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ORIGINAL RESEARCH

MJM BIOLABS

In-vitro Synergistic Antimicrobial Activity of *Melia azedarach* and *Solanum incanum* Ethanolic Leaf Extracts Against the *Staphylococcus aureus*

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Abstract

The increasing challenges of adverse drug reactions, costly medicines, and antimicrobial resistance necessitate the exploration of safe and effective antimicrobial agents. This study aimed to evaluate the antimicrobial activity of ethanol leaf extracts from *Melia azedarach* and *Solanum incanum* against *Staphylococcus aureus*, a clinically significant pathogen known for its resistance to conventional antibiotics. The research design employed was an experimental study with three concentrations of the plant extracts (1g/ml, 1.4g/ml, and 2g/ml) tested against *Staphylococcus aureus*, and the standard antimicrobial agent, ciprofloxacin, served as a positive control, while ethanol served as a solvent control. The samples were prepared, and the experiments conducted in triplicate, following established protocols. The choice of *Melia azedarach* and *Solanum incanum* was scientifically justified based on their well-documented traditional medicinal uses and previous reports of antimicrobial activity. Ethical approval was obtained from the Kabarak University Research Ethics Committee (KUREC), and a research permit was obtained from the National Commission for Science, Technology, and Innovation (NACOSTI). The results demonstrated significant anti-staphylococcal activity for both extracts, with *Melia azedarach* exhibiting superior efficacy compared to *Solanum incanum* at all concentrations tested. Moreover, the combined extracts displayed enhanced antimicrobial effects against *Staphylococcus aureus* compared to the individual extracts. Phytochemical analysis revealed the presence of tannins, coumarins, flavonoids, and anthraquinones in both extracts, with higher levels of steroidal glycosides in Solanum incanum. We recommend further phytochemical and pharmacological investigations to identify the active antimicrobial constituents in these plants. These findings hold promise in the development of novel therapeutic agents to combat infectious diseases, offering *Melia azedarach* and *Solanum incanum* as potential sources for drug development.

Keywords: *Solanum incanum, Melia azedarach, Staphylococcus aureus*, Disk diffusion method, MIC, Phytochemicals.

INTRODUCTION

Antimicrobial resistance has escalated into a formidable global public health crisis, presenting a pressing challenge in diverse countries (Smith et al., 2019). Among the alarming rise of resistant pathogens, methicillin-resistant Staphylococcus aureus (MRSA) stands as a menacing multidrug-resistant bacterium, jeopardizing the effectiveness of conventional antibiotic treatments for staphylococcal infections (Tong et al., 2015). *Staphylococcus aureus*, a gram-positive bacterium, inflicts a spectrum of ailments, ranging from mild to severe, encompassing skin infections, meningitis, pneumonia, sepsis, toxic shock syndrome, and endocarditis (Tong et al., 2015). This relentless increase in antimicrobial resistance has prompted a global call to action, necessitating urgent research to explore alternative and innovative antimicrobial agents to combat these challenges effectively.

With the limited efficacy of existing treatments against drug-resistant strains of *Staphylococcus aureus*, urgent exploration of alternative antimicrobial agents is imperative. In recent years, there has been a notable surge in interest in natural and semi-synthetic medicinal products derived from medicinal plants, showcasing their potential in combating drug-resistant microbes (Ogawa et al., 2019). Notably, *Melia azedarach* and *Solanum incanum* have garnered attention as potential sources of plant-derived antimicrobial agents, exhibiting promising activity against various bacterial strains (Zahoor et al., 2015). *Melia azedarach*, commonly known as the Chinaberry tree, and *Solanum incanum*, known as bitter apple, are widely distributed in Kenya and have been well-documented in ethnobotanical studies for their antimicrobial properties. The rich diversity of phytochemical compounds in these plant extracts, including alkaloids, tannins, flavonoids, and phenolic compounds, underpins their potential as effective antimicrobial agents (Zahoor et al., 2015). As scientific interest in medicinal plants continues to grow, harnessing the antimicrobial potential of *Melia azedarach* and *Solanum incanum* presents an exciting avenue in the quest to combat drug-resistant infections and develop novel therapeutic options.

