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Phytochemical constituents of tea tree oil and garlic and their antibacterial activity against bacteria isolated from high vaginal swabs

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Abstract

Bacterial vaginal infections are common and often recurrent issues primarily caused by an imbalance in the vaginal flora. With rising antibiotic resistance, alternative treatments like natural substances are being explored. The aim of this study was to determine the phytochemical constituents and antibacterial activity of garlic and tea tree oil on high vaginal swab (HVS) bacterial isolates. A total of 75 HVS samples were collected and each was inoculated on Nutrient agar, Blood agar, MacConkey agar and de Man Rogosa Sharpe agar, and incubated at 37 °C for 24 hours. Pure isolates were identified through standard methods and molecular characterization. Garlic and tea tree oil were analysed for bioactive components using qualitative phytochemical methods. Antibacterial susceptibility testing was also carried out on the compounds using agar well diffusion method. Broth dilution method was used to determine the Minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC) of the isolates. The primary bacterial isolates identified out of the 75 bacterial isolates obtained were *Lactobacillus* spp, *Escherichia coli*, *Staphylococcus aureus* and *Shigella flexneri* which had a frequency distribution of 21, 35, 14 and 5 respectively. The qualitative phytochemical analysis showed presence of allicin, flavonoids, reducing sugar, saponins and tannins in garlic while tea tree oil showed presence of flavonoids, terpenoids and terpinen-4-ol. The inhibition zone diameters (IZD) for garlic ranged from 9.50±0.35 mm at 12.5 mg/ml against *S. flexneri* to 23.00±0.28 and 23.00±0.71 mm at 200 mg/ml against *E. coli* and *S. aureus* respectively, while the IZD values for tea tree oil ranged from 8.00±0.00 mm at 25 mg/ml against *Lactobacillus* spp to 25.00±0.78 mm at 200 mg/ml against *S. aureus*. The MIC values for garlic were 12.5 to 50 mg/ml, whereas tea tree oil showed MIC values of 12.5 to 100 mg/ml. Minimum bactericidal concentration (MBC) values for garlic ranged from 50 to 200 mg/ml and for tea tree oil from 25 to 200 mg/ml. These findings indicated that tea tree oil is more effective than garlic against the bacterial isolates. Both garlic and tea tree oil exhibit potent antibacterial effects against the isolates. These substances could be potential alternatives or adjuncts to conventional treatments for bacterial vaginal infections. *In vivo* studies and clinical trials are recommended to validate these findings and develop appropriate therapeutic formulations.

Keywords: Garlic; Tea Tree Oil; Antibacterial Activity; High Vaginal Swabs

1. Introduction

A high vaginal swab (HVS) is a medical procedure in which a sterile cotton swab is used to collect a sample of vaginal discharge from the vaginal canal. This sample is then sent to a laboratory for analysis to diagnose infections, identify potential pathogens and assess the vaginal microbiota. It is important to note that while a high vaginal swab is a valuable diagnostic tool, it may not provide a definitive diagnosis in all cases [1].

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Bacterial infections of the female reproductive system, particularly those identified through high vaginal swabs, pose significant challenges to women's health worldwide. These infections, which often involve pathogens such as *Escherichia coli*, *Staphylococcus aureus* and *Shigella flexneri*, can lead to severe reproductive health issues, including pelvic inflammatory disease, infertility and adverse pregnancy outcomes if not promptly and effectively treated [2]. The growing resistance of these pathogens to commonly used antibiotics exacerbates the problem, making it increasingly difficult to manage these infections using conventional antimicrobial therapies [3].

