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Phytochemical Screening and Selected Biological Activities of *Solanum incanum* L. Fruit Extracts

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Abstract:

Numerous studies have documented the use of the fruit of *Solanum incanum* in traditional medicine. This study aims to investigate the phytochemical composition, antioxidant capacity, and antimicrobial efficacy of *S. incanum* fruit extracts. Methanolic and aqueous extracts were prepared using a rotary shaker method. Phytochemical screening revealed the presence of alkaloids, saponins, phenols, tannins, flavonoids, steroids, and carbonyl compounds in the methanol extract. In contrast, the aqueous extract possessed alkaloids, saponins, phenols, tannins, flavonoids, and carbonyl compounds. Fourier-transform infrared (FT-IR) spectroscopy analysis of the methanol extract indicated the presence of hydroxyl (–OH), aromatic/alkene (C–H), carbonyl (C=O), C=C, and C–O functional groups. However, in the aqueous extract, the FT-IR spectra showed the presence of hydroxyl (–OH), aliphatic C–H (methane group), carbonyl (C=O), amine (N–H), and C–O groups. The antioxidant activity of both extracts was assessed using the DPPH (2, 2-diphenyl-1-picrylhydrazyl) radical scavenging assay. The methanol extract exhibited significantly higher antiradical activity compared to the aqueous extract, indicating greater antioxidant potential. Antimicrobial activity was evaluated using the agar well diffusion method against three human bacterial pathogens—*Staphylococcus aureus*, *Escherichia coli*, and *Pseudomonas aeruginosa*—and one fungal pathogen, *Candida albicans*. The aqueous extract demonstrated superior antimicrobial activity against all tested pathogens, followed by the methanol extract. The antimicrobial efficacy of the aqueous extract is probably attributable to the presence of bioactive antimicrobial peptides (AMPs), which are essential components of the innate immune defense system in many organisms.

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INTRODUCTION

Solanaceae is a flowering plant family of great economic and medicinal importance. It comprises 98 genera and approximately 2,700 species that range from annual or perennial herbs to shrubs and trees. These species are widely distributed across all continents except Antarctica (Nath *et al.*, 2017; Afroz *et al*., 2020). Among them are vital cash crops such as Potatoes (*Solanum tuberosum*), Tomatoes (*S. lycopersicum*), Eggplants (*S. melongena*), and Peppers (*Capsicum annuum*) (Ghatak *et al.*, 2017; Afroz *et al.*, 2020). Additionally, the family includes several significant medicinal plants that have played an important role since the early stages of drug discovery, especially genera like *Atropa* L., *Hyoscyamus* L., *Withania* Pauquy, *Capsicum* L., and *Nicotiana* L., which continue to hold importance in traditional herbal medicine systems such as Ayurveda and Traditional Chinese Medicine (Shah *et al.,* 2013; Chowanski *et al.*, 2016; Afroz *et al.*, 2020). Members of the Solanaceae family are rich in bioactive secondary metabolites, including alkaloids, flavonoids, glycosides, lactones, lignans, steroids, phenols, sugars, terpenoids, and antimicrobial peptides (AMPs). These compounds contribute notably to their antimicrobial properties (Afroz *et al.*, 2020).

In Yemen, the Solanaceae family is represented by 38 species across 10 genera, including the genus *Solanum* L., which comprises 15 species (Al Khulaidi, 2013). Among these, *Solanum incanum* L. which is the most common species, growing in a variety of habitats across the country (Al-Hubaishi and Müller-Hohenstein, 1984; Wood, 1997; Ebad *et al.*, 2006) and widely used in traditional Yemeni medicine (Al Dubaie and Al Khulaidi, 2005).