The leaf extracts of *Melia azedarach* and *Solanum incanum* are rich reservoirs of phytochemical compounds, including alkaloids, tannins, flavonoids, and phenolic compounds, which contribute to their potent antimicrobial effects (Zahoor et al., 2015). Extensive research has demonstrated the broad-spectrum antibacterial activity of *Melia azedarach*, as evidenced by its inhibitory effects against *Proteus mirabilis*, *Enterobacter aerogenes*, *Pseudomonas aeruginosa*, *Escherichia coli*, *Staphylococcus aureus*, and *Bacillus subtilis* (Abbas et al., 2017; Zahoor et al., 2015). *Solanum incanum* extracts have also exhibited notable bactericidal and nematicidal properties (Anwar, 2018), complemented by their traditional use in wound healing, pain relief, and treatment of protozoan infections (Mwonjoria et al., 2014; Qureshi et al., 2019; Waweru et al., 2017). These observations underscore the potential therapeutic value of these plant extracts in addressing drug-resistant bacterial infections and inspire further investigation into their antimicrobial mechanisms and potential applications in modern medicine. As the global urgency to combat antimicrobial resistance grows, the exploration of *Melia azedarach* and *Solanum incanum* as valuable sources of novel antimicrobial agents becomes increasingly imperative.

Despite individual investigations of their antibacterial activity, no study has yet explored the potential synergistic effects of combining leaf extracts of *Melia azedarach* and *Solanum incanum* against *Staphylococcus aureus*. Therefore, this study aims to evaluate the combined antimicrobial activity of these plant extracts against Staphylococcus aureus, with the goal of identifying potential novel therapeutic options in the fight against antimicrobial-resistant infections.

METHODOLOGY

Location of the Study

The study was conducted at Kabarak University, utilizing the facilities of the School of Pharmacy laboratory for Pharmacognosy (extraction) and the Pharmaceutical Microbiology lab for evaluation. Fresh *Melia azedarach* leaves were collected from Kabarak University's botanical garden, and *Solanum incanum* leaves were sourced from Rafiki area, Rongai constituency, Nakuru County, Kenya (latitude: -0.2735, longitude: 35.9886). Collected samples were then taken to the Pharmacognosy laboratory

for identification by the school botanist, and herbarium specimens were prepared (Herbarium voucher number: KU/HB/2023/001).

Sample Preparation

The collected leaves were washed with distilled water and dried under shade at room temperature for 2 days. After drying, the leaves were pounded using a mortar and pestle to reduce them to a coarse powder. Separate quantities of 150 grams of Solanum incanum powder and 150 grams of Melia azedarach powder were weighed and added to labeled conical flasks, with each flask containing 300 ml of ethanol. The flasks were then placed on a rotary shaker for three days to facilitate the extraction process. Following extraction, the plant extracts were filtered using Whatman filter paper, and the ethanol solvent was evaporated using a warm water bath to obtain concentrated extracts. Subsequently, paper discs with a diameter of 5 mm were generated by punching dry and clean Whatman filter paper. These discs were soaked in the obtained extracts of different concentrations, as required for the Minimum Inhibitory Concentration (MIC) determination, and allowed to absorb the extract for 24 hours to ensure proper impregnation. This process prepared the test discs for subsequent antimicrobial evaluation.

Media and Inoculum Preparation

Media and inoculum preparation was conducted with strict adherence to standard protocols. Nutrient broth, used for growing the staphylococcal isolates and for determining the Minimum Inhibitory Concentration (MIC), was prepared by dissolving 8.4 grams of nutrient agar in 300ml of distilled water and heated to boiling to ensure complete dissolution. Similarly, Mueller Hinton Agar (MHA) was prepared for culturing bacteria during the disc diffusion method, by dissolving 15.2 grams of MHA in 400ml of distilled water and heating it to boil for complete dissolution. Both media were subjected to sterilization through autoclaving at 121°C for 15 minutes, followed by cooling to 45°C. Media sterility checks were performed by incubating samples of each medium alone at 37°C for 24 hours to verify their ability to support growth. Additionally, media viability checks were carried out by inoculating samples of each medium with bacteria to confirm their efficacy in supporting bacterial growth. *Staphylococcus aureus* isolates were sourced from the reservoir stored in the Pharmaceutical Microbiology Laboratory, School of Pharmacy, Kabarak University. The bacteria were grown in nutrient broth and incubated at 37°C for 24 hours, following the CLSI (2022) guidelines.

Phytochemical Analysis

Seven classes of phytochemicals were tested including: tannins, flavonoids coumarins anthraquinones, saponins, phytosterols and alkaloids (Mutungi et al. 2023; Ahmed and Sulaiman 2018).

