concentration of 10 %. Furthermore, the mass of the pure ointment base was calculated by subtracting the mass of the solution (100 g) and the mass of the solute 10 g) to obtain 90 g.

Determination of the antibacterial activity of *C. mindanaense*

The following procedures were adapted from the study of Okunye et al. (2020):

1. To assess the antibacterial activity of *C. mindanaense*, the antibacterial ointment was applied to a Petri dish inoculated with *E. coli* bacteria.
2. The effect of the extract on the bacteria was monitored using a measuring tool (ruler) to observe any reactions or changes in bacterial growth.
3. Continuous observations were made to track the size of the inhibition zone, allowing for an assessment of the extract's effectiveness.

Measures

This study adopted the Agar diffusion method, as reported by (Salina et al., 2018). The method involves placing the plates at 37 °C for 22 hours. Antibacterial activities are then assessed based on the diameters of the zone of inhibition (ZOI) in mm through each area with a ruler. An experienced medical laboratory technologist with more than two years of experience in bacterial infection studies, who specifically examined the effectiveness of the kaningag-based antibacterial ointment in inhibiting *E. coli* growth. Based on the outcomes of Minejima et al. (2019), this research aimed to establish the antibacterial activity of the kaningag ointment, monitoring the period in days for the inhibition of bacterial growth. Also, the ZOI was taken to determine the degree of the antibacterial activity of the ointment through the criteria of Hudzicki (2016), wherein an inhibition zone size less than 14 mm is resistant, while those 15-17 mm wide are moderate, and a zone of 18 mm or above is sensitive.

The main objective of this evaluation was to assess the potential efficacy and the inhibition zone of the kaningag (*C. mindanaense* Elm.) ointment for its antibacterial effect on wound healing, particularly against *E. coli*. Experts' assessments focus on parameters rated on a specified standard scale to determine the effectiveness of the leaf extract in an ointment for managing wounds caused by *E. coli* bacteria. Post-assessment, the compiled evaluations were organized using an interpretation table and subjected to systematic analysis. This analytical approach aimed to display the effects of the extracts on pivotal wound healing parameters necessary for potential future applications against *E. coli* bacteria.

Table 1. *E. coli*'s Growth Inhibition Interpretation

Mean Score Interval	Descriptive Equivalent	Interpretation
≥18 mm	Susceptible	The antibacterial activity showed an excellent image of the zone of inhibition.
15.00 mm - 17.00 mm	Moderate	The antibacterial activity showed a fair image of the zone of inhibition.
≤14 mm	Resistant	The antibacterial activity showed a poor image of the zone of inhibition.

Analysis and Interpretation

Mean. It indicates the average size of the inhibition zones, measured in millimeters, which reflects the extent of bacterial growth inhibition achieved by each concentration.

Independent-Samples Kruskal-Wallis Test. It is a non-parametric statistical test used to compare three or more independent groups to determine if there are statistically significant differences between them (DATAtab Team, 2025). This nonparametric alternative is commonly applied in decision-making scenarios where comparing the equality of multiple means is necessary, particularly when the data consists of precise values (Sherwani et al., 2021). The independent samples Kruskal-Wallis test can be utilized in this study to compare the antibacterial effectiveness of different concentrations of kaningag (*C. mindanaense* Elm.) leaf extract against *E. coli*, especially when the data is non-normally distributed. This test determines whether there are significant differences among treatment groups, allowing for further post hoc analysis, such as Dunn's test, to identify which specific concentrations vary in effectiveness.

Dunn's Test. It is used to conduct pairwise comparisons between independent groups and identify which groups show statistically significant differences at a given significance level (α) (Bobbitt, 2020). It can be used in this study as a post hoc analysis following the Kruskal-Wallis test to determine which specific concentrations of kaningag leaf extract exhibit significant differences in antibacterial effectiveness. By applying multiple comparison adjustments, this test ensures accurate identification of the most effective concentrations while minimizing the risk of statistical errors.

Ethical Considerations

This research adhered to strict ethical guidelines to promote responsible and conscientious study practices. These measures were taken to maintain the study's credibility, ensure the appropriate handling of biological materials, and comply with scientific, environmental, and institutional regulations.

Biosafety and security. This is important, especially in ensuring that the safety of the researchers and others involved during this study was not compromised. According to Iberdrola (2021), it is a field full of complexities that involves danger, which is why certain rules, regulations, and protective measures should be implemented to prevent potential threats from exposure to infectious microorganisms.

Environmental responsibility. This should be considered, especially if the experiments done in the study could cause a significant effect on the environment. In this study, researchers assure that from the extraction to the experimentation of the leaves and the culturing of the bacteria, the environment was not ignored nor harmed.

