

Antibacterial Activity of Crude Leaf Extracts from Selected Medicinal Plants against *Shigella Flexineri*

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Abstract

This study aimed at determining the effects of the selected medicinal plants (*Tagetes minuta*, *Aloe secundiflora*, *Vernonia lasiopopus*, and *Bulbine frutescens*) extracts against Clinical isolate of *Shigella flexineri*. *Shigella flexineri* is a gram-negative bacterium that is associated with gastrointestinal disturbances leading to diarrhoea in human beings. The antibacterial activity of the medicinal plant extracts against *Shigella flexineri* was determined using the Kirby Bauer method. The extracts showed antimicrobial activity against *Shigella flexineri* with *Bulbine frutescens* extract (minimum inhibitory concentration 3.2 µg/ml; maximum bactericidal concentration 6.2 µg/ml) being the most active when compared to the others. *Tagetes minuta* (minimum inhibitory concentration 7.4 µg/ml; maximum bactericidal concentration 12.6 µg/ml) extract was less active when compared to the other extracts. *Bulbine frutescens* had the largest average zone of inhibition 19.50 ± 1.05 mm while *Vernonia lasiopopus* and *Aloe secundiflora* had the least zone of inhibition of 18.17 ± 1.47 mm both. Ciprofloxacin (5µg/ml) was used as a positive control producing an average zone of inhibition of 22 ± 1.84 mm while negative controls (water and dimethyl sulphoxide) showed no zone of inhibition. The preliminary qualitative screening for phytochemical showed the presence; of saponins, tannins, alkaloids, and flavonoids. The study provides insight into the antibacterial activity of the medicinal plant extracts and if they can be used in the treatment of infections caused by *Shigella flexineri* as an antibacterial agent.

Keywords: *Shigella flexineri*; Phytochemicals; Kirby Bauer; Medicinal plants; Zone of inhibition

Introduction

Medicinal plants are used by almost 80% of the world's population for their basic health care because of their low cost and ease of availability. From the dawn of civilization, people have developed a great interest in plant-based drugs and pharmaceutical products [1]. In the last few decades, many bacterial organisms have continued to show increasing resistance against current antimicrobial agents [2]. Herbal drugs made from medicinal plants have been used from ancient times to treat various diseases and their antimicrobial properties make them a rich source of many potent drugs [3]. The use of herbal medicinal plants has always played a positive role in the control or prevention of diseases such as diabetes, heart disorders, and various cancers [4]. Some medicinal plants have been used in the production of various drugs singly or combination and even as principal raw material for the production of other conventional medicines [5].

The genus *Tagetes* belongs to the Asteraceae family which presently comprises 56 species, 27 biennials, and 29 perennials. *Tagetes* species are grown all over the world as multipurpose plants. The most common species are *Tagetes minuta*, *Tagetes patula*, *Tagetes erecta*, and *Tagetes tenuifolia* [6]. *Tagetes* species and chemotypes from its genus have been largely examined for biologically active metabolites that can be used in industry and medicine [7]. Compounds that have antimicrobial activity in the *Tagetes minuta* plant are said to be accumulated in the organs of the plant and their essential oils have not only antimicrobial effects but also insecticidal properties [8]. Plant parts such as flowers and leaves have been known to contain flavonoids that are scavengers for free radicals which enhances the antimicrobial activity of the *Tagetes minuta* extracts [9]. Some of the *Tagetes minuta* phytochemicals from the plant such as carotenoids have also been used in pharmacological preparations and they have been found to contain anti-aging and anticancer effects [10]. The plant extracts have been used in treating intestinal and stomach problems [11-12]. *Tagetes minuta* extracts such as its volatile oil and other components have been used in the flavoring of food products and as perfumes. The plant has also shown inhibitory