Table 1:

List of Phytochemical Classes and the Tests Used to Evaluate their Presence

Phytochemical class	Detection test	Procedure
Tannins	Iron chloride test	The ethanolic extract was dissolved in distilled water then filtered. 3 drops of ferric chloride were added. Formation of grey color indicates presence of tannins
	Lead acetate test	To the ethanolic extracts, 3 drops of lead acetate were added. Formation of white buffy precipitate indicates presence of tannins
	Iodine test	Dilute iodine was added to the ethanolic extracts. Appearance of transient red colour indicates presence of tannins
	Acetic acid test	A few drops of Acetic acid were added to small amounts of the ethanolic plant extracts and the colour change observed. A green or blue color indicates presence of tannins.
Flavonoids	Alkaline test	Extracts were treated with a few drops of sodium hydroxide solution. Formation of intense yellow color, which becomes colorless on the addition of dilute acid, indicates the presence of flavonoid
	Shinoda test	A few fragments of magnesium turnings and concentrated sulphuric acid were added to the ethanol extract. Appearance of red to pink color after few minutes indicates presence of flavonoids
	Lead acetate test	Extracts were treated with a few drops of lead acetate solution. Formation of a yellow colour precipitate indicates the presence of flavonoids.
	Ammonia test	Dilute ammonia was added to a portion of the extract then concentrated sulphuric acid added. Formation of a yellow brown colour
Coumarins	Iron chloride test	Few drops of FeCl3 solution was added to the ethanolic extracts. Formation of dark green colour indicates presence of coumarins
Anthraquinones	Bontragers test	The extracts were boiled with dilute sulphuric acid and chloroform added to the filtrate and shaken well. The lower layer obtained then few drops of ammonia added. The lower ammoniacal layer takes pink color
	Modified Bontragers test	3 drops of Ferric chloride were added to the sulphuric acid extract and boiled for 2 minutes and then filtered and chloroform added to the filtrate and shaken well. The lower layer was obtained then few drops of ammonia added. The lower ammoniacal layer takes pink color
Saponins	Froth test	5m of extracts was shaken for about 1 minute, and then allowed to stand for 10 minutes. Formation of persistent froth indicates presence of Saponins
Phytosterols	Salkwoski test	Extract treated with chloroform and filtrates were treated with a few drops of conc. Sulphuric acid, shaken and allowed to stand. Appearance of golden yellow colour indicates the presence of triterpenes
	Lieberman's test	Extracts were treated with chloroform and filtered. The filtrates are treated with a few drops of acetic anhydride, boiled and cooled. Conc. Sulphuric acid is added. The formation of brown ring at the junction indicates the presence of phytosterols.
Alkaloids	Mayer's test	Extracts were treated with Mayer's reagent (potassium mercuric iodide). Formation of a yellow-colored precipitate indicates the presence of alkaloids
	Wagner's test	Extracts were treated with Wagner's reagent (iodine in potassium iodide). The formation of a brown/reddish precipitate indicates the presence of alkaloids
	Dragendorff test	Extracts were treated with Dragendroff's reagent (solution of potassium bismuth iodide). Formation of a red precipitate indicates the presence of alkaloids
	Hager's test	Extracts were treated with Hager's reagent (saturated picric acid solution). Formation of yellow colour indicates the presence of alkaloids

Minimum Inhibitory Concentration (MIC) Determination

Minimum inhibitory concentrations (MICs) were determined through serial dilution of the plant extracts at four different concentrations. Fourteen grams of Melia azedarach leaf extract and 10 grams of *Solanum incanum* leaf extract was separately diluted in 5 ml of dimethyl sulfoxide (DMSO) in the first pair of test tubes. Subsequently, 2 ml of DMSO was added to the remaining three test tubes. In a stepwise manner, 1 ml of the mixture from the first test tube was transferred to the second test tube and thoroughly mixed. Then, 1 ml from the second test tube was drawn and transferred to the third test tube, and the same process was repeated for the fourth test tube. This resulted in the preparation of four different concentrations for Solanum incanum (2g/ml, 1g/ml, 0.5g/ml, 0.25g/ml) and Melia azedarach (2.8g/ml, 1.4g/ml, 0.7g/ml, 0.35g/ml) extracts, respectively. The serial dilutions allowed for a range of concentrations to be tested, enabling accurate determination of the MIC values for each extract.

