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Phytochemical Screening and Antibacterial Activity Evaluation of Seed Oils from *Tylosema esculentum*, *Schinziophyton rautanenii*, *Bauhinia petersiana*, and *Citrullus lanatus* (Thunb.) Mansf. in Botswana

Mpho Granny Batlhophi[1], Freddy Bwacha[1], Barati Sandi Phirinyane[2], Olekile Tibe[1], Emang Molojwane[1], Rosemary Ikalafeng Kobue-Lekalake[2], Ompelege Keolopile Matenanga[2], Bonno Sekwati-Monang[2*], Geremew Bultosa[2], Eyassu Seifu[2], Moenyane Molapisi[2], John Gwamba[2], Kethabile Sonno[2], Gaone Mokhawa[2], Tsaone Phakama[2], Modiri Dirisca Setlhoka[2], Gulelat Desse Haki[2]

[1]Department of Biological Sciences and Physical and Chemical Sciences, Botswana University of Agriculture and Natural Resources, Gaborone, Botswana
[2]Department of Food Science and Technology, University of Agriculture and Natural Resources, Gaborone, Botswana
Email: *bmonang@buan.ac.bw

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Abstract

Antimicrobial Resistance (AMR) poses a global health threat, necessitating exploration of natural alternatives such as plant-derived compounds. In Botswana, indigenous seed oils remain underutilised due to limited data on antimicrobial potential. This study aimed to evaluate the phytochemical composition and antibacterial activity of seed oils from four underutilised plants: *Schinziophyton rautanenii* (*mongongo or manketti*), *Tylosema esculentum* (*morama*), *Bauhinia petersiana* (*mogose*), and *Citrullus lanatus* (Thunb.) Mansf (*kgwengwe*). Seeds were collected from three districts in Botswana, from which seed oils were prepared for antibacterial activity. Qualitative analysis for phytoconstituents was conducted through standard assay tests. Antibacterial activity against *S. aureus* ATCC 25923 and *E. coli* ATCC 25922 was assessed using the EUCAST-guided disk diffusion method, with tests performed in triplicate. Data were analysed using one-way ANOVA followed by Tukey's HSD and Student's t-tests ($p \leq 0.05$). Phytochemical analysis revealed the presence of terpenoids in three oils, *T. esulentum*, *S. rautanenii*, and *B. petersiana*, with steroids in *C. lanatus* (Thunb.) Mansf. and saponins in *S. rautanenii*. All seed oils exhibited antibacterial activity against gram-negative *S. aureus*, with inhibition zones of

16.0 ± 0.00 (*B. petersiana*), 9.33 ± 0.58 (*T. esculentum*), 7.00 ± 0.00 (*S. rautanenii*), and 6.47 ± 0.06 (*C. lanatus* (Thunb.) Mansf.), showing significant differences ($p < 0.001$). *C. lanatus* (Thunb.) Mansf. yielded the lowest antibacterial activity. No activity was observed against gram-negative *E. coli*. Positive controls (penicillin, gentamicin, tetracycline) confirmed the assay validity. These findings highlight the potential of these seed oils, particularly *B. petersiana*, as suitable plant-based antibacterial against gram-positive pathogens like *S. aureus*, a WHO high-priority AMR bacterium. Future studies should isolate active compounds, determine Minimum Inhibitory Concentration (MIC)/Minimum Bactericidal Concentration (MBC) values, evaluate efficacy against resistant strains and other foodborne pathogens, and explore optimised extraction methods to enhance yields and applications in food preservation and pharmaceuticals.

Keywords

Antibacterial Activity, *E. coli*, Methanol, Seed-Oils, *S. aureus*, Under-Utilized

1. Introduction

Seed oils contain phytochemicals with applications in food industries [1] [2], perfumery where their aromatic compounds enhance the flavour and fragrance profiles [3], and they have significant health benefits and potential therapeutic properties owing to their bioactive compounds, especially terpenoids, flavonoids, and alkaloids, which have antimicrobial properties [4]. Given the increase in antibiotic resistance, phytochemistry offers promising avenues for alternative therapeutic compounds to combat infectious diseases, with significant benefits in pharmacology and antimicrobial drug development. Global health challenges underscore the need for accessible and effective therapies, particularly in developing nations where medical resources may be limited [5]. Accordingly, WHO recommends plant-derived medicines [6], because of the antimicrobial properties of their organic compounds [7]. In Botswana, traditional plant-based medicines play a central role in healthcare [8]. However, with the rise in antibiotic-resistant microbial life forms, the value of plants with medical significance and commercial applications needs to be enhanced using research-based information streams.