Previous studies have explored the antimicrobial effects of methanol and aqueous extracts from various parts of *S. incanum*, including the stem, leaves, and fruits. These investigations identified several bioactive secondary metabolites. Almoulah et al. (2017) reported that methanolic leaf extracts of *S. incanum* exhibited antibacterial activity against both Gram-positive bacteria, such as *Staphylococcus aureus* and *Bacillus subtilis*, as well as Gram-negative bacteria, including *Escherichia coli* and *Pseudomonas aeruginosa*. Moreover, Kipngeno et al. (2014) found that crude methanol extracts of *S. incanum* fruit contained phytochemical components (alkaloids, flavonoids, tannins, phenols, and saponins), that exhibit an antimicrobial activity against a strain of *Staphylococcus* and *Trichophyton rubrum*. However, Sahle and Okbatinsae (2017) analyzed the phytochemical composition of *S. incanum* ethanol and aqueous fruit extracts, they discovered that the ethanol extract contained carbohydrates, proteins, alkaloids, phenols, flavonoids, glycosides, steroids, triterpenes, and tannins, while the aqueous extract contained similar compounds, with saponins as a primary component. Both extracts exhibited significant antimicrobial activity against *E. coli*, *Salmonella typhimurium*, and *Candida albicans*.

In addition, Jepkoech and Gakunga (2017) reported the presence of alkaloids, flavonoids, cardiac glycosides, steroids, and triterpenes in both methanol and aqueous extracts, which showed antimicrobial activity against *S. aureus*. Furthermore, Indhumathi et al. (2014) demonstrated that ethanol extracts of *S. incanum* fruit had antibacterial activity against both Gram-positive (*S. aureus*, *B. subtilis*) and Gram-negative (*P. aeruginosa*, *S. paratyphi*, and *Vibrio cholerae*) bacteria. Additionally, Séré et al. (2022) illustrated that both acetone and water extracts of *S. incanum* possessed antibacterial activity against *Pasteurella* species.

Earlier studies have mainly focused on the antimicrobial effects of various extracts; however, they have not elucidated the reasons underlying the inhibitory activity of the aqueous extract against Gram-positive and Gram-negative bacteria, as well as fungal pathogens. Therefore, this paper attempts to explain and discuss the antimicrobial activity of *S. incanum* fruit aqueous extract against bacterial (gram-positive and gram-negative) and fungal pathogens by investigating the functional groups

in *S. incanum* fruit water and methanol extracts, and combining it with the antimicrobial effect of the extracts.

MATERIALS AND METHODS

Collection, Classification, and Preparation of plant material for extraction

During the period 15/3/2025 to 15/4/2025, plant specimens and ripe fruit samples of *S. incanum* (Fig.1) were collected from Sana'a University, New Campus (latitude: 15°21'52.14"N, longitude: 44°10'57.51"E, altitude: 2265 m above sea level) located in Sana'a Governorate, Yemen. The plant specimens were identified and classified using available taxonomical references, specifically Wood (1997) and Al-Khulaidi (2013). Three plant samples were assigned herbarium voucher numbers (BHSS 2001 and BHSS 2045) and deposited in the Herbarium at the Faculty of Science, Sana'a University, for future reference. The collected fruits were thoroughly washed with distilled water, cut into pieces, and air-dried in a shaded area for three weeks. After drying, the fruits were ground into a fine powder using a blender. The powdered fruits were stored in a refrigerator in airtight cellophane containers as stock samples until required for further analysis (Chirchir *et al.*, 2014; Ibrahim *et al.*, 2023b).

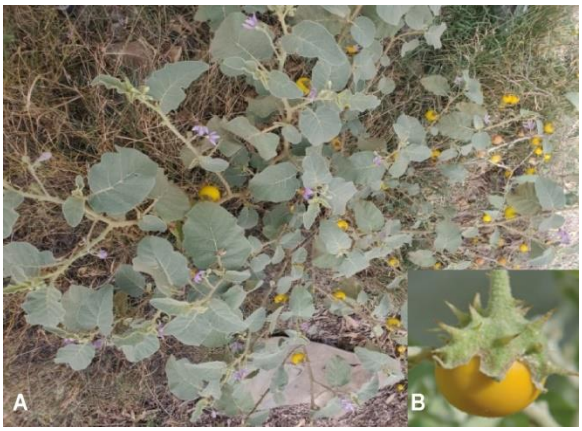

Fig. 1. *Solanum incanum* L.: A. General view, B. Ripe Fruit.