In response to the rising threat of antibiotic resistance, there has been a renewed interest in alternative therapeutic agents, particularly those derived from natural sources. *Melaleuca alternifolia*, commonly known as tea tree oil (TTO) and *Allium sativum*, known as garlic, are two such natural agents that have been extensively studied for their antimicrobial properties. Tea tree oil, extracted from the leaves of the Australian tea tree, contains a variety of bioactive compounds, with terpinen-4-ol and alpha-terpineol being the most notable for their antimicrobial effects [4]. These compounds have been shown to exhibit significant antibacterial activity against a broad spectrum of pathogens, including those that are resistant to conventional antibiotics [5]. Garlic, on the other hand, has a long history of use in traditional medicine across various cultures. It is particularly rich in sulphur-containing compounds, such as allicin, which are responsible for its characteristic odour and its potent antimicrobial properties [6]. Allicin and its derivatives have been found to inhibit the growth of a wide range of bacteria, including both Gram-positive and Gram-negative species [7]. Moreover, studies have demonstrated that garlic can enhance the immune response and has potential as a complementary therapy in the treatment of bacterial infections [8].

Despite the growing body of evidence supporting the antimicrobial properties of tea tree oil and garlic, their application in the context of bacterial infections isolated from high vaginal swabs remains relatively underexplored. High vaginal swabs are routinely used in clinical settings to diagnose infections of the female reproductive tract and identifying effective natural treatments for these infections could have significant implications for public health, particularly in regions with high rates of antibiotic resistance and limited access to healthcare [9]. This study, conducted in a teaching hospital in the South Eastern part of Nigeria, seeks to address this gap by evaluating the phytochemical constituents of tea tree oil and garlic and assessing their antibacterial activity against bacteria isolated from high vaginal swabs. By focusing on these natural agents, this research aims to contribute to the development of alternative therapeutic strategies for managing vaginal infections, offering a potential solution to the growing problem of antibiotic resistance.

2. Material and methods

2.1. Study area

The research was conducted at Chukwuemeka Odumegwu Ojukwu University Teaching Hospital, Awka, Anambra State, Nigeria.

2.2. Selected compounds

Garlic and tea tree oil were purchased from Eke-Awka market in Awka South Local Government Area of Anambra State, Nigeria. The samples were then transferred into sterile containers and transported to General Microbiology Laboratory, Nnamdi Azikiwe University, Awka, Anambra state for processing and analysis as described by [10].

2.3. Specimen collection

With the permission of the Ethical Committee of Chukwuemeka Odumegwu Ojukwu University Teaching Hospital, Awka, Anambra state, 75 high vaginal swabs were aseptically and properly collected from women at the gynecological department using sterile swab sticks by a professional healthcare personnel.

2.4. Processing of the high vaginal swab specimens and culture

The media preparation was done according to the manufacturer's instruction. It was then aseptically dispensed into Petri-dishes. Each swab was aseptically inoculated directly onto Nutrient Agar, Blood agar, MacConkey agar and MRS (de Man Rogosa Sharpe) agar. The agar plates were then incubated at 37 °C for 24 hours. Discrete colonies were further selected and subcultured onto petri dishes containing freshly prepared Nutrient agar using streak plate method to obtain pure cultures. The pure cultures were stored on Nutrient agar slants in Bijou bottles at 4 °C until ready for biochemical tests and identification [11].

2.5. Processing of the garlic

The collected bulbs were washed with distilled water and were allowed to dry at 60 °C for 7 days using a laboratory oven, after which the cloves were grinded into powdery form, using a grinder and were kept in a cool and dry area in the refrigerator until it was required for use. Ten (10) grams of the powder was mixed with 100 ml of distilled water in a sterile conical flask and left for 3 days with intermittent shaking. The mixtures were filtered using filter paper and concentrated in water bath at 70 °C for 3 hours. The extract was kept in a sterile container and refrigerated at 4 °C for further experiment [12].

2.6. Identification of the isolate

The isolates were identified using standard methods such as; Gram staining, Citrate utilization and Sugar fermentation test [13], Catalase test [14], Coagulase test [15], Urease and Motility test [16] and Molecular characterization [17].

2.7. Phytochemical analysis

Qualitative phytochemical analysis of garlic and Tea tree oil [18] [19] was carried out to detect the presence or absence of their bioactive components.