In Southern Africa, local communities rely on *B. petersiana*, *C. lanatus* (Thunb.) Mansf., *S. rautanenii*, and *T. esculentum* for food and medicine. For example, *T. esculentum*, known as the morama bean in Botswana (English name gemsbok bean), is a member of the Fabaceae family, which produces pod seeds and bears tubers used in medicine by traditional healers [9]. Some people use it to combat drug-resistant infections, as an anti-inflammatory food component, and to lessen gastrointestinal illnesses of viral and bacterial origin [10]. *C. lanatus* (Thunb.) Mansf. or kgwengwe (Tswana) is a non-woody climbing vine used by indigenous people in Botswana as a remedy for urinary tract infections [11]. Although *C. lanatus*

(Thunb.) Mansf. has been implicated in improving human health, but it remains largely uncultivated. *S. rautanenii* is a large deciduous tree, commonly known as mongongo or manketti, and belongs to the Euphorbiaceae family. The nuts of *S. rautanenii* are rich in oil and are consumed in Namibia. Various parts of the plant are traditionally used for medicinal purposes, such as the treatment of fungal dermatological conditions, stomach discomfort, and measles [12]. *B. petersiana* or Kalahari camel foot plant, locally termed mogose, belongs to the family Fabaceae. It is rarely used for therapeutic purposes, except for the management of colds, sterility, and menstrual discomfort in the South African traditional health environment [13]. In Botswana, *B. petersiana* seeds are generally consumed as pickled relishes or baked into porridge meal powder [14].

Recent ethnobotanical studies have documented the food value and medicinal uses of various parts of *T. esculentum*, *S. rautanenii*, *B. petersiana*, and *C. lanatus* (Thunb.) Mansf. [15]. Unfortunately, most users are uninformed about the exact nutrients and antimicrobial components of plants. The practical understanding of the medical use of plants and the skills required to prepare concoctions are typically left to traditional healers, who retain the curative knowledge of the plants [9]. Existing research is primarily centred on the phytochemical and antimicrobial activities of extracts from sources other than plant seed oils. Therefore, antimicrobial compounds in seed oils remain medically underutilized [15] because of limited research on their efficacy against food-contaminating bacteria. Hence, the study was conducted to bridge this gap by examining the phytochemical compounds in seed oils of four economically important, edible, and medicinal plants native to Botswana and assessing their antibacterial properties against *S. aureus* (ATCC 25923) and *E. coli* (ATCC 25922). *E. coli* and *S. aureus* are among the World Health Organization (WHO) priority pathogens for Antimicrobial Resistance (AMR), classified under the critical and high priority categories, respectively [16]. In Sub-Saharan Africa, including Botswana, a large proportion of the population relies on traditional herbal medicine, with many reporting tangible health benefits from such remedies [17]. Building on this ethnopharmacological relevance, we hypothesize that the selected four seed oils contain bioactive phytochemicals with antibacterial activity against *S. aureus* and *E. coli*. The findings of this study could contribute to the development of sustainable, plant-based antimicrobial agents and promote community-driven commercialization models that preserve and valorise indigenous knowledge systems.

2. Materials and Methods

2.1. Sample Collection and Preparation

The sampling areas (**Figure 1**) are within Shakawe village, Northwest District (18.3673˚S, 21.8390˚E), Malwelwe village, Kweneng District (23.9871˚S, 25.2487˚E), and Lonetree village, Ghanzi District (20˚27'0"S, 24˚52'60"E).

Seeds were collected at the end of the rainy season (March-May). The fruits (seedpods) for *S. rautanenii*, *T. esculentum*, *B. petersiana*, and *C. lanatus* (Thunb.)

Mansf melons were harvested directly from the plants. The melons and the seedpods were transported to the laboratory for identification (using an identification key) by a Botanist at Botswana University of Agriculture and Natural Resources. Mongongo seeds were de-shelled in a conventional hand-forging way. Hard coat of morama seeds was removed using a rock-crushing process. Mogose seedpods were sun-dried until they opened naturally, ejecting their seeds. Kgwengwe seeds were removed from the melons and sundried. The dried, clean seeds were uniformly mashed using a mortar and pestle and then placed in an airtight Ziploc polyethylene bag. The prepared samples were kept at 4˚C - 7˚C in a refrigerator until further use (Table 1).