activity against some pathogens and insects. Studies carried out have shown that leaf extracts from most of the *Tagetes* species including *Tagetes minuta* contain flavonoids that have shown antimicrobial potential against both Gram-positive and Gram-negative bacteria. Extracts from *Tagetes minuta* leaf flowers and stem extracted using methanol have been shown to contain secondary metabolites including terpenes which are thought to be responsible for antibacterial activities. Aloes are perennial succulent xerophytes that develop water storage tissues in leaves to survive in areas with low or erratic rainfall [13]. The plant is mainly found in cultivation, having no naturally occurring population although closely related Aloes do grow in northern parts of Africa [14]. The plant is an almost sessile perennial herb that has leaves 30-50 centimetres long and 10 centimetres broad at the base, bright yellow tubular flowers 25-35centimetres in length arranged in a slender loose spike [15]. The genus *Aloe* is common in Kenya; with about 60 taxa recognized [16]. *Aloe secundiflora* has been used in treating ailments including; chest problems, polio, malaria, and stomach ache by herbalists in the Lake Victoria region [17]. *Aloe secundiflora* leaf components have been credited for antibacterial, antifungal antiviral, and anthelmintic medicinal properties [18]. *Aloe* extracts have been used for many centuries for their curative and therapeutic properties. *Aloe* products have also been used in pharmaceuticals, cosmetic, and food industries [19]. *Bulbine* is a genus of plants in the family Xanthorrhoeaceae and subfamily asphodeloideae and its members are well known for their medicinal value [20]. *Bulbine frutescens* wild

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and *Bulbine natalensis* baker is the most common species known [21]. The *Bulbine* plant has been used for medicinal purposes in the early stages of the 18th century by Dutch and British settlers of South Africa in treating various ailments [22]. Many species have bulb-shaped tubers. It's chiefly found in South Africa with a few species extending to the tropics of Africa and Australia [23]. They are succulent plants with most of the species having yellow flowers whereas some of them have white, orange, or pink flowers. *Bulbine frutescens* is mostly grown as an ornamental plant in the flower garden at homes in South Africa [24]. The leaves of the plant have been used in the treatment of wounds thought to be infected with bacterial pathogens and it has shown antibacterial properties [25]. Some of the species of the plant found in South Africa have been used for blood cleansing, treatment of ringworms, and gravel rush by some local communities such as the Xhosa [26]. A decoction of bulbs and roots of some of the species has been used in the treatment of some of the venereal diseases in women and stomach upsets [28]. *Vernoniaeae* is a tribe that has about 1300 species and is in the family *Asteraceae* (*Compositae*) which mostly contains herbaceous plants [29]. *Vernonia* shrubs grow in tropical Africa and have a height of about 2-5 metres, elliptical leaves of up to 20 centimetres, and a rough bark [31]. The plants in this genus usually have a bitter taste and in English, they are called bitter leaf [32]. Some of the common African names of plants in this genus are *Olusia* (Luo), *Mululuza* (Luganda), *Onugu* (Igbo), *Grawa* (Amharic), and *Chusar-Doki* (Hausa) [33]. *Vernonia lasiopos* decoctions from the stems and leaves have been traditionally been used by herbalists in East Africa to treat, malaria, worms, and gastrointestinal problems [34]. In the Kikuyu community, it's traditionally known as *Mucatha* and it has been used in treating diarrhoea problems [35]. Studies carried out have shown some of the phytochemical components found in its extracts have antimicrobial capability [36]. Its extracts have also been used in treating some of the sexually transmitted diseases in southern parts of Africa [37]. In North America, some of the species of the genus *Vernonia* such as *Vernonia altissima*, *Vernonia fasciculata*, and *Vernonia flaccidifolia* have been found to contain effective properties for them to be used as blood purifiers, uterus toner, and also contain sesquiterpene lactone which can also help in preventing atherosclerosis. In Brazil, *Vernonia condesata* commonly known to locals as *necroton* or *figatil* has been used in traditional medicine to treat analgesic, anti-thermal, anti-anemic, anti-inflammatory and as an antibacterial agent. *Shigella* is a genus of Gram-negative, rod-shaped facultative bacteria responsible for shigellosis. Only a few cells of the bacteria can cause infections and its clinical manifestations include exothermic reactions, diarrhea, abdominal pain and sometimes vomiting. Shigellosis epidemic tends to occur in developing countries due to poor sanitation and the transmission rate from one person to another is more frequent especially when it's due to water or food contamination. Shigellosis is responsible for most of the diarrhea episodes and causes approximately over one million deaths annually [38]. Over time the increase in the use of conventionally produced antimicrobials against *Shigella flexineri* has led to the development of resistance. The increase in resistance has led to the need for an alternative to the antimicrobials produced leading to the need to extensively study extracts from medicinal plants with antimicrobial activities [39].