Preparation of plant extracts

Methanol and aqueous (water) fruit extracts were prepared independently; by employing the methodology Illustrated by Ibrahim et al. (2023a) (after modification), 30g of air-dried fruit powder was extracted by 300 ml of methanol and distilled water (individually) using the rotary shaker method. The extracts were filtered using Whatman filter paper, the supernatant of each extract was collected, then solvents were evaporated to obtain the final volume of *S. incanum* fruit methanol and aqueous crude extracts independently, extracts were kept in a sealed container at 4°C for further investigation.

Phytochemical Screening

The methanolic and aqueous extracts of *S. incanum* fruit were examined through qualitative phytochemical screening to identify the presence of various bioactive compounds. Standard phytochemical tests, as outlined by Venkatesan et al. (2009) and Ibrahim et al. (2022), were employed to detect alkaloids, saponins, phenols, tannins, flavonoids, steroids, and carbonyl compounds. The screening procedures for each phytochemical group were as follows:

Determination of Alkaloids

To detect the presence of alkaloids in the *S. incanum* fruit extracts, 0.2 g of each extract was individually mixed with 2% sulfuric acid in a test tube. The mixture was gently warmed for 2 minutes and then filtered. A few drops of Dragendorff's reagent were added to the filtrate. The formation of an orange-red precipitate indicated the presence of alkaloids in the extract.

Determination of Saponins

To examine the presence of saponins, 0.2 g of each *S. incanum* fruit extracts was separately mixed with 5 ml of distilled water in a test tube. The mixture was then heated to boiling. The formation of a stable, creamy froth or mass of small bubbles indicated the presence of saponins in the extract.

Determination of Phenols and Tannins

To determine the presence of phenols and tannins in the fruit extracts of *S. incanum*, a small amount of each extract was individually mixed with distilled water and heated in a water bath. After heating, the mixture was filtered, and the resulting filtrate was treated with a few drops of ferric chloride ($FeCl_3$) solution. The appearance of a dark green coloration indicated the presence of phenols and tannins compounds in the extract**.**

Determination of Flavonoids

To examine the presence of flavonoids, 0.2 g of each *S. incanum* fruit extract was separately dissolved in a diluted Sodium hydroxide solution. If the mixture turns colorless upon the addition of diluted Hydrochloric acid, it indicates the presence of flavonoids in the extract.

Determination of Steroids

To detect the presence of steroids in the fruit extracts of *S. incanum*, 2 mg of each extract was independently heated with acetic anhydride until it reached boiling. After boiling, 1 ml of concentrated sulfuric acid was carefully added. A color change from violet to blue or green indicates the presence of steroids in the extract.

Determination of Carbonyl Groups

To determine the presence of carbonyl groups in *S. incanum* fruit extracts, 0.2 g of each extract was independently treated with 2 ml of 2,4-Dinitrophenylhydrazine (2,4-DNPH) reagent. The formation of yellow crystals indicates the presence of carbonyl groups in the extract.

Determination of Chemical Functional Groups by Fourier Transform Infrared (FT-IR) analysis

Functional groups, presented in the methanol and aqueous extracts of *S. incanum* fruit were identified separately using IR spectra. A portion powder from each crude extract was mixed with Potassium Bromide (KBr) salt (individually) using a mortar and pestle. This mixture was then pressed into a thin pellet with a KBr press. The pellets were placed in the Perkin-Elmer FT-IR spectrometer 410 compartment and scanned across the IR range from 4000 to 400 cm^{-1} (Chen *et al.*, 2008; Ibrahim *et al.*, 2023b).