Table 1. Details of the plants from which the oils were extracted.

Plant species	Vernacular name	Family name	Place of harvest (Village district)
Tylosema esculentum	Morama	Fabaceae	Malwelwe (Kweneng)
Schinziophyton rautanenii	Mongongo	Euphorbiaceae	Shakawe (Northwest)
Bauhinia petersiana	Mogose	Fabaceae	Malwelwe (Kweneng)
Citrullus lanatus (Thunb.) Mansf.	Kgwengwe	Cucurbitaceae	Lonetree (Ghanzi)

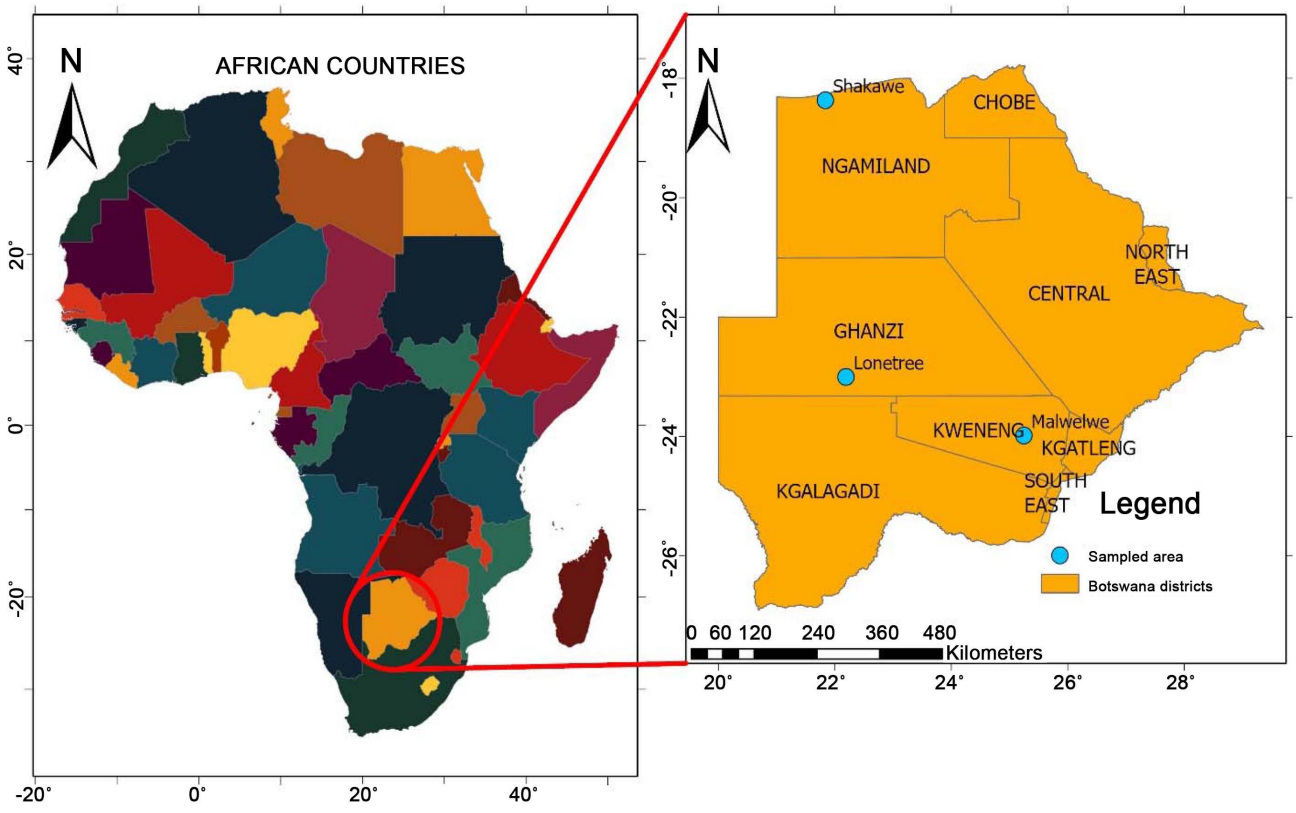

Figure 1. Map of Botswana showing the sampling areas. (Source: ArcGIS Pro (Esri, Redlands, CA, USA))

2.2. Sample Extract Preparation

The extraction of seed oil used for phytochemical profiling was conducted with a Soxhlet extractor for 8 h at 60 ˚C per cycle, using 50 mL methanol (99%, Lab Alley, LC/MS Grade) as solvent. The oils were obtained after the solvent was removed under reduced temperature and pressure using a Heidolph Hei-VAP Expert control rotatory evaporator (Walpersdorfer Str. 12, 91126 Schwabach, Germany), with refluxing at 70˚C to remove any excess solvent used for oil extraction. The extracted seed oils were stored in an airtight glass bottle in a refrigerator freezer at 2˚C until used for analyses [15].