Materials and Methods

Plant material collection

The fresh plant leaves of *Aloe secundiflora*, *Bulbine frutescens*, *Vernonia lasiopos*, and *Tagetes minuta* were collected at Kenyatta University Arboretum. Voucher specimens were prepared and

deposited in the university herbarium in Plant Sciences Department for future reference. The plant's leaves were brought to the laboratory and thoroughly washed in running water to remove debris and dust particles and then rinsed using distilled water and finally air-dried.

Preparation of plant extract

The air-dried plants leaves were ground into powder soaked in methanol for 72 hours and placed in a Gallenkamp shaker at 65 revolutions per minute. The contents were homogenized and filtered using Whatman filter paper no. 1. The filtrate was poured into a round bottom flask concentrated using a vacuum evaporator and stored in a labeled amber glass bottle at room temperature away from light and heat before being used for an antibacterial efficacy test.

Preparation of Media

The media used was Muller Hinton agar and it was prepared according to commercially given instructions.

Preparation of Muller-Hinton agar

38 milligram of Muller-Hinton agar powder was added into one liter of distilled water in a flat-bottomed conical flask. The mixture was heated with frequent agitation and boiled for one minute to completely dissolve the media. The flask was then tightly closed using cotton wool and further covered with aluminum foil. The mixture was autoclaved for 15 minutes at 121 °C after which it was left to cool down to room temperature. The media was poured into the Petri dishes in a laminar flow to give a uniform depth of 3-4 millimeters. The Petri dishes containing the media were then placed in a sterile plastic bag and stored at a temperature of 2-8 °C before use.

Test bacterial organism

The microorganism used was a clinical isolate of *Shigella flexineri* obtained from Kenyatta University Health Centre Laboratory, Nairobi. The isolate was tested against methanolic leaf extracts from *Tagetes minuta*, *Aloe secundiflora*, *Bulbine frutescens*, and *Vernonia lasiopos*.

Antimicrobial susceptibility testing

The microorganism used was a clinical isolate of *Shigella flexineri* obtained from Kenyatta University Health Centre Laboratory, Nairobi. *Shigella flexineri* was tested against methanol extracts of *Tagetes minuta*, *Aloe secundiflora*, *Bulbine frutescens*, and *Vernonia lasiopos*. *Shigella flexineri* inoculum was concentrated by comparing it with a 0.5 McFarland standard. Discs of 6 milliliters were prepared from Whatman no.1 filter paper. The discs were sterilized by autoclaving. After sterilization, the moisture discs were dried on a hot air oven at 50 °C [40]. The various solvent extracts discs prepared were impregnated with the extracts from the highest concentration of 1000 µg/ml to the lowest concentration of 1 µg/ml. The antimicrobial efficacy test was carried out using the Kirby Bauer method [41]. Muller Hinton agar was used in the spread plate technique where *Shigella flexineri* was spread using a sterilized cotton wool swab and exposed to extracts impregnated discs in milligrams per microliter from *Aloe secundiflora*, *Tagetes minuta*, *Vernonia lasiopos*, and *Bulbine frutescens*. The discs were placed with equal distance between them on agar plates inoculated with *Shigella flexineri*. Positive control discs contained ciprofloxacin while negative control discs were impregnated with dimethyl sulphoxide and distilled water. The Petri dishes were incubated at 37°C for 24 hr. Zones of inhibition formed were measured in milli meters and their average determined. The experiment was carried out in duplicates and the diameter of zones of inhibition formed was measured.