Assessment of Antioxidant Activity in *S. incanum* Fruit Extracts via 1, 1-Diphenyl-2-picrylhydrazyl (DPPH) Free Radical Scavenging Assay

The antioxidant activity of the *S. incanum* fruit extracts (methanol extract and aqueous extract) was determined by 1,1-diphenyl-2-picrylhydrazyl free radical scavenging activity (DPPH) assay as described by Al-Naqeb (2015) and Ibrahim et al. (2023b). In this analysis, ascorbic acid was used as a standard antioxidant agent; DPPH solution (50 mg DPPH in 100 ml methanol) served as the control, and each *S. icanum* fruit extract (methanol extract and aqueous extract) was independently dissolved in methanol. To determinate the radical scavenging activity of *S. incanum* fruit extracts, 0.5 ml of a methanol solution of DPPH was added to different concentrations (0.5, 1, 1.25, 1.5, 2 & 2.5 µg/ml) of ascorbic acid methanol solution and to various concentrations (2.5, 5, 7.5 &10 µg/ml) of *S. incanum* fruit extracts (methanol extract and aqueous extract) methanol solutions individually. Control and treated samples (ascorbic acid and both extracts) with DPPH solution were incubated in the dark at room temperature for 30 minutes, and then the absorbance was recorded at 517 nm using a Genova Life Science Analyser - Protein. The radical scavenging effect was calculated in percentage by the formula: Radical scavenging effect (%) = Ac – As/Ac × 100, where Ac = absorbance of the control and As = absorbance of the test sample (ascorbic acid and both extracts).

Determination of Antimicrobial Activities

Tested Pathogens

The antimicrobial activity of *S. incanum* fruit extracts (methanolic and aqueous) was evaluated against four clinically relevant human pathogens: one Gram-positive bacterium

(*Staphylococcus aureus*), two Gram-negative bacteria (*Escherichia coli* and *Pseudomonas aeruginosa*), and one fungal pathogen (*Candida albicans*). All microbial strains were obtained from the National Central Public Health Laboratory (NCPHL) in Sana'a, Yemen.

Antibacterial activity

The Agar well-diffusion method on Mueller Hinton Agar (MHA) media was applied to determine the activity of *S. incanum* fruit extracts (methanol and aqueous) against the selected bacterial pathogens (Iqbal *et al.,* 2015; Hussain *et al.*, 2016; Shahzad *et al.,* 2017). Five petri dishes containing MHA media were prepared for each bacterial pathogen, with two plates assigned to each extract type and one plate used as a positive control. Approximately 106 CFU/ml of each tested bacterium were cultured in each petri dish (Ibrahim *et al.*, 2023b). Using a sterilized cork borer, 4 to 3 wells (5 mm in diameter) were made in the agar of each petri dish. One well at the center was filled with 50 µl of dimethyl sulfoxide (DMSO) as negative control. In contrast, the remaining wells (located at the corners) were filled with the different concentrations of the plant extracts (Ibrahim *et al.*, 2022). To assess the antibacterial activity of *S. incanum* fruit extracts (methanol and aqueous), a stock solution of each extract at 100% concentration was prepared by dissolving 100 g of each extract in 100 ml of DMSO individually. Serial dilutions of each stock solution were made using DMSO to obtain concentrations of 50%, 25%, 12.5%, and 6.25%. Then, 50 µl of each concentration (100%, 50%, 25%, 12.5%, and 6.25%) of both methanol and aqueous extracts was individually pipetted into the designated wells (Ibrahim *et al.*, 2023a). For the positive control, a separate Petri dish was prepared containing the standard antibacterial agents Ampicillin and Ofloxacin. The petri dishes were incubated at 37°C for 24 hours, and then the diameters of the inhibition zones were measured in millimeters (Hiregoudar *et al.*, 2011). This procedure was performed in triplicate for each bacterial species, and the results were expressed as the mean inhibition zone diameter (Ibrahim *et al.*, 2023b). In addition, the Minimum Inhibitory Concentration (MIC) of each plant extract was determined against the three selected human pathogenic bacteria: *S. aureus*, *E. coli*, and *P. aeruginosa* as described by Hiregoudar *et al.* (2011) and Ibrahim *et al.* (2022).