2.8. *In vitro* evaluation of the antimicrobial activity

2.8.1. Preparation of stock solution

Stock solutions of the garlic was prepared by weighing out 2 g of each of the test agents using electronic weighing machine and dissolved in 10 ml of sterile water in test tubes to give a stock concentration of 200 mg/ml of the extract [20]. Tea tree oil was dissolved with 95 % ethanol. The desired concentration (5 %v/v) was obtained by dissolving 1 ml of tea tree oil in 19 ml of the ethanol. It was then stored in an airtight glass vial and stored in a cool, dark place to protect it from light and heat.

2.8.2. Preparation of 0.5 McFarland standards

A 0.5 McFarland standard was prepared by dissolving 0.5 g of barium chloride dihydrate ($BaCl_2 \cdot 2H_2O$) in 50 ml of distilled water to obtain 1 % barium chloride solution. Then 0.5 ml of concentrated sulfuric acid (H_2SO_4) was added to 49.5 ml of distilled water carefully to make a 1% (v/v) solution. About 0.05 ml (50 µL) of the 1 % barium chloride solution was then measured using a sterile pipette and 9.95 ml of the 1 % sulfuric acid solution was measured in another container. The 0.05 ml of barium chloride solution was then added to the 9.95 ml of sulfuric acid solution and mixed thoroughly for uniform turbidity. The mixture formed a fine precipitate of barium sulfate ($BaSO_4$), which causes the turbidity [21].

2.8.3. Sensitivity test using agar-well diffusion method

Sensitivity test was carried out on garlic and tea tree oil to determine their inhibitory properties towards the HVS bacterial isolates. The sensitivity test was conducted using agar-well diffusion method. Plates of Mueller Hinton Agar were aseptically prepared and left to gel. Each plate was seeded with 0.2 ml of the standardized inoculum and a sterile glass rod was used to spread the inoculum onto the plate surface. Using a 6 mm cork borer, 6 mm diameter wells were bored through the medium. Then, 0.2 ml of the test solutions of garlic and tea tree oil were then added into the wells. Sterile water and ethanol were used as negative controls for garlic and tea tree oil respectively. The experimental set up was then incubated at 37 °C for 24 hours and the zones of inhibition were measured after the incubation period. The effectiveness of the antimicrobial agent was indicated by the clear zones around the wells [11].

2.8.4. Determination of MIC and MBC using broth dilution method

From the stock concentration of 200 mg/ml of the test agents, various concentrations of the test agents were made in Nutrient broth by double fold serial dilution to obtain, 100 mg/ml, 50 mg/ml, 25 mg/ml and 12.5 mg/ml. Each dilution in a test-tube was inoculated with 0.5 ml of the broth culture of test isolates diluted to 0.5 McFarland standards. All the tubes were incubated at 37 °C for 24 hours. The lowest concentration showing no visible growth was recorded as the minimum inhibitory concentration (MIC) for each organism [22].

To determine the minimum bactericidal concentration (MBC), 0.2 ml was taken from concentrations where no growth was observed (based on the MIC results) and transferred to fresh, antimicrobial-free growth medium (Nutrient agar) incubated for 24 hours at 37 °C. After incubation, the lowest concentration at which no viable bacterial cells were observed (i.e., the concentration that kills the bacteria) was recorded as minimum bactericidal concentration (MBC) for each organism [22].