Disc Diffusion Method

The prepared Mueller Hinton Agar (MHA) medium was aseptically added to sterile culture plates and allowed to solidify. Using a sterile wire loop, bacterial isolates obtained from the staphylococcal inoculum were streaked onto the MHA culture medium in a zigzag pattern. Subsequently, paper discs impregnated with the plant extracts at different concentrations were carefully placed on top of the streaked culture media. To ensure sufficient spacing, appropriate distances were maintained between the discs themselves and between the discs and the walls of the petri dishes. As positive and negative controls, Gentamicin and dimethyl sulfoxide (DMSO) were used, respectively. Following preparation, the cultures were incubated at 37°C for 24 hours. After the incubation period, the diameter of the zone of inhibition around each disc was measured using a Vernier caliper and the values were tabulated for further analysis. This methodology allowed for accurate assessment of the antimicrobial activity of the plant extracts against the test organism, *Staphylococcus aureus*.

Ethical Considerations

Ethical considerations were meticulously addressed throughout the course of this study. Approval was obtained from the School of Pharmacy, ensuring adherence to institutional guidelines and regulations. Additionally, the study received ethical clearance from the Kabarak University Research Ethics Committee (KUREC), which ensures the protection of human subjects and upholds ethical standards in research. Furthermore, the National Commission for Science, Technology, and Innovation (NACOSTI) granted the necessary research permit, affirming compliance with national regulations and guidelines for conducting research involving human subjects or biological samples. The researchers followed rigorous ethical protocols to safeguard the rights and well-being of study participants, ensure informed consent, and maintain data confidentiality. Ethical considerations extended to the handling of plant materials, ensuring sustainable collection practices and preservation of the local biodiversity.

Data Analysis

Data analysis was performed SPSS, and the results were presented descriptively and inferentially. The antimicrobial activity of *Melia azedarach* and *Solanum incanum* extracts against *Staphylococcus aureus* was assessed by measuring the zone of inhibition around each disc. The obtained data were subjected to statistical tests, such as analysis of variance (ANOVA) and post-hoc tests, to determine significant differences between the various concentrations of the plant extracts. The minimum inhibitory concentration (MIC) values were determined through serial dilution experiments. Additionally, the phytochemical composition of the extracts was analyzed using descriptive statistics. The results were presented in tables, graphs, and figures.

RESULTS

Solvent extraction

Table 2 below depicts the mass of plant extract obtained after extraction. A total of 14 grams and 10 grams of plant extract was obtained from the plant specimens' *Melia azedarach* and *Solanum incanum* respectively.

Table 2:

Mass of Leaf Extract Obtained from the Extraction Process for Each Plant

Plant extract	Extract mass
Melia azedarach	14 grams
Solanum incanum	10 grams

Phytochemical Analysis

Phytochemical analysis of *Solanum incanum* and *Melia azedarach* leaves extracts showed the presence of Anthraquinones, saponins, flavonoids, coumarins, steroidal glycosides, tannins and traces of alkaloids, which could have contributed to the antimicrobial activities of the plant extracts.

Table 3:

Phytochemical Analysis of Ethanol Leaf Extracts of Melia azedarach and Solanum incanum

Test	Observation	Inference
ANTHRAQUINONES		
Bontragers test *Solanum incanum* -Pink colour	*Melia azedarach*- Formation of pink color	+++ (high presence)
	+++ (high presence)	
Modified Bontragers test *Solanum incanum* -Pink colour	*Melia azedarach* -Formation of pink colour	++ (moderate presence)
	+ (low presence)	
SAPONINS		
Froth test *Solanum incanum* -Formation of persistent froth	*Melia azedarach*- Formation of persistent froth	+ (low presence)
	+ (low presence)	
FLAVONOIDS		
Alkaline test	*Melia azedarach*-Brown colour which disappears on adding H2SO4	+++ (high presence)
	Solanum incanum -Brown colour which disappears on adding H2SO4	+++ (high presence)
Shinoda test *Solanum incanum* -Formation of red colour	*Melia azedarach*- Formation of red pink colour	+ (low presence)
	+++ (high presence)	
lead acetate *Solanum incanum* -Formation of yellow colour	*Melia azedarach*- Formation of yellow colour	+++ (high presence)
	+++ (high presence)	