Antifungal activity

The antifungal activity of *S. incanum* fruit extracts (methanol and aqueous) against C*. albicans* was determined using the agar well-diffusion method on Sabouraud Dextrose Agar (SDA) as demonstrated by Kalim et al. (2016) and Ibrahim et al. (2023a). Five Petri dishes containing SDA were prepared (two testing plates for each extract and one positive control plate). A sterilized swab was used to spread 100 microliters of *C. albicans* suspension (105 CFU/ml) over the SDA in each plate. Four to three wells (5 mm in diameter) were made in the agar media using a sterilized cork borer, one in the center, containing DMSO as a negative control. In contrast, the remaining wells (located at the corners) were filled with the different concentrations of the plant extracts (Ibrahim *et al.*, 2022). To determine the antifungal activity of *S. incanum* fruit (methanol and aqueous) extracts, a stock solution of *S. incanum* fruit extracts (methanol and aqueous) with a concentration of 100% for each extract was prepared individually by dissolving 100g of each *S. incanum* fruit extract in 100ml of DMSO. A serial dilution (50%, 25%, 12.5%, and 6.25%) was prepared from the stock solution of each extract using DMSO. 50 µl of each extract concentration (100%, 50%, 25%, 12.5%, and 6.25%) was pipetted into a well, while 50 µl of DMSO served as a negative control. For the positive control, a separate Petri dish was prepared containing the standard antifungal agents; Miconazole, Nystatin, and Ketoconazole. Plates were incubated at 37°C for 24 hours before measuring the inhibitory zone for each concentration. This process was repeated three times, and the inhibition zone diameter (mm) results were obtained as a mean (Ibrahim *et al.* 2023a). Furthermore, the Minimum Inhibitory

Concentration (MIC) of each extract against *C. albicans* was determined by assay as described by Mahesh et al. (2011) and Ibrahim et al. (2023b).

RESULTS

Phytochemical Screening

According to the findings of the Phytochemical investigation presented in Table 1, both methanol and aqueous extracts of *S. incanum* fruit contain several active pharmacological components, including alkaloids, saponins, phenols, tannins, flavonoids, and carbonyl-containing compounds. However, steroids were detected in the methanol extract and were absent in the aqueous extract (Table 1).

Table 1. Qualitative Phytochemical Constituents of *S. incanum* Methanol and Aqueous Fruit Extracts.

Sr. No.	Phytochemical components	Type of *S. incanum* Fruit Extract	
		Methanol	Aqueous
1	Alkhaloids	+ve	+ve
2	Saponins	+ve	+ve
3	Phenols+ Tannins	+ve	+ve
4	Flavonoids	+ve	+ve
5	Steroids	+ve	-ve
6	Carbonyl group	+ve	+ve

+ve: Present; -ve: Absent

Determination of Function Groups by Fourier transform infrared (FT-IR) assay

The FT-IR analyses identify the chemical and functional groups present in the methanol fruit extract of *S. incanum*, as detailed in Table 2 and illustrated in Figure 2. A broad peak observed at 3431.71 cm^{-1} indicates the presence of Hydroxyl group (O-H), suggesting that pharmaceutical compounds, such as phenols and their derivatives, are present in the extract (Figure 2). In contrast, the peaks at 2922.59 and 2854.13 cm^{-1} are attributed to the methylene (C-H) functional group, which is typically found in Aromatic and Alkenes compounds, indicating their presence in the methanol fruit extract (Figure 2). Moreover, the overtone/combination bands and the weak absorption between 1870.61 and 1846.51 cm^{-1} support the existence of Aromatic rings in the methanol fruit extract (Figure 2). Furthermore, the peak at 1734.66 cm^{-1} indicates the presence of the carbonyl (C=O) group, suggesting that some carbonyl compounds are present in the methanol fruit extract, whereas the absorptions at 1626.66 and 1421.28 cm^{-1} confirm the presence of C=C in the methanol fruit extract. In addition, the peaks at wavelengths 1064.51 and 669.178 cm^{-1} indicate the presence of -C-O &-C-H (out-of-plane, oop) respectively (Figure 2). On the other hand, Table (2) and Figure (3) exhibit the presence of the Hydroxyl group (OH) at the peak 3421.1 cm^{-1}, suggesting the presence of medicinal compounds such as phenols in the aqueous fruit extract of *S. incanum*. However, the medium absorption at 2924.52 cm^{-1} corresponds to the methylene (-CH) functional group of Aliphatic compounds in the aqueous fruit extract. Additionally, the peaks at 1734.6 and 1627.63 cm^{-1} indicate the presence of -C=O and N-H groups in the aqueous fruit extract respectively. This supports the presence of amino acids and

peptides in *S. incanum* aqueous fruit extract. Moreover, the peaks at 1053.91and 669.178 cm^{-1} support the presence of C-O and -C-H (out-of-plane bending) groups, respectively.