2.3. Phytochemical Qualitative Tests

Qualitative screening tests for phytochemicals were performed on the four seed oil samples, and all tests were performed in triplicate.

2.3.1. Terpenoids (Salkowski Test)

An extract (0.5 ml) was mixed with 2 ml of chloroform, followed by 3 ml of concentrated sulfuric acid to form a layer. The formation of a reddish-brown coloration at the interface indicated the presence of terpenoids [18].

2.3.2. Steroids (Salkowski's Test)

One millimetre of the extract was dissolved in 10 ml of chloroform, and an equal volume of concentrated sulfuric acid (H_2SO_4) was slowly added to the side of the test tube. A change in the upper layer to a red colour and the H_2SO_4 layer to a yellow colour with green fluorescence was indicative of the presence of steroids [19].

2.3.3. Flavonoids (Ammonia Test)

Two millimetres of the extract was filtered, and then 5 ml of dilute ammonia and 1 ml of concentrated H_2SO_4 were added slowly. The development of a yellow colour that disappears during standing indicates the presence of flavonoids [20].

2.3.4. Tannins (Lead acetate Test)

Three drops of 1% lead acetate were added to 2 mL of extract. The formation of a yellowish precipitate indicated the presence of tannins [21].

2.3.5. Coumarins (Alkaline Reagent)

Three millimetres of 10% NaOH was added to 2 ml of the extract, and a yellow colour indicated the presence of coumarins [20].

2.3.6. Saponins (Foam Test)

Five millimetres of distilled water was added to 2 ml of the extract, and the mixture was shaken vigorously. The formation of stable persistent foam indicates the presence of saponins [19].

2.3.7. Oxalates

A few drops of glacial ethanoic acid were added to 3 ml of extract. The presence

of greenish-black coloration is evidence of oxalates [19].

2.3.8. Phenols (Ferric Chloride Test)

2 ml of the extract was treated with 5% aqueous ferric chloride, and the formation of a deep blue or black colour indicated the presence of phenols [19].

2.4. Antibacterial Activity Testing

Seed oil used for antibacterial activity testing was extracted by cold maceration using ground seed (10 g) in 50 mL of methanol (99%, Lab Alley, LC/MS Grade) and filtered after incubation in an incubator shaker at 25˚C for 24 h. The supernatants were stored in a refrigerator, after which the retentates were reextracted. Residual methanol was removed from each supernatant using Heidolph™ Hei-VAP Expert control rotatory evaporator (Walpersdorfer Str. 12, 91126, Schwabach, Germany), and each undiluted seed oil extract was used to impregnate sterile blank antimicrobial susceptibility disks under sterile conditions. The disks were left to dry for six days at room temperature, and once completely dry, antimicrobial tests were performed. In addition to considering the toxicity of the solvent in the biological assay [22], the oils were extracted using methanol because most antimicrobial agents are aromatic hydrocarbons (termed arenes), which dissolve easily in either methanol or ethanol [23]. Methanol's polarity is considered dominant over ethanol; thus, methanol establishes a higher strength in intermolecular hydrogen bonding, acting as an amphoteric molecule concerning donating and accepting hydrogen [24].

2.5. Test Organism

The antibacterial activity of seed oils was tested against *Staphylococcus aureus* ATCC 25923 (gram-positive) and *Escherichia coli* ATCC 25922 (gram-negative) purchased from Caymen Heights Pty (Ltd.), Botswana. The cultures were incubated at 37˚C for 18 h, and the purity of the cultures was checked after 18 h of incubation. Bacterial strains were maintained on nutrient agar slants at 4˚C before use.