3. Results and discussion

In this study, a total of 75 bacterial isolates were obtained from high vaginal swabs, of which four bacterial species were identified and analysed: *Lactobacillus spp.*, *Escherichia coli*, *Staphylococcus aureus* and *Shigella flexneri*. The distribution of these isolates (Table 1) revealed significant insights into the microbial landscape associated with vaginal infections in the population studied. *E. coli* was the most prevalent isolate. This finding aligns with previous studies that have identified *E. coli* as a common uropathogen and a frequent cause of bacterial vaginosis and other genital infections [23]. The high prevalence of *E. coli* in this study highlights its role in vaginal infections, which could be attributed to its ability to colonize the vaginal environment and ascend from the rectal flora, a pathway well-documented in the literature [24]. *Lactobacillus* species are typically considered beneficial commensals in the vaginal microbiota, playing a crucial role in maintaining vaginal health by producing lactic acid, which lowers the pH and inhibits the growth of pathogenic bacteria [25]. However, their presence in this context could be indicative of a shift in the vaginal flora or a response to an underlying infection, which merits further investigation. The presence of *S. aureus* in vaginal swabs is of particular concern due to its association with toxic shock syndrome and its ability to produce various virulence factors, including toxins and biofilms [26]. The relatively high frequency of *S. aureus* in this study is consistent with findings from other research in similar settings, where *S. aureus* has been identified as a significant pathogen in vaginal and urinary tract infections [27]. Finally, *S. flexneri* was the least common isolate. Although *Shigella* species are primarily known for causing gastrointestinal infections, their presence in the vaginal swabs may reflect faecal contamination or an unusual pathogenic colonization of the vaginal environment [28]. The low prevalence of *S. flexneri* in this study is consistent with its generally rare occurrence in non-gastrointestinal infections.

Table 1 Frequency (%) distribution of bacterial isolates from high vaginal swabs

S/N	Isolates	Number of Isolates	Frequency (%)
1	*Lactobacillus* spp.	21	28
2	*Escherichia coli*	35	46.7
3	*Staphylococcus aureus*	14	18.7
4	*Shigella flexneri*	5	6.7
	Total	**75**	**100**

The qualitative phytochemical analysis of garlic presented in Table 2 revealed the presence of several bioactive compounds which are known for their therapeutic properties. Allicin, a sulfur-containing compound, is a primary active ingredient in garlic and is well-documented for its antibacterial and antifungal properties [6]. Flavonoids are known for their antioxidant activities and potential health benefits, including anti-inflammatory and antimicrobial effects [29]. The presence of reducing sugars in garlic may contribute to its potential health benefits, including its role in glucose regulation [30]. Saponins have been reported to possess antimicrobial properties and may play a role in garlic's overall therapeutic effects [31]. Tannins are known for their antimicrobial and astringent properties, which may enhance garlic's ability to inhibit microbial growth [32]. The analysis showed negative results for alkaloids, cardiac glycosides, phlobatannins and steroids. This is consistent with existing literature, where garlic is noted for its distinct profile of active compounds without the presence of these specific phytochemicals [33].

The qualitative phytochemical analysis of tea tree oil (TTO) identified the following phytochemicals (Table 3).Flavonoids in TTO contribute to its antioxidant and antimicrobial properties, which have been well documented in various studies [4]. Terpenoids, including terpinen-4-ol, are major components of TTO and are crucial for its antimicrobial activity. Terpinen-4-ol, in particular, has been shown to have potent antibacterial properties [5]. Terpinen-4-ol is one of the most significant constituents of TTO, responsible for much of its antimicrobial and therapeutic efficacy [4]. The analysis showed negative results for alkaloids, saponins and tannins which align with the known composition of TTO. These findings support the specificity of TTO's antimicrobial activity attributed primarily to terpenoids and related compounds [34].

Table 2 Qualitative phytochemical analysis of garlic

S/N	Qualitative analysis	Result
1	Alkaloids (Dragendorff test)	-
2	Allicin	+
3	Flavonoids	+
4	Cardiac glycosides (Keller-kiliani test)	-
5	Phlobatannins	-
6	Reducing sugar (fehling's test)	+
7	Saponins	+
8	Steroids	-
9	Tannins (Ferric chloride test)	+

Key:-= absent; + = present

Table 3 Qualitative phytochemical analysis of tea tree oil

S/N	Qualitative analysis	Result
1	Alkaloids (Dragendorff test)	-
2	Flavonoids (Aluminum chloride test)	+
3	Saponins (froth test)	-
4	Tannins (ferric chloride test)	-
5	Terpenoids (salkowski test)	+
6	Terpinen-4-ol	+