Table 2. FT-IR Spectral Analysis of *Solanum incanum* Methanol and Aqueous Fruit Extracts.

NO.	Function Group	Wavelength of Absorption cm^{-1}	
		Methanol Fruit Extract	Aqueous Fruit Extract
1	-OH	3431.71	3432.67
2	-C-H (Aromatic/Alkenes)	2922.59 & 2854.13	2924.52
3	Overtone/ Combination-Aromatic ring	1870.61 &1846.51	Absent
4	-C=O	1734.66	1734.66
5	-C=C	1626.66 & 1421.28	Absent
6	N-H	Absent	1627.63
7	-C-O	1064.51	1053.91
8	-C-H (out of plane-oop)	669.178	669.178

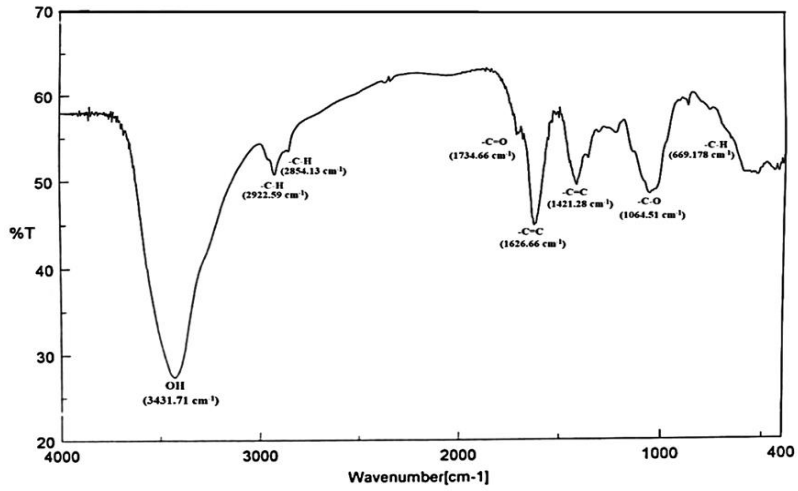

Fig. 2. FT-IR spectrum analysis of *S. incanum* methanol fruit extract.

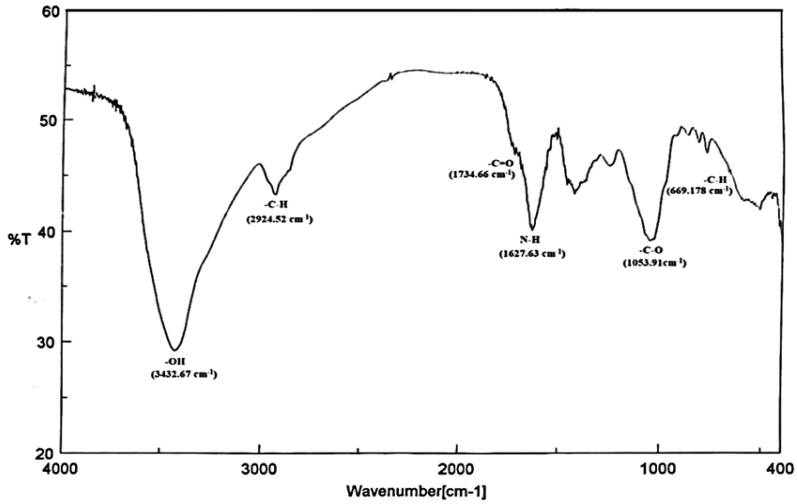

Fig. 3. FT-IR spectrum analysis of *S. incanum* aqueous fruit extract.