Minimal inhibitory concentration (MIC) and Minimum bactericidal concentration (MBC) determination

Minimal inhibitory concentration (MIC) was determined using the broth tube method, [43]. 100µl of 250 µg/ml of methanol extract was added to 100µl of sterile bacteriological peptone in the first well of the 96 well microplates and mixed well with a micropipette. 100µl of this dilution was transferred subsequently to wells two folding each dilution of the original extract. This was done to the extracts of Aloe secundiflora, Bulbine frutescens, Vernonia lasiopos, and Tagetes minuta. An inoculum of 100µl (0.5 McFarland standard) of an overnight clinical culture of *Shigella flexineri* was added in each of the wells. Triplicates of each microplate were made and the procedure was repeated. The plates were then incubated at 37°C for 24 hrs. After incubation 40µl of 0.2 mg/µl of INT were added in each of the wells and the plates were examined after an additional 60 minutes of incubation. Growth was indicated by a red colour (conversion of INT to formazan). The lowest concentration at which the colour was invisible as compared to the next dilution was taken as the minimum inhibitory concentration [44]. Minimum bactericidal concentration (MBC) was determined by taking 100µl of suspension from micro plate wells that demonstrated no growth and inoculated on agar plates. The plates were incubated at 37°C for 24 hrs. In the case where there was no bacterial growth and value not greater than the minimum inhibitory concentration, the concentration was used as the maximum bacterial concentration.

Phytochemical analysis

The presence of saponins, tannins, flavonoids, and alkaloids in the crude extract was determined.

Tannins: Each of the extracts were weighed to 0.5mg and dissolved in 1 ml of distilled water. Filtration was carried out after 2ml of FeCl₃ was added. If there was the presence of a blue or black precipitate then it indicated the presence of tannins.

Flavonoids: Each of the extracts was weighed to 0.5mg dissolved in 1 ml of ethanol and filtered. 2ml of 1% HCL and magnesium ribbon was added to the filtrate. If there was the formation of a pink or red colour it indicated the presence of flavonoids.

Alkaloids: Each of the extracts was weighed to 0.5mg dissolved in 1ml of methanol and filtered. 1% HCL was added to the filtrate and the solution heated. Mayor's reagent was added drop wise and if there was the formation of any colored precipitate it indicated the presence of alkaloids.

Saponins: Each of the extracts was weighed to 0.5mg dissolved in 1 ml of methanol and filtered. Distilled water was added and shaking was done for a few minutes. If there was persistence frothing then it indicated the presence of saponins.

Data Analysis

The data were expressed as means and standard deviations. SAS version 19.0 package was utilized in conducting ANOVA test to determine significant differences in antimicrobial activity of selected plant extracts against *Shigella flexineri*. Two-way ANOVA was carried out to determine if there was any interaction between the plant extracts and *Shigella flexineri*. $P \leq 0.05$ is considered significant. Turkey's posthoc was utilized to assess the difference within individual means of the zones of inhibition. A P -value ≤ 0.05 was considered statistically significant.

Results

The extracts from *Tagetes minuta*, *Aloe secundiflora*, *Bulbine frutescens*, and *Vernonia lasiopos* all showed significant antimicrobial activity when tested against the clinical isolate of *Shigella flexineri*. The extract from *Bulbine frutescens* was more active against *Shigella flexineri* as compared to others producing the highest average zone of inhibition (19.50 ± 1.05 mm). *Aloe secundiflora* and *Vernonia lasiopos* extract were the least active both producing an average zone of inhibition of 18.17 ± 1.47 mm. The extract from *Tagetes minuta* also showed a considerable antimicrobial activity producing a zone of inhibition of 19.00 ± 1.41 , Ciprofloxacin antibiotic discs used as positive control produced a zone of inhibition of 22.00 ± 1.84 mm. The negative controls of dimethyl sulphoxide (DMSO) did not produce any zones of inhibition (0.0 ± 0.00). The results showed that *Bulbine frutescens* produced the highest zone of inhibition hence it was a more potent antibacterial agent when compared to the other plant extracts (Table 1).

The value of average zones of inhibition \pm standard error after one-way ANOVA followed by Tukey's HSD test. A value followed by the same superscript within the same column is not significantly different ($P > 0.05$).

Key: DMSO - Dimethyl sulphoxide, Plant extracts concentration (1000µg/ml) Antibiotic standard discs of; Ciprofloxacin (5µg/ml) (+ve control).