Key: -= absent; + = present

The microscopic and biochemical test results presented in Table 4 provide critical insights into the identification and characterization of bacterial isolates from high vaginal swabs. *Lactobacillus* spp. is known for its beneficial role in the vaginal microbiota, where it maintains a low pH and suppresses pathogen growth [25]. The negative catalase and coagulase tests are consistent with its classification as a non-pathogenic, Gram-positive rod [35]. The positive fermentation of multiple sugars supports its role in creating an acidic environment favourable for its growth and detrimental to potential pathogens [36]. *E. coli* are a common cause of urinary tract infections and bacterial vaginosis [23]. The positive catalase and indole tests, combined with motility and fermentation of a wide range of sugars, confirm its identity [37]. The negative citrate test is typical for most *E. coli* strains, aligning with its biochemical profile [38]. *S. aureus* is known for its pathogenic potential, including its role in toxic shock syndrome and other infections [26]. The positive coagulase test is a key identifying feature, along with the positive catalase test and the ability to ferment mannitol [39]. The negative citrate and indole tests are consistent with its biochemical profile [40]. *S. flexneri* is primarily associated with gastrointestinal infections but can be present in unusual sites due to faecal contamination [28]. Its profile of negative catalase and citrate tests, along with positive motility, aligns with known characteristics of *Shigella* species [41]. Overall, the biochemical profiles support the correct identification of these bacterial isolates and provide a basis for understanding their pathogenic potential.

Table 4 Microscopic and biochemical test of the isolates

S/N	Gram stain	Rod/Cocci	Catalase	Citrate test	Coagulase	Indole test	Urease test	Motility	Glucose	Fructose	Sucrose	Dextrose	Mannitol	Lactose	Maltose	D-xylose	Raffinose	Galactose	Bacterial Isolate
1	+	Rods	-	-	-	-	-	-	+	+	+	+	+	+	+	-	-	+	*Lactobacillus spp*
2	-	Rods	+	-	-	+	-	+	+	+	+	+	+	+	+	+	-	+	*Escherichia coli*
3	+	Cocci	+	-	+	-	-	-	+	+	+	+	+	+	+	-	-	+	*Staphylococcus aureus*
4	-	Rods	-	-	-	-	-	-	+	+	-	+	-	+	+	+	-	+	*Shigella flexneri*

Key:-= negative; + = positive

The antibacterial activities of garlic and tea tree oil against *Lactobacillus spp.* were evaluated by measuring the inhibition zone diameters at various concentrations. The results are summarized in Table 5. Garlic exhibited significant antibacterial activity against *Lactobacillus spp.* across all tested concentrations, with the largest inhibition zone at 200 mg/ml. This finding supports the well-documented antimicrobial properties of garlic, attributed primarily to its active compound, allicin [6]. The reduction in inhibition zone diameter with decreasing garlic concentration is consistent with the concentration-dependent nature of antimicrobial activity. At the lowest concentration of 12.5 mg/ml, garlic did not inhibit *Lactobacillus* spp., indicating that a minimum concentration is necessary to exert an antibacterial effect. TTO also demonstrated antibacterial activity, with the largest inhibition zone observed at 200 mg/ml. However, its inhibitory effect was less pronounced compared to garlic at the same concentration. This difference might be due to the lower concentration of active antimicrobial compounds such as terpinen-4-ol in tea tree oil compared to the allicin in garlic [4]. The inhibition zones decreased with lower concentrations, aligning with the expected trend where antimicrobial activity diminishes as the concentration decreases.

The observed results highlight garlic as a more potent antibacterial agent against *Lactobacillus spp.* compared to TTO, potentially due to the higher efficacy of its key components. These findings are consistent with previous studies that have reported the superior antibacterial activity of garlic over various essential oils, including TTO [34, 5]. However, the effectiveness of TTO should not be underestimated, as it still demonstrates significant antibacterial properties, making it a valuable alternative or supplementary antimicrobial agent.