Assessment of Antioxidant Activity in *S. incanum* Fruit Extracts via 1, 1-Diphenyl-2-picrylhydrazyl (DPPH) Free Radical Scavenging Assay

This process is based on the reduction of the purple-colored stable free radical DPPH to the yellow-colored diphenyl picrylhydrazyl DPPH-H, which is a non-radical form, when an antioxidant agent is present. Figures 4 and 5 illustrate the free radical scavenging activity of the methanolic and aqueous extracts of *S. incanum* fruits respectively, as measured by the DPPH assay. The methanolic extract demonstrated significantly higher antioxidant activity, with scavenging percentages of 63.1%, 70.5%, 77%, and 80.8% at concentrations of 2.5 µg/ml, 5 µg/ml, 7.5 µg/ml, and 10 µg/ml, correspondingly. However, the aqueous extract exhibited markedly lower activity at the same concentrations, with scavenging values of 7.7%, 17.6%, 32.2%, and 44.6% accordingly. On the other hand, the standard antioxidant reference, ascorbic acid, showed 15% radical scavenging at a concentration of 0.5 µg/ml, increasing to 31.4%, 45.7%, 55.6%, 64%, and 83% at concentrations of 1 µg/ml, 1.25 µg/ml, 1.5 µg/ml, 2 µg/ml, and 2.5 µg/ml, respectively.

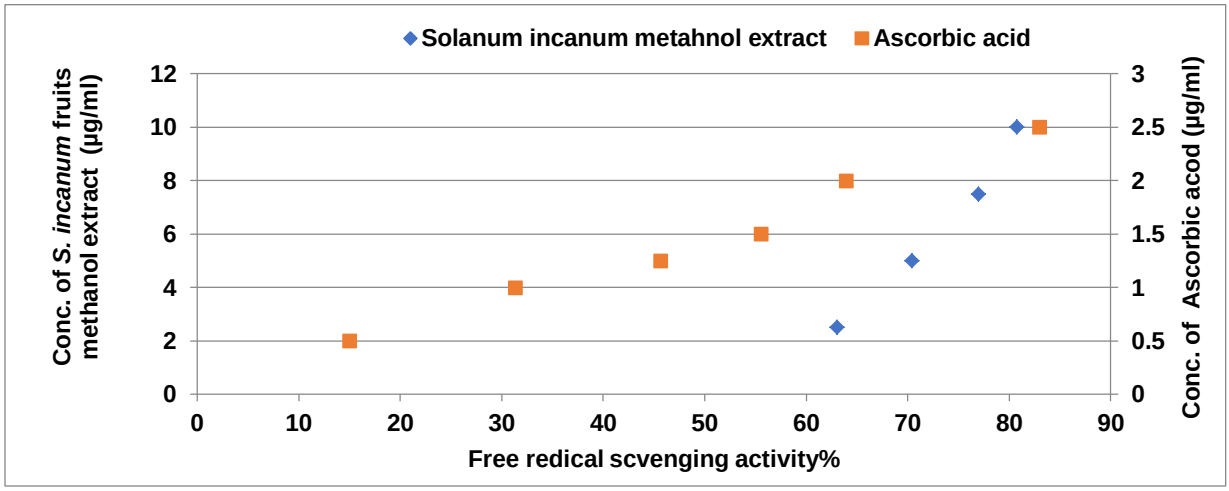

Fig. 4. Antioxidant Activity of *S. incanum* Fruit Methanolic Extract and Ascorbic Acid.