Integrity and transparency. The integrity in research is a set of ethical and moral standards that stand as a pillar for the conduct of research (Zhaksylyk, 2023). The results of this study were fully and honestly made. Every result from this study was analyzed carefully and documented without any biases that would otherwise jeopardize the whole paper. It was also checked to see if every citation was addressed correctly to the right author, making sure to avoid any form of plagiarism. Every outcome shown is presented without favoritism or one-sidedness to remain transparent and objective.

RESULTS AND DISCUSSION

This chapter deals with the presentation, analysis, and interpretation of data. The first part describes the antibacterial effectiveness of various concentrations of kaningag (*Cinnamomum mindanaense* Elm.) leaf extract, along with the control treatments—pure ointment base and commercial topical antimicrobial ointment. The second part presents the significance of differences between concentrations in inhibiting the growth of *Escherichia coli*, including comparisons with the control treatments and statistical analysis using Dunn's Test and Independent-Samples Kruskal-Wallis Test.

Antibacterial Effectiveness of Different Kiningag Leaf Extracts in Antibacterial Ointment Against *E. coli*

The study determined the effectiveness of kiningag (*C. mindanaense* Elm.) leaf extracts as an active ingredient in the antibacterial ointment for inhibiting the growth of *E. coli* with four different treatments: Treatment 1 – 2.5 %; Treatment 2 – 5 %; Treatment 3 – 7.5 %; and Treatment 4 – 10 %, all consisting of kiningag leaf extract. The researchers determined the antibacterial property of each concentration by seeing the size of the zone of inhibition per treatment in each replication (R1, R2, R3).

Table 2 presents the antibacterial activity of different concentrations of kiningag leaf extract on the inhibition of *E. coli*. As shown in the table, the results demonstrated a distinct correlation between the extract and its antibacterial effectiveness, with increasing concentrations leading to greater activity. In contrast to T2, T3 and T4, T1, which contains 2.5 % of kiningag leaf extract, showed no antibacterial effect, with a mean inhibition zone of 0 mm and a standard deviation of 0.00, thereby classifying it as resistant. T2 with 5 % of the extract, demonstrated a mean zone of 18 mm and a standard deviation of 1.73, indicating a susceptible response. T3, containing 7.5 %, had a mean of 16 mm and a standard deviation of 1.00, showing a moderate result. The highest antibacterial effect was observed in T4, which contains 10 % of the extract, with a mean of 31 mm and a standard deviation of 9.54, classifying it as susceptible. Hence, the researchers obtained the following results.

Table 2. Antibacterial Effectiveness of Different Kiningag Leaf Extracts in Antibacterial Ointment against *E. coli*

Treatments	Zone of Inhibition (in mm)			Mean	SD	Description
	R1	R2	R3			
T1	0	0	0	0	0.00	Resistant
T2	19	19	16	18	1.73	Susceptible
T3	16	15	17	16	1.00	Moderate
T4	40	32	21	31	9.54	Susceptible

These findings primarily contradicted the study by Agton et al. (2019), which stated that the kiningag (*C. mindanaense* Elm.) leaf extract did not exhibit any zone of inhibition and antibacterial activity against *E. coli*. This gram-negative bacterium is more likely to develop MDR (Prastiyanto et al., 2024). However, other studies supported the idea that certain parts of various *Cinnamomum* species contain high levels of bioactive phytochemical molecules (Mohamed et al., 2020). These bioactive phytochemical molecules include antimicrobial active compounds like cinnamaldehyde and eugenol, which are the main reasons for its antibacterial activity (Parisa et al., 2019). Additionally, this finding has already been verified by Magleo et al. (2017), who stated that *C. mindanaense* bark extract showed antibacterial activity. Due to the literature gap of *C. mindanaense*, this finding showed the great significance of kiningag leaf extract as an antibacterial agent against *E. coli*.

Antibacterial Effectiveness of Control Treatments on *E. coli*

The study included the antibacterial effectiveness of the positive and negative control treatments, using a commercial topical antimicrobial ointment in the form of ointment as the positive control and an ointment base as the negative control, against *E. coli*. An efficient amount of the commercial topical antimicrobial ointment was directly squeezed from a 3.5 g tube, scooped with a sterile cotton applicator, and applied to the center area of the disc. This process was repeated for each replicate, using a new sterile cotton applicator for each application to prevent cross-contamination. The discs were then incubated at 37°C for 22 hours. After incubation, the zone of inhibition of each disc was measured in millimeters. The negative control, an ointment base, was applied in the same manner and incubated under the same conditions as the positive control. The zone of inhibition was also measured after incubation. Table 3 shows that the mean inhibition zone of the positive control among the replicates reflects the average antibacterial effectiveness, indicating that it has

antibacterial activity against *E. coli* because of its measurable zone of inhibition. In contrast, the negative control's mean showed no inhibition zone, indicating that the ointment base alone has no antibacterial effect.