2.6. Antibacterial Assay

The antibacterial activity of the four seed oils was evaluated using the modified disk diffusion method [25] [26]. Bacterial strains, *S. aureus* ATCC 25923 and *E. coli* ATCC were grown in 100 mL Mueller-Hinton broth (Oxoid, UK) at 37˚C for 18 h with shaking (150 rpm) in an incubator shaker. The overnight cultures were adjusted with saline solution to a turbidity equivalent to 0.5 McFarland standard ($\approx 10^5$ CFU/mL) using a BioSan DEN-1 densitometer with externally powered LED light source. Whatman antibiotic blank assay discs (6 mm diameter) were immersed in 2 mL 100% seed oils overnight and air-dried under sterile conditions. The discs were then placed on Mueller-Hinton agar (4 mm, thickness) previously inoculated by evenly swabbing the bacterial suspension ($\approx 10^5$ CFU/mL) over the agar surface, using an Oxoid antimicrobial susceptibility disc dispenser, and gen-

tly pressed with sterile forceps to ensure full contact with the agar surface. Solvent-only discs served as negative controls, while penicillin (10 μg), tetracycline (30 μg), and gentamicin (10 μg) discs were used as positive controls. All tests were performed in triplicate. The plates were left at room temperature for 30 min to allow diffusion of the oils into the agar and incubated at 37˚C for 18 h. The diameter of the inhibition zones was measured and recorded to assess the antibacterial activity. The mean inhibition zone diameters were interpreted according to CLSI guidelines.

2.7. Statistical Analysis

Triplicate measurements of inhibition zones were performed following CLSI guidelines (M02 for disk diffusion testing). Data were analysed using Microsoft Excel's Data Analysis Toolpak (Windows version). Results are reported as mean ± standard deviation, coefficient of variation (CV, %), and 95% Confidence Intervals (CI). To assess differences among groups (for example, different antibiotics or oils), one-way ANOVA was conducted, and significant results ($p \leq 0.05$) were followed by post hoc pairwise comparisons using Tukey HSD and Student's t-tests with Bonferroni correction to control for multiple comparisons. A p-value ≤ 0.05 (two-tailed) was considered statistically significant.

3. Results

Qualitative phytochemical screening using methanol as an extraction solvent was performed on four seed oils to identify potential bioactive compounds before antibacterial testing. Methanol was preferred based on its higher solute-elution strength, enabling a quicker and more effective extraction process [27]. Methanol is a popular choice in compositional analysis for the extraction of different bioactive substances, including terpenoids, flavonoids, and phenols [27]. The results indicate the presence of three phytochemical substances: steroids, saponins, and terpenoids. While three of the seed oils (*S. rautanenii*, *B. petersiana*, and *T. esculentum*) contained terpenoids, the seed oil of *S. rautanenii* also contained saponins with *C. lanatus* (Thunb.) Mansf. containing only steroids (**Table 2**). The presence of steroids, saponins, and terpenoids in a seed oil can potentially influence the antibacterial activity of the seed oil extract. According to Olasehinde *et al.* [28], a plant oil extract containing phytochemicals, steroids, saponins, and terpenoids is of medicinal importance, showing antibacterial activity against *S. aureus* and *E. coli*.

Table 2. Qualitative test for secondary metabolites (phytochemicals) in four methanol-extracted seed oils.

S/N	Secondary product	*T. esculentum*	*S. rautanenii*	*B. petersiana*	*C. lanatus* (Thunb.) Mansf.
1	Steroids	-	-	-	+
2	Terpenoids	+	+	+	-

Continued

3	Tannins	-	-	-	-
4	Flavonoids	-	-	-	-
5	Oxalate	-	-	-	-
6	Saponins	-	+	-	-
7	Coumarins	-	-	-	-
8	Phenols	-	-	-	-

+ presence of phytochemicals and - represents absence of phytochemicals in seed oil.

The antibacterial activity of four seed oils was evaluated against *S. aureus* and *E. coli* using the disc diffusion method (Table 3). All seed oils inhibited *S. aureus*, but none showed activity against *E. coli*. One-way ANOVA revealed significant differences in antibacterial activity against *S. aureus* among the seed oils ($P < 0.001$). Post-hoc analysis using Tukey's HSD and Student's t-tests with Bonferroni correction identified significant pairwise differences (denoted a-e in Table 3). Replicate variability was low (0.89% - 6.19%), with narrow confidence intervals, indicating high precision. *B. petersiana* showed the highest activity against *S. aureus* (16 ± 0.0 mm), although this was lower than antibiotic controls. In contrast, the seed oils were inactive against *E. coli*, while antibiotics showed significant inhibition ($P < 0.001$). Tetracycline produced the largest inhibition zones against *S. aureus* (36.67 ± 0.58 mm) and *E. coli* (25 ± 0.00 mm).