The analysis of the interaction between the plant extracts and *shigella flexineri* showed that the average zone of inhibition formed by the plant's extracts when used against *shigella flexineri* was significantly different from those formed by antibiotic, methanol, and DMSO ($P < 0.05$; (Table 2). Moreover, zones formed by *Bulbine frutescens* extract against *shigella flexineri* was significantly different from those formed by the other plant extracts (17.19 ± 0.42 mm) ($P < 0.05$) Table 2. The interaction between *shigella flexineri* and the plant extracts was significant in Table 2. The value of average zones of inhibition \pm standard error of the mean (SEM) after two-way ANOVA was followed by Tukey's HSD test. A value followed by the same superscript within the same column is not significantly different ($P > 0.05$).

Key: Antibiotic (ciprofloxacin), DMSO - dimethyl sulphoxide

When the plant extracts were used in low concentrations against *Shigella flexineri*, *Bulbine frutescens* was the most active with a minimum inhibitory concentration (MIC) of 3.2µg/ml and maximum bactericidal concentration (MBC) of 6.2µg/ml. *Tagetes minuta* extract was the less active; MIC of 7.4µg/ml and MBC of 12.6µg/ml in comparison to all other plant extracts. The other two extracts, *Vernonia lasiopos*, and *Aloe secundiflora* also had a pronounced antimicrobial activity against *Shigella flexineri* with; MIC of 3.3µg/ml and 3.7µg/ml; MBC of 7.1µg/ml and 8.0µg/ml respectively (Figure 1).

Table 1: Efficacy test of the plant's leaf extracts against *Shigella flexineri*.

| Plant extracts | Zone of Inhibition (mm) |
|---------------------------|-------------------------|
| <i>Tagetes minuta</i> | 19.00 ± 1.41^{bc} |
| <i>Aloe secundiflora</i> | 18.17 ± 1.47^c |
| <i>Bulbine frutescens</i> | 19.50 ± 1.05^{bc} |
| <i>Vernonia lasiopos</i> | 18.17 ± 1.47^c |
| Controls | Zone of inhibition (mm) |
| Ciprofloxacin | 22 ± 1.8^b |
| Methanol | 31.67 ± 2.88^a |
| 4% (DMSO) | 0.0 ± 0.00^d |
| <i>P value</i> | 0.0001 |

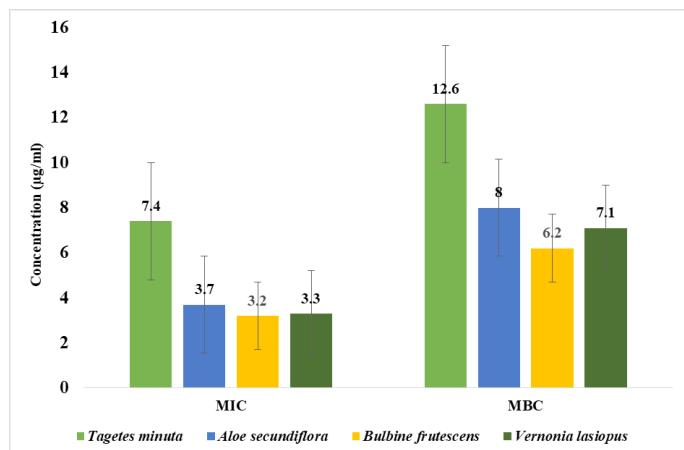


Figure 1: Minimum inhibitory concentration (MIC) and Maximum bactericidal concentration (MBC) of plant extracts against *Shigella flexineri*.

Table 2: Interactions between the plants extract and *Shigella flexineri*.

| Extract | Zone of inhibition±SEM |
|---|----------------------------|
| <i>Aloe secundiflora</i> | 16.69 ± 0.40 ^{cd} |
| <i>Bulbine frutescens</i> | 17.19 ± 0.42 ^c |
| <i>Tagetes minuta</i> | 16.06 ± 0.56 ^d |
| <i>Vernonia lasiopos</i> | 15.81 ± 0.61 ^d |
| Controls | Zone of inhibition±SEM |
| Antibiotic | 23.5 ± 0.43 ^b |
| Methanol | 31.17 ± 0.34 ^a |
| DMSO | 0.00 ± 0.00 ^e |
| Test microorganism | Zone of inhibition±SEM |
| <i>Shigella flexineri</i> | 18.69 ± 1.45 ^a |
| P values of the main factors and their interactions | |
| Extract | <0.001 |
| Test microorganism | <0.001 |
| Extract*Test microorganism | <0.001 |

The plants extract from *Tagetes minuta*, *Aloe secundiflora*, *Bulbine frutescens*, and *Vernonia lasiopos*, when qualitatively analyzed for the presence of phytochemicals showed the presence of saponins, alkaloid, tannins, and flavonoids (Table 3).