Table 3. Antibacterial Property of Commercial Treatment on *E. coli*

Treatments	Zone of Inhibition (in mm)			Mean	SD	Description
	R1	R2	R3			
Commercial Topical Antimicrobial Ointment	15	21	24	20	4.58	Susceptible
Ointment base	0	0	0	0	0.00	Resistant

The findings support the study of Yasodha et al. (2019), who demonstrated the effectiveness of a commercial topical antimicrobial ointment against *E. coli*, showing a comparable inhibition zone that suggests a consistent antimicrobial response. Similarly, Temüz et al. (2024) further confirmed the broader activity of commercial topical antimicrobial ointment against various bacterial strains, including *E. coli*. Additionally, Akinyemi (2020) states that tetracyclines, including this commercial topical antimicrobial ointment, are characterized by their exceptional chemotherapeutic efficacy against a wide range of Gram-positive and Gram-negative bacteria. The main use of tetracycline is attributed to its effectiveness in treating infectious diseases caused by *E. coli* and *Haemophilus influenzae*.

The negative control ointment base findings aligned with the study of Segueni et al. (2022), who reported that wounds treated with petroleum jelly alone exhibited a greater degree of inflammation, which appeared to be lessened in wounds treated with 30% ethanol extract of propolis (EEP) and propolis ointment. Additionally, Boudjelal et al. (2020) observed that while wounds treated with petroleum jelly showed some contraction, petroleum jelly itself has no therapeutic properties and keeps the wound moist. However, it is an effective base ointment carrier for transporting active constituents in topical animal medications (Grevialde & Ladiana, 2018). Furthermore, petroleum jelly offers advantages such as resistance to oxidation and prevention of microbial contamination, as it lacks water necessary for microbial growth (Kamrani et al., 2024).

Significant Difference in the Antibacterial Effectiveness of Different Kaningag Leaf Extracts in Antibacterial Ointment Compared to the Control Treatments in the Inhibition of *E. coli*

Table 4 presents the results of the comparative analysis on the antibacterial effectiveness of the experimental and control groups, analyzed based on the zone of inhibition. To assess the statistical validity of the study, a normality test using the Kolmogorov-Smirnov test was conducted to check if the data followed a normal distribution. The test revealed a significant departure from normality ($W = 0.215$, $p = 0.028$), indicating that the data on the inhibitory property of the treatment did not meet the assumptions required for parametric analysis. Consequently, this suggests alternative statistical methods may be needed to analyze the data properly. Moreover, the Independent-Samples Kruskal-Wallis Test was employed for the analysis. This nonparametric alternative is commonly applied in decision-making scenarios where comparing the equality of multiple means is necessary, particularly when the data consists of precise values (Sherwani et al., 2021).

It can be noted that the test statistic value for the overall zone of inhibition is 14.813, with 5 degrees of freedom and a p-value of 0.011, which is less than 0.05. This means that the study needed to reject the null hypothesis. This indicates a significant difference in the inhibitory property of the different extract concentrations of kaningag and the control treatments towards *E. coli* when observed using the zone of inhibition. Further, this means that different treatments have varying degrees of effectiveness in inhibiting *E. coli*.

Table 4. Significant Difference in the Antibacterial Effectiveness of Different Kuningag Leaf Extracts in Antibacterial Ointment Compared to the Control Treatments in the Inhibition of *E. coli*

Variables Reviewed	Test Statistic	df	p-value	Decision	Interpretation
Zone of Inhibition	14.813	5	0.011	Reject H_o	Significant Difference

To determine which of the three concentrations significantly differ from the others, a post hoc analysis was conducted, specifically pairwise comparisons of sample means using Dunn's test. According to Bobbitt (2020), Dunn's Test conducts pairwise comparisons between independent groups and identifies which groups show statistically significant differences at a given significance level (α).