Table 5 Inhibition zone diameters of garlic and tea tree oil against *Lactobacillus* spp

Concentration (mg/ml)	Garlic (mm)	Tea tree oil (mm)
200	23.00±0.71	17.00±0.00
100	21.00±0.35	13.25±0.35
50	16.00±0.28	10.00±0.57
25	11.00±0.00	8.00±0.00
12.5	-	-

Garlic showed strong antibacterial activity against *E. coli* (Table 6), with the largest inhibition zone measured at 200 mg/ml. This finding aligns with previous research indicating that garlic, particularly its active compound allicin, has significant antimicrobial properties against *E. coli* [6]. The inhibition observed at 12.5 mg/mL, although reduced, indicates that garlic retains some antibacterial activity even at lower concentrations. The inhibition zones for TTO decreased with concentration, similar to garlic, but the zones were generally slightly smaller. This result suggests that while tea tree oil is effective, it may be less potent than garlic in inhibiting *E. coli*. TTO's antimicrobial efficacy is often attributed to its key compounds such as terpinen-4-ol [4], but the comparative efficacy might be influenced by the concentration of these active components in the oil.

Table 6 Inhibition zone diameters of garlic and tea tree oil against *Escherichia coli*

Concentration (mg/ml)	Garlic (mm)	Tea tree oil (mm)
200	23.00±0.28	23.00±0.00
100	20.00±0.71	19.00±0.57
50	16.00±0.00	16.00±1.41
25	12.50±0.71	13.50±0.35
12.5	10.50±0.71	10.50±0.71

The IZD values recorded for *S. aureus* demonstrate the antibacterial effectiveness of both garlic and TTO, as observed in this study (Table 7). The comparison between the IZD values of garlic and TTO indicates that both substances are potent against *S. aureus*. However, the observed differences in their IZDs suggest variability in their mechanisms of action and efficacy. Garlic's higher or comparable IZD may be attributed to its unique ability to disrupt bacterial cell walls, leading to the leakage of cell contents, whereas tea tree oil likely disrupts the permeability of the bacterial cell membrane, causing cell death. These findings are consistent with prior research that highlights the potential of natural antimicrobial agents in combating bacterial infections, especially in the context of rising antibiotic resistance [42] [4]. The results underscore the relevance of garlic and tea tree oil as alternative or complementary therapies for infections caused by *S. aureus*.

Table 7 Inhibition zone diameters of garlic and tea tree oil against *Staphylococcus aureus*

Concentration (mg/ml)	Garlic (mm)	Tea tree oil (mm)
200	23.00±0.71	25.00±0.78
100	20.00±1.41	23.00±1.41
50	15.50±0.35	19.00±0.00
25	13.00±1.41	17.50±0.71
12.5	11.00±0.71	14.50±0.35

The results of the IZD observed in Table 8 showed that both garlic and tea tree oil effectively inhibited *S. flexneri*, with TTO showing slightly higher inhibition at the highest concentration. This supports the use of both agents as effective treatments for infections caused by *S. flexneri*. The results agree with existing literature on the antimicrobial properties of garlic and tea tree oil, which have been reported to possess broad-spectrum antibacterial effects [34] [5]. The slight superiority of tea tree oil in this instance reinforces its potential as an effective alternative or supplementary antimicrobial agent.

Table 8 Inhibition zone diameters of garlic and tea tree oil against *Shigella flexneri*

Concentration (mg/ml)	Garlic (mm)	Tea tree oil (mm)
200	21.50±0.71	22.00±0.28
100	18.00±0.14	17.00±0.71
50	14.00±0.71	15.00±0.42
25	10.00±0.57	11.25±0.35
12.5	9.50±0.35	9.00±0.00