Table 3. Antibacterial activity of seed oils and antibiotic controls against *S. aureus* and *E. coli*.

	S. aureus			*E. coli*		
Sample	Mean ± SD (mm)	CV (%)	95% CI	Mean ± SD (mm)	CV (%)	95% CI
Seed oils						
T. esculentum	9.33 ± 0.58^{b}	6.19	7.90, 10.77	0.00^{a}	N/A	
B. petersiana	16.00 ± 0.00^{c}	0.00	16.00, 16.00	0.00^{a}	N/A	
S. rautanenii	7.00 ± 0.00^{a}	0.00	7.00, 7.00	0.00^{a}	N/A	
C. lanatus Thunb. Mansf.	6.47 ± 0.06^{a}	0.89	6.32, 6.61	0.00^{a}	N/A	
Antibiotic (μg/mL)						
Tetracycline (30)	36.67 ± 0.58^{d}	1.57	35.23, 38.10	25.00 ± 0.00^{c}	0.00	25.00, 25.00
Penicillin (10)	27.33 ± 0.58^{e}	2.11	25.90, 28.77	NT		
Gentamicin (10)	NT			23.30 ± 0.58^{b}	2.47	21.90, 24.77

Values represent the mean inhibition zones (mm) from three replicates per sample. SD: standard deviation; CV: Coefficient of Variation; 95% CI: Confidence Interval; NT: Not Tested; N/A: not applicable because of zero means. For *S. aureus*, means ± SD with different superscript letters (a-e) differ significantly ($P < 0.05$). For *E. coli*, statistical analysis was performed only for antibiotics Gentamicin and Tetracycline due to zero means in seed oils.

4. Discussion

The aim of conducting the study was to analyse the phytochemical compounds in

seed oils of *S. rautanenii*, *T. esculentum*, *B. petersiana*, and *C. lanatus* (Thunb.) Mansf. and to evaluate their antibacterial activity against *S. aureus* and *E. coli*. *S. aureus* and *E. coli* are key species implicated in food contamination and pose a significant threat to human health [29], for example, carbapenem resistant *E. coli* has been reported to harbour colistin resistance gene (*mrc*-1), and colistin is considered the last resort for treating multidrug-resistant bacteria, creating strains that approach pan resistance—a global health crisis [30]. Therefore, *E. coli* and *S. aureus* are classified among the critical and high-priority groups of pathogens for the development of new antibacterial agents [31]. To address the escalating threat of pan-resistance, calls for intensive research to discover novel antibacterial agents and strategic global interventions to stop the spread of resistance. Our study identified terpenoids, steroids, and saponin substances with diverse biological activities, such as antibacterial activity, making the seed oils promising candidates as antimicrobial agents. Terpenoids were detected in the seed oils of *S. rautanensis*, *T. esculentum*, and *B. petersiana*, but were absent in *C. lanatus* (Thunb.) Mansf. (**Table 2**). Phenols were not detected in our study, although a similar study involving *T. esculentum* seed bean oil reported the presence of phenolic acids [32], which are carboxylic acid-containing types of phenols [33]. Variations in extraction time and temperature may be the cause of the discrepancy; for example, extracts high in phenol are susceptible to degradation when exposed to light and oxygen-rich conditions for long periods [34]. We also did not find tannins, coumarins, saponins, and flavonoids in *B. petersiana* seed oil although reported in the plant [14]. Our results also differ from other research that identified additional phytochemicals in the four seed oils analysed [15]. Discrepancies in phytochemical profiles may arise from variations in environmental factors such as temperature and soil composition, solvent selection, extraction and drying methods, duration of extraction, or extract concentration [35]. While the absence of phenols and other bioactive compounds in our seed oils may limit their antibacterial activity, the presence of terpenoids, steroids, and saponins warrants investigations into their antibacterial potential against *E. coli* and *S. aureus*. Research indicates that plants synthesize terpenoids for microbicidal purposes through secondary metabolic pathways [36]-[38], and constitute the largest group of specialized metabolites, characterized by diverse properties and varying chemical compositions [39]. Terpenoids are among the primary families of antibacterial compounds that combat various pathogens due to their lipophilic properties. By easily permeating the phospholipid molecules that shield microbial cells, terpenoids break down their cell membrane [40]. Similarly, steroids from *Ulva fasciata* and *Bersama abyssinica* have shown efficacy against *S. aureus* [41] [42], whereas saponins enhance cell wall permeability by interacting with membrane components [43] [44], an effect amplified when combined with antibiotics [45]. However, there is no direct comparison of the observed bioactivity concerning saponins and steroids of the seed oils; to our knowledge, we are the first to evaluate the antibacterial activity of the seed oils.