Table 5 presents the results of post hoc comparisons conducted using Dunn's test. The analysis revealed several significant differences. T1 was significantly different from T4 ($p = 0.002$), indicating that T4 was better than T1; T1 was significantly different from the Positive control treatment ($p = 0.032$), suggesting that the Positive control treatment was better than T1; T4 was significantly different from the Negative control treatment ($p = 0.002$), with T4 being better than the Negative control treatment; and the Positive control treatment was significantly different from the Negative control treatment ($p = 0.032$), with the Positive control treatment being better than the Negative control treatment. These results suggested that T4 exhibits a higher level of susceptibility to the treatment or condition being tested compared to the other concentrations (T1, T2, T3) and the control groups (positive and negative). Its response was also comparable to the positive control (terramycin), which has a known effect (with a mean of 20mm and a standard deviation of 4.58), suggesting that T4 may be an effective concentration with a potential yielding similar results to a standard and proven treatment.

Meanwhile, no significant differences were observed between the remaining pairwise comparisons, including T1 vs. T2 ($p = 0.061$), T1 vs. T3 ($p = 0.172$), T1 vs. Negative ($p = 1.000$), T2 vs. T3 ($p = 0.612$), T2 vs. T4 ($p = 0.242$), T2 vs. Positive ($p = 0.785$), T2 vs. Negative ($p = 0.061$), T3 vs. T4 ($p = 0.093$), T3 vs. Positive ($p = 0.435$), and T3 vs. Negative ($p = 0.172$). These indicated no meaningful differences between these groups, indicating a comparable inhibitory effectiveness.

Table 5. Post Hoc Comparisons using Dunn's Test

	Test Statistic	p	Decision	Interpretation
Between T1 and T2	-8.000	0.061	Fail to Reject H_o	Not Significant
Between T1 and T3	-5.833	0.172	Fail to Reject H_o	Not Significant
Between T1 and T4	-13.000	0.002	Reject H_o	Significant
Between T1 and Positive	-9.167	0.032	Reject H_o	Significant
Between T1 and Negative	0.000	1.000	Fail to Reject H_o	Not Significant
Between T2 and T3	2.167	0.612	Fail to Reject H_o	Not Significant
Between T2 and T4	-5.000	0.242	Fail to Reject H_o	Not Significant
Between T2 and Positive	-1.167	0.785	Fail to Reject H_o	Not Significant

Between T2 and Negative	8.000	0.061	Fail to Reject H_o	Not Significant
Between T3 and T4	-7.167	0.093	Fail to Reject H_o	Not Significant
Between T3 and Positive	-3.333	0.435	Fail to Reject H_o	Not Significant
Between T3 and Negative	5.833	0.172	Fail to Reject H_o	Not Significant
Between T4 and Positive	3.833	0.369	Fail to Reject H_o	Not Significant
Between T4 and Negative	13.000	0.002	Reject H_o	Significant
Between Positive and Negative	9.167	0.032	Reject H_o	Significant

The post hoc analysis using Dunn's Test revealed significant differences between treatments. T4, the highest concentration, demonstrated significantly greater antibacterial activity than T1, the lowest concentration, and the negative control treatment (petroleum jelly). Additionally, the effectiveness of T4 was comparable to that of the positive control (terramycin). Meanwhile, no significant differences were observed among the other concentrations (T1, T2, T3), suggesting that these treatments exhibited similar antibacterial activity.

These findings suggested that kaningag leaf extract, particularly at higher concentrations (T4), has strong potential as an effective antibacterial agent against *E. coli*. The results also indicated its efficacy comparable to standard treatments like the commercial topical antimicrobial ointment. This supported the study by Nuñez et al. (2021), which highlighted the traditional medicinal use of kaningag (*C. mindanaense* Elm.) due to its bioactive compounds, including cinnamaldehyde and cinnamic acid, both of which are also known for their antimicrobial properties. Furthermore, Albuero et al. (2018) emphasized the pharmacological significance of *Cinnamomum* species, particularly for their therapeutic potential in combating bacterial infections.

Moreover, the results of this study directly contradicted and disproved the findings of Agton et al. (2019), which claimed that kaningag leaf extract exhibited no antibacterial activity against *E. coli*. The significant antibacterial effect observed in this study, particularly at higher concentrations, highlighted kaningag leaf extract as a promising natural antibacterial agent. Given that *E. coli* is known for its adaptability and increasing resistance to conventional antibiotics, these findings underscore the potential of *C. mindanaense* as an alternative treatment in antibacterial therapy. Further research is needed to explore its full therapeutic potential, optimize its application, and assess its efficacy against other bacterial strains.

Summary

This study aimed to evaluate the antibacterial effectiveness of leaf extracts from kaningag (*C. mindanaense* Elm.) at different concentrations as an active ingredient in antibacterial ointment in inhibiting *E. coli* growth. The results indicated that kaningag leaf extract, especially at higher concentrations (T4), exhibits significant potential as a powerful antibacterial agent against *E. coli*. The findings revealed a clear relationship between the extract and its antibacterial potency, showing that higher concentrations enhanced effectiveness.