Test	Observation	Inference
Ammonia test *Solanum incanum* -Formation of yellow brown colour	*Melia azedarach*-Formation of yellow brown colour	+++ (high presence)
	+++ (high presence)	
COUMARINS		
$FeCl_3$ test	*Melia azedarach*-Formation of dark green colour	+++ (high presence)
	Solanum incanum -Formation of dark green colour	+++ (high presence)
STEROIDAL GLYCOSIDES		
Salkwoski test *Solanum incanum* -Formation of brown ring	*Melia azedarach*-Formation yellowish fluorescence	++ (moderate presence)
	+++ (high presence)	
Liebermans test *Solanum incanum* -Formation of brown layer	*Melia azedarach*-Formation of brown colour at lower ring	++ (moderate presence)
	+++ (high presence)	
TANNINS		
$FeCl_3$ test *Solanum incanum* -Formation of green colour	*Melia azedarach*-Formation of green colour	+++ (high presence)
	+++ (high presence)	
Lead acetate *Solanum incanum* -White brown buffy precipitate	*Melia azedarach*-White brown buffy precipitate	+++ (high presence)
	+++ (high presence)	
Iodine test *Solanum incanum* -No yellow colour	*Melia azedarach*-Formation of yellow colour	+++ (high presence)
	___ (no presence)	
ALKALOIDS		
Mayers test *Solanum incanum* -Slight yellow ppt	*Melia azedarach*-Slight yellow ppt	+ (low presence)
	+ (low presence)	
Wagners test *Solanum incanum* -Slight brown ppt	*Melia azedarach*-Slight brown ppt	+ (low presence)
	+ (low presence)	
Hagers test *Solanum incanum* -Slight yellow color	*Melia azedarach*-Slight yellow colour	+ (low presence)
	+ (low presence)	
Dragendorfs test *Solanum incanum* -Slight red ppt	*Melia azedarach*-Slight red ppt	+ (low presence)
	+ (low presence)	

Minimum Inhibitory Concentration Results

The MIC of *Solanum incanum* extract against *Staphylococcus aureus* was 1g/ml while for *Melia azedarach* extract against was 1.4g/ml as shown in table 4 below. Similarly, for the combined extracts, the MIC was 50% for *Staphylococcus aureus*.

Table 4:

Minimum Inhibitory concentration of Ethanol Leaf Extracts of Melia azedarach and Solanum incanum Against Staphylococcus aureus

Solanum incanum		*Melia azedarach*		Combined plant extracts	
Concentration (g/ml)	observation	Concentration (g/ml)	Observation	Concentration (g/ml)	observation
2	Clear	2.8	Clear	100	Clear
1	Clear (MIC)	1.4	Clear (MIC)	50	Clear (MIC)
0.5	Turbid	0.7	Turbid	25	Turbid
0.25	Turbid	0.35	Turbid	12.5	Turbid

Disc Diffusion Results

Solanum incanum ethanolic extract showed activity against *Staphylococcus aureus*. Highest zone of inhibition was observed at 2g/ml, with a diameter of zone of inhibition of 7.04mm. Lowest zones of inhibition were observed at 0.25g/ml (5.5mm). *Melia azedarach* showed highest zone of inhibition at 7.5mm at a concentration of 2.8g/ml. Highest zones of inhibition for the combined extract was 9.56mm at 100% extract concentration while lowest diameter was found at 12.5% concentration (6.84mm). The combined extracts of *Solanum incanum* and *Melia azedarach* produced larger diameters of zones of inhibition as compared to individual diameter of zones of inhibition for each plant extract. The diameters of the zone of inhibition decreased with decreasing concentrations of the extracts, which means the susceptibility of the bacteria decreases with decreasing concentration of the extract.

Table 5:

In-Vitro Antimicrobial Activity of Ethanol Leaf Extracts of Melia azedarach and Solanum incanum Against Staphylococcus aureus

Solanum incanum		*Melia azedarach*		**Combined plant extracts**	
Concentration (g/ml)	Average zone of inhibition (mm)	Concentration (g/ml)	Average zone of inhibition (mm)	Concentration (g/ml)	Average zone of inhibition (mm)
2	7.04	2.8	7.50	100	9.56
1	6.92	1.4	6.96	50	8.12
0.5	6.78	0.7	6.02	25	7.65
0.25	5.50	0.35	5.72	12.5	6.84
Positive control (gentamicin)		26.68 mm			
Negative control (DMSO)		-			