The antibacterial evaluation demonstrated that all seed oils exhibited varying degrees of antimicrobial activity against *S. aureus* but showed no activity against *E. coli*. Among the seed oils, *B. petersiana* oil displayed the highest activity against *S. aureus* (16.00 ± 0.00 mm), significantly outperforming *T. esculentum* (9.33 ± 0.58 mm), *S. rautanenii* (7.00 ± 0.00 mm), and *C. lanatus* (Thunb.) Mansf. (6.47 ± 0.06 mm) ($P < 0.05$). However, these oils were less effective than the positive controls: penicillin (36.67 ± 0.58 mm) and tetracycline (27.33 ± 0.58 mm), which is expected given the optimized mechanisms of antibiotics targeting bacterial cell wall synthesis and protein production. The lack of activity against *E. coli* is consistent with the known resistance of gram-negative bacteria to many natural compounds, likely due to their outer membrane, which restricts the penetration of hydrophobic molecules present in seed oils. This resistance is attributed to the hydrophilic Lipopolysaccharide (LPS) layer in gram-negative bacteria, which impedes the entry of lipophilic compounds such as terpenoids, unlike the more vulnerable gram-positive *S. aureus* [46]-[48]. However, some studies have reported the activity of terpenoids against *E. coli* when combined with antibiotics or extracted from other plant species [28] [49] [50], suggesting that the inefficacy observed in our study may reflect limitations in the chemical composition or extraction method of the seed oils.

Our results support the potential of *B. petersiana* as a candidate for further investigation of natural antimicrobial agents, although its efficacy remains lower than that of conventional antibiotics. Future studies should explore the active components of *B. petersiana* (phenolic compounds or fatty acids) and test modified formulations to enhance their activity against gram-negative pathogens. Additionally, the limited testing of antibiotics (Penicillin not tested against *E. coli* and Gentamicin not tested against *S. aureus*) restricts direct comparisons across all treatments, warranting further investigation. To overcome these limitations, future research should focus on determining the Minimum Inhibitory Concentration (MIC) and Minimum Bactericidal Concentration (MBC) of the oils, following the CLSI guidelines, to assess their bacteriostatic or bactericidal potential [51]. Exploring alternative extraction methods, such as combining solvents to enhance the yield and diversity of bioactive compounds, could also improve efficacy [27]. Moreover, testing the seed oils against a broader range of foodborne pathogens, such as *Campylobacter jejuni*, *Salmonella spp.*, *Listeria monocytogenes*, and *Bacillus cereus*, would provide a more comprehensive understanding of their antimicrobial spectra [52]. Characterizing the specific secondary metabolites responsible for the observed effects, elucidating their mechanisms of action, and evaluating their toxicity and therapeutic dosages across age groups are critical next steps for practical applications in food preservation and pharmaceutical development.

5. Conclusion

Phytochemical screening of seed oils from *S. rautanenii*, *T. esculentum*, *B. peter-*

siana, and *C. lanatus* (Thunb.) Mansf. revealed the presence of key secondary metabolites, saponins, steroids, and terpenoids, known for their antimicrobial potential. These bioactive compounds likely contributed to the observed antibacterial activity against *S. aureus*, while no activity was observed against *E. coli*. The differential sensitivity between *S. aureus* and *E. coli* highlights the structural barriers of gram-negative bacteria and underscores the importance of testing across diverse microbial groups. Notably, *B. petersiana* exhibited the highest efficacy against *S. aureus*, suggesting its potential as a potent natural antimicrobial agent. To advance the practical use of these seed oils, further research is needed to isolate and characterise the specific bioactive compounds responsible for the observed effects, determine their MIC and MBC, and assess the synergetic effects with conventional antibiotics. Comprehensive toxicological and clinical studies are also necessary to establish safety and efficacy profiles for potential therapeutic use. Expanding research to include multiple extraction techniques and a wider range of pathogens will enhance understanding of these indigenous oils as suitable plant-based antimicrobial agents.

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Conflicts of Interest

All authors declared that they have no conflict of interest.

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