In T1, which contained 2.5% kaningag (*C. mindanaense* Elm.) leaf extract, no antibacterial activity was observed, classifying it as resistant. T2, with a 5% concentration, exhibited an 18mm mean inhibition zone, indicating susceptibility. Meanwhile, T3, containing 7.5%, showed a moderate effect with a mean inhibition zone of 16mm. The strongest antibacterial activity was recorded in T4, which had a 10% extract concentration, producing a mean inhibition zone of 31mm with a standard deviation of 9.54, also classified as susceptible.

Notably, the results indicated its efficacy comparable to standard treatments like commercial topical antimicrobial ointments. It demonstrated significantly different antibacterial effects, underscoring their

potential as natural antibacterial agents. The strong antibacterial activity observed in this study, especially at higher concentrations, emphasizes the potential of kaningag (*C. mindanaense* Elm.) leaf extract as a natural antibacterial agent. Since *E. coli* is highly adaptable and increasingly resistant to standard antibiotics, these results highlight *C. mindanaense* as a promising alternative for antibacterial treatment. Additional research is required to fully explore its therapeutic potential, refine its applications, and evaluate its effectiveness against other bacterial strains.

CONCLUSION

In this study, we examined how the different concentrations of kaningag (*C. mindanaense*) leaf extract ointment affected the growth of *E. coli*. The results showed clear differences across the different concentrations of the treatments. From the data gathered, the following conclusions emerged:

1. The antimicrobial effectiveness of the treatments varied significantly. The most effective treatment was T4, which exhibited a mean zone of inhibition of 31 mm, indicating strong antibacterial activity. In contrast, T1 was the least effective, showing no zone of inhibition (0 mm), suggesting minimal or no antibacterial effect at that concentration.
2. The antibacterial effectiveness of the control treatments against *E. coli* was evaluated. The results indicated that the positive control (commercial topical antimicrobial ointment) displayed an expected average zone of inhibition of 20 mm against *E. coli*. In contrast, the negative control (ointment base) did not display any zone of inhibition, therefore, *E. coli* is resistant to the negative control. This result also indicates that the ointment base has no antibacterial properties.
3. The study explored the antibacterial efficacy of varying concentrations of kaningag (*C. mindanaense* Elm.) leaf extracts against *E. coli*. The results revealed significant differences in the inhibitory effects of the different concentrations on *E. coli* growth. This indicates that kaningag leaf extracts possess notable antibacterial properties, particularly at specific concentrations. Consequently, these findings suggest that such extracts could represent a viable alternative for antibacterial treatments to combat *E. coli* infections.

RECOMMENDATIONS

With the outcomes presented in the conclusion, the researchers recommend the following:

1. Department of Health (DOH) officials should further explore the inhibitory effect of kaningag (*C. mindanaense* Elm.) leaf extract on *E. coli*. The DOH officials should also look more deeply into the alternatives that could be provided by other means in the fight against antimicrobial-resistant bacteria, developing a safer and feasible option for the public.
2. The Department of Science and Technology (DOST) should further investigate kaningag (*C. mindanaense* Elm.) as a natural antibacterial agent and explore its potential in creating cost-effective medicinal products. The DOST should also encourage local cultivation of kaningag to support sustainable agriculture and eco-friendly healthcare alternatives.
3. Pharmaceutical companies should delve into the different ways they could utilize kaningag and other plants to improve their potential further in making new pharmaceutical products.
4. Healthcare professionals should collaborate to find more plant-based alternatives like kaningag and investigate the different natural ways of providing treatment to improve the care they give to others.
5. It is highly encouraged that local communities research the effectiveness of alternative medicines. By forming cooperatives to grow kaningag, they could create a low-cost income source, support traditional herbal practices, and develop viable products for health and economic benefit.
6. Researchers suggest that patients who suffer from wound infections due to *E. coli* should evaluate the

efficacy of kaningag-based products. Patients, overall, should also assess whether the said product undergoes thorough research and quality testing before procuring it.

7. Future researchers should search for other ways to extract the kaningag leaves. A proper timeline must be implemented, with delays being factored in, to give them enough time to finish the experiments. They should also observe adequate sterilization and wear standard protective gear to avoid contamination when preparing the ointment. They should also consider increasing the concentrations of the leaf extracts added. Also, they must highlight that once the ointment has been made, it should be sent to the laboratories immediately to prevent the risk of the efficacy going down. Lastly, future researchers should test the ointment on human skin to observe its effectiveness against wound infection.

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