Key: (+) present

Discussion

The increase of antimicrobial resistance has led to the need for the invention of new antimicrobial agents. The use of plant extracts to test for antimicrobial activity has been brought forward as one of the ways of achieving this goal. The plants used in the study have been said to be of medicinal value. This study evaluated the use of the plant's leaf extracts in treating *Shigella flexineri* and tested if they are effective or not. Furthermore, qualitative analysis to test for the phytochemical's presence was also done. This is because phytochemicals have been said to be responsible for some of the antimicrobial activity by extracts from plants with medicinal value. From the findings, the leaf extracts from the plants were found to have antimicrobial activity when used against *Shigella flexineri*.

Antimicrobial activity

Medicinal plant extracts from various studies have shown that plants from similar genera with *Aloe secundiflora* have been shown to have antimicrobial activity. In this study, the antimicrobial activity

Table 3: Phytochemical tests on the plant extracts.

| Name of test | Plants leaf extracts | | | |
|------------------|----------------------|-----------------------|---------------------|-------------------|
| | <i>T.minuta</i> | <i>A.secondiflora</i> | <i>B.frutescens</i> | <i>V.lasiopus</i> |
| Saponins test | + | + | + | + |
| Tannin's test | + | + | + | + |
| Alkaloid's test | + | + | + | + |
| Flavonoid's test | + | + | + | + |

of leaf extracts from medicinal plants was tested against *Shigella flexineri*. It was interesting to note that the leaf extract from *Aloe secundiflora* showed antimicrobial activity against *Shigella flexineri*. Findings from the study were similar to those obtained in Nigeria [45] from activities against medicinal plants. The leaf extract from *Aloe secundiflora* had antimicrobial activity against *Shigella flexineri* and other bacterial pathogens such as; *Escherichia coli*, *Salmonella typhi*, *Staphylococcus aureus*, and *Enterococcus faecalis*. The findings were similar to those obtained in a study carried out on Gram-negative and Gram-positive bacteria in India and England. These findings were also similar to those obtained in a previously carried out study in Kenya [46] which found, methanol extracts of *Aloe secundiflora* along the lake region in Kenya to be effective against *Shigella flexineri* and other bacterial pathogens such as *Escherichia coli*, *Salmonella typhi*, and *Staphylococcus aureus* among others. The leaf extract from *Tagetes minuta* showed antimicrobial activity against *Shigella flexineri*. The findings were similar to a previously carried out study in Pakistan who found out that extracts from *Tagetes minuta* had antimicrobial activity against not only *Shigella flexineri* but also other bacterial organisms such as *Salmonella typhi*, *Escherichia coli*, and *Staphylococcus aureus*. Furthermore, the minimum inhibitory concentration against these test microorganisms ranged from 4 to 100µg/ml, the average zones of inhibition formed were ≥ 17.00mm and the standard antibiotic used (Ciprofloxacin) produced zones of inhibition ≥ 20.00mm. This means that if the concentration of *Tagetes minuta* could be standardized, it could be used as an alternative therapy for Ciprofloxacin, against *Shigella flexineri*. *Bulbine frutescens* extract showed antimicrobial activity against *Shigella flexineri*. Its antimicrobial activity was significantly higher when compared to the other plant extracts. This finding was contrary to the one in South Africa [47] who found out that, extract from *Bulbine frutescens* had no antimicrobial activity against gram-negative bacterial pathogens. These differences could be due to the different geographical and environmental conditions during the growth of the plant, the method of extraction used, and the age of the plant. The extract from *Vernonia lasiopos* showed antimicrobial activity against *Shigella flexineri*. The findings concurred with those previously obtained in Kenya who found out that leaf extracts from *Vernonia lasiopos* had antimicrobial activity against gram-negative bacterial pathogens.