The MIC values for garlic and tea tree oil against the bacterial isolates (Table 9) revealed that Garlic exhibited variable MIC values across the different bacterial isolates. The lowest MIC of 12.5 mg/ml was observed against *S. aureus*, indicating that garlic is highly effective against this pathogen. This result agrees with the well-documented potent antimicrobial activity of garlic, particularly due to its active compound allicin, which has been shown to inhibit Gram-positive bacteria effectively [6]. The MIC for *E. coli* was slightly higher at 25 mg/ml and for *Lactobacillus* spp. and *S.*

flexneri, the MIC was 50 mg/ml, suggesting that garlic is less effective against these bacteria compared to *S. aureus*. These findings align with previous studies that report garlic's variable effectiveness against different bacterial strains [34]. TTO showed a broader range of MIC values. The MIC for *E. coli* and *S. aureus* was 12.5 mg/ml, indicating high efficacy against these pathogens. The lower MIC values for TTO compared to garlic suggest that TTO might be more effective against these specific bacteria, consistent with the strong antimicrobial properties attributed to its major components such as terpinen-4-ol [4]. The highest MIC of 100 mg/ml was recorded for *Lactobacillus* spp., suggesting TTO is less effective against this bacterium compared to others.

Table 9 MIC (mg/ml) Determination of garlic and tea tree oil against the isolate

Antimicrobial agent	***Lactobacillus* spp**	***Escherichia coli***	***Staphyococcus aureus***	***Shigella flexneri***
Garlic	50	25	12.5	50
Tea tree oil	100	12.5	12.5	25

In the result depicted in Table 10, it was observed that garlic showed a high MBC for *S. aureus* and *S. flexneri*, indicating that garlic requires higher concentrations to achieve bactericidal effects compared to other antimicrobial agents [6]. The MBC for *E. coli* and for *Lactobacillus* spp., were at higher concentrations, reflecting that garlic is less effective as a bactericidal agent at lower concentrations against these bacteria. These results are consistent with garlic's known variable bactericidal efficacy depending on the bacterial strain [34]. TTO exhibited lower MBC values compared to garlic, indicating more effective bactericidal activity. The MBC for *S. aureus* was 25 mg/ml, the lowest among the tested bacteria, which underscores TTO's potent bactericidal activity against this pathogen [4]. The MBC for *E. coli* indicated a stronger bactericidal effect than garlic. The MBC for *S. flexneri* and *Lactobacillus* spp. demonstrated that TTO is also effective at higher concentrations against these bacteria. These findings are similar with the literature that highlights TTO's broad-spectrum antimicrobial properties and its efficacy at relatively lower concentrations compared to other agents [5]. In summary, both garlic and tea tree oil exhibited bactericidal activity across the different bacterial isolates, with tea tree oil generally demonstrating lower MBC values and hence greater bactericidal potency. These results support the use of both antimicrobial agents, with tea tree oil showing superior effectiveness in achieving bactericidal activity at lower concentrations.

Table 10 MBC (mg/ml) Determination of garlic and tea tree oil against the isolates

Antimicrobial agent	***Lactobacillus* spp**	***Escherichia coli***	***Staphylococcus aureus***	***Shigella flexneri***
Garlic	200	100	50	200
Tea tree oil	200	50	25	100

4. Conclusion

The findings of this study revealed that both garlic and tea tree oil exhibited significant antimicrobial properties, though their effectiveness varied by bacterial strain and concentration. Both garlic and tea tree oil present viable options for antimicrobial therapy, each with unique advantages. Garlic remains a potent natural antimicrobial agent, especially against Gram-positive bacteria, while tea tree oil offers broad-spectrum activity and effectiveness at lower concentrations. These findings support further exploration into the therapeutic applications of these phytochemicals, suggesting their potential roles in developing natural antimicrobial treatments.

Compliance with ethical standards

Disclosure of conflict of interest

No conflict of interest to be disclosed.

Statement of ethical approval

Ethical clearance was gotten from Chukwuemeka Odumegwu Ojukwu University Teaching Hospital, Awka, Anambra State, Nigeria.

Statement of informed consent

Informed consent was obtained from all individual participants included in the study.

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