DISCUSSION

The present studied aimed at exploring the combined effects of ethanolic leaf extracts from *Melia azedarach* and *Solanum incanum* on the bacterium *Staphylococcus aureus*. This was done through extraction, phytochemical screening, MIC determination and conducting disk diffusion analysis. The extraction process yielded 14 grams of *Melia azedarach* and 10 grams of *Solanum incanum* extract. Phytochemical analysis revealed that on average, ethanolic leaf extract of both *Melia azedarach* and *Solanum incanum* have moderate to high amounts of tannins, coumarins, flavonoids and anthraquinones. Flavonoids are well-known for not only antimicrobial properties but also antioxidant and anti-inflammatory actions which makes them useful in managing numerous conditions (Ghasemzadeh et al., 2015). Although, coumarins are recognized for their anti-thrombotic and anti-inflammatory actions,

they have also been demonstrated to have antifungal and antibacterial actions (Flores-Morales et al., 2023). Further, tannins have potent astringent action but also do possess antimicrobial properties as reported by Kaczmarek (2020). Therefore, the presence of these bioactive compounds in the extracts in such high amounts may suggest they are responsible for the anti-staphylococcal activity observed during the disk diffusion analysis. The remaining phytochemicals: alkaloids and saponins, were low in both the leaf extracts. Evidently, both alkaloids and saponins also do possess antimicrobial properties which might suggest a contributory role in the overall antimicrobial activity of the plant extract. Notably, steroidal glycosides were present in moderate amounts in the leaf extract of *Melia azedarach* but present in high amounts in leaf extract of *Solanum incanum*. This parallels study done by Sbhatu & Abraha (2020) that reported that steroidal glycosides are in high concentrations in *Solanum incanum* as glycol alkaloids. To note however, the amount of phytochemicals available in a plant at a given time is influence by multiple factors including prevailing climatic conditions, age of the plant and geographical location. Therefore, it is essential to acknowledge variations in phytochemical concentrations and approach these study finds with caution.

The MIC test is use to indicate minimum concentration required to inhibit growth of a microbe. In this study, the MIC of *Solanum incanum* extract against *Staphylococcus aureus* was 1g/ml, while that of *Melia azedarach* extract was 1.4g/ml. Further, the combined extracts exhibited minimum inhibitory concentration at 50% for *Staphylococcus aureus*. These results suggest that a higher concentration of the extracts is needed to inhibit *Staphylococcus aureus*. Indeed, as denoted by the results for disk diffusion, the is an increase in anti-staphylococcal activity for concentrations above the MIC. At the highest concentration, *Melia azedarach* had more activity as compared to *Solanum incanum* and this is also seen at the least concentration of the two extracts used. This suggests that in overall, *Melia azedarach* has more anti-staphylococcal activity than *Solanum incanum*. Notably, the combined anti-staphylococcal effect of both plant extracts was higher than any of the single plant's anti-staphylococcal effect but not additive of each other. This suggest a synergistic action when the two extracts are combined towards the staphylococcal bacteria. Although the anti-staphylococcal activity was much lower compared to gentamicin, we suggest that based on the trajectory of activity as we increase extract concentration, the plant extract may be equipotent to the drug thus providing an alternative treatment modality.

CONCLUSION

To summarize, this study reports that the leaf extract of both *Melia azedarach* and *Solanum incanum* have moderate to high levels of tannins, coumarins, flavonoids and anthraquinones which possess antibacterial effect. In contrary, akaloids and saponins were in low amounts in both extracts while steroidal glycosides were in moderate amounts in *Melia azedarach* but high in *Solanum incanum*. Further, the MIC for *Melia azedarach, Solanum incanum* and the combined plant extract was 1.4g/ml, 1g/ml and 50% respectively against *Staphylococcus aureus*. Both plant extracts demonstrated significant anti-staphylococcal activity with *Melia azedarach* showing more anti-staphylococcal activity than *Solanum incanum*. Thus, using the combined leaf extract of both plants produces more anti-staphylococcal activity compared to using individual plant extracts.

RECOMMENDATIONS

Based on the discussion and conclusions above, we recommend further phytochemical and pharmacological studies with a view of identifying active principles of the two plants that can provides leads for antibacterial drug development. (You cannot recommend use at this preliminary stage!!). the use of the combined extract as an alternative modality for treating staphylococcal infections; especially when formulated as shampoos and gels for skin infections attributed to *Staphylococcus aureus*. Further studies should be done to identify and isolated the specific phytochemicals responsible for the bacterial activity in order. Additionally, evaluations of anti-staphylococcal effect at concentrations almost levelling that of gentamicin should be conduct to validate whether they are equipotent.

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