Phytochemicals

The extract from *Aloe secundiflora* showed that the plant leaf contained pharmacologically active components. The extract contained flavonoids, saponins, alkaloids, and tannins which may be responsible for the antimicrobial activity. Similar studies previously carried out have shown that some of the pharmacologically active components have antimicrobial activity. These findings concurred with a previously carried out study in Kenya who carried out qualitative analysis of phytochemical components of *Aloe secundiflora* extract used against bacterial and fungal pathogenic microbes from a plant collected along the Kenya lake region found it too contained tannins, saponins, flavonoids, and alkaloids. Furthermore, similar findings were

obtained in a study carried out in India who confirmed the presence of flavonoids, saponins, tannins, and alkaloids in Aloe extract. Qualitative analysis for the presence of phytochemicals in *Tagetes minuta* extract showed the presence of flavonoids, saponins, alkaloids, and tannins. This pharmacologically active component might be responsible for *Tagetes minuta* extract antimicrobial activity against *Shigella flexineri*. The study findings were similar to those obtained in a study carried out in Pakistan in Argentina who also confirmed the presence of phytochemicals flavonoids, saponins, tannins, and alkaloids in *Tagetes minuta* extract used against Gram-positive and Gram-negative bacteria. The extract from *Bulbine frutescens* contained pharmacologically active compounds namely saponins, tannins, alkaloids, and saponins which could be responsible for antimicrobial activity. This study concurred with those of a previously done study in South Africa by [48] who found out the presence of alkaloids, saponins, tannins, and flavonoids in plant extract from *Bulbine frutescens*. However, the findings were contrary to those obtained in a previously carried out study in South Africa by who found out that, the extract from *Bulbine frutescens* did not contain any of the four phytochemicals. This could be due to diverse plant metabolites associated with a geographical and ecological difference from where the plant was obtained and also the age of the plant used. The extract from *Vernonia lasioporus* had active pharmacological compounds; flavonoids, saponins, tannins, and alkaloids which could be responsible for the antimicrobial activity. These findings were similar to those of a previously carried out study by who found out that, extract from *Vernonia lasioporus* contained alkaloids, flavonoids, saponins, and tannins when used against Gram-positive and Gram-negative bacteria. In Nigeria also found out that extract from the plant from the family *Vernonia* contained flavonoids, alkaloids, tannins, and saponins. However, the findings were contrary to those obtained in a study carried out in the Southwestern region in Nigeria by who found out the extract from *Vernonia lasioporus* did not contain alkaloids. This difference may be associated with the geographical and environmental factors of the area from which the plant was collected.

Conclusion

All the plant's leaf extracts showed significant antibacterial action against *Shigella flexineri*. This demonstrates that the extracts from the plants have the potential to be employed as an antibacterial agent against *Shigella flexineri* and related bacterial pathogens of the nature (Gram-negative). There is also a requirement to purify the plant extracts further to identify the key bioactive components of phytochemicals that are responsible for this antibacterial action. This will help in the supply of a natural source for treating illness caused by a bacterial pathogen (gram-negative) and others of its kind which have been gradually gaining resistance against commercially available antibiotics.

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Ethics approval

The ethical approval for the use of clinical isolates of *Shigella flexineri* obtained from Kenyatta University Health Centre Laboratory was obtained from the university research and ethical approval committee.

Declaration

I declare that this manuscript submitted for publication is from my original research work. All the sections in this manuscript, the concept, and the argument developed by other authors have been referenced to show the material has been used to support my manuscript.

Consent of publication

I hereby give consent for the publication of this manuscript because it's my original work and it does not contain any specific individual data.

Availability of data and Material

The data and material can only be shared on request by the author but it depends on the reasons for such request being clearly articulated pending a decision of acceptance or rejection from the respondent author.

Competing interests

The authors declare there are no competing interests.

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Author's contributions

Opinde H.R. – Research Student.

Nyamache A.K. – Research Supervisor and Advisor.

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Authors' information

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