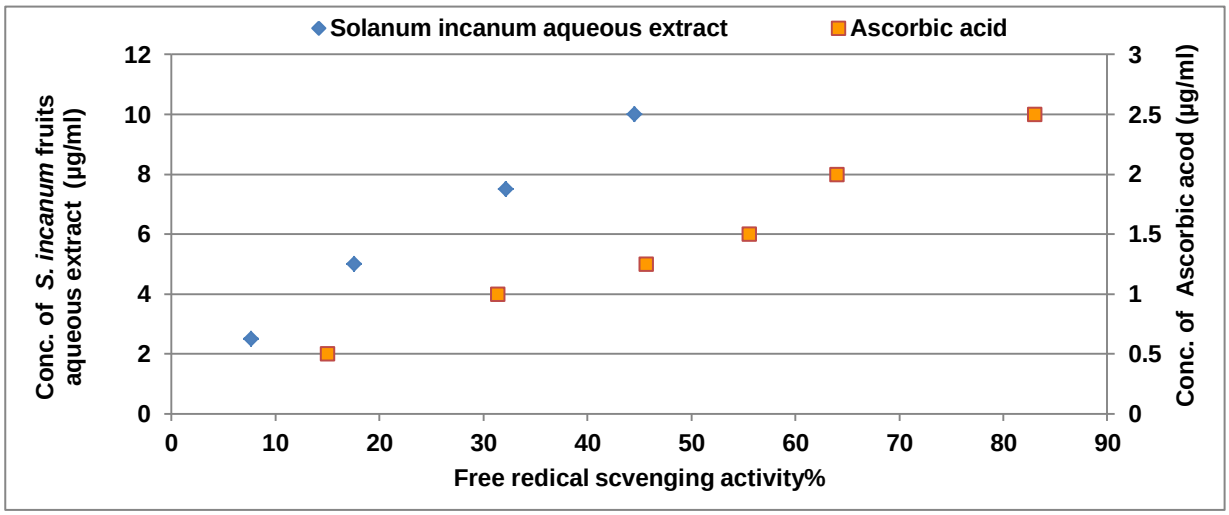

Fig. 5. Antioxidant Activity of *S. incanum* Fruit Aqueous Extract and Ascorbic Acid.

Antimicrobial Activities

According to Table 3, the methanol extract of *S. incanum* fruit showed antibacterial activity against two Gram-negative bacteria (*E. coli* and *P. aeruginosa*) and antifungal activity against one pathogenic fungus (*C. albicans*). Against *E. coli*, the extract showed a mean inhibition zones of 1, 6, 7, and 16 mm at concentrations of 12.5%, 25%, 50%, and 100%, respectively, while against *P. aeruginosa*, the extract exhibits a mean inhibition zone of 4 and 5 mm at 50% and 100% concentrations, accordingly. Moreover, the extract illustrated antifungal activity against *C. albicans*, with mean inhibition zones of 4.5, 6, 6.5, and 6 mm at the same concentrations (12.5%, 25%, 50%, and 100%, respectively).

Table 3. Inhibition zone diameters (mm) of *S. incanum* methanol fruit extract against tested Pathogens.

Tested Microorganism (Bacterial Pathogens)	**Type**	**Extraction Concentration %**					**-ve Control (DMSO)**	**+ve Control (Antibiotics)**		**MIC%**
		100%	50%	25%	12.5%	6.5%		OFOX	AMP	
		Inhibition Zone Diameter (mm)								
S. aureus	G +ve	-	-	-	-	-	-	20.5	24	-
E. coli	G -ve	16	7	6	1	-	-	17	27.5	12.5
P. aeruginosa	G -ve	5	4	-	-	-	-	-	16	50
Tested Microorganism (Fungal Pathogen)		**Extraction Concentration%**					**-ve Control (DMSO)**	**+ve Control (Antibiotics)**		**MIC%**
		100%	50%	25%	12.5%	6.5%		MIC	NS	KT
		Inhibition Zone Diameter (mm)								
C. albicans		6	6.5	6	4.5	-	-	3	25	-

G +ve = Gram positive; G-ve = Gram negative; AMP = Ampicllin; OFOX = Ofloxacin; MIC = Miconazol; NS= Nystation & KT= Ketoconazole

However, no antibacterial activity was detected against the selected Gram-positive bacterium (*S. aureus*). On the other hand, the aqueous *S. incanum* fruit extract, as indicated in Table 4, exhibits antibacterial activity against all three tested bacteria; one Gram-positive bacterium; *S. aureus* (with an average inhibition zone of 4 mm at 100% concentration), and two Gram-negative bacteria; *E. coli* (with average inhibition zones of 1, 6, 11, and 13 mm at concentrations of 12.5, 25, 50 and 100%, correspondingly) and *P. aeruginosa* (with average inhibition zones of 3, 7, 10, 12, and 8 mm at concentrations of 6.5, 12.5, 25, 50, and 100%, respectively). In addition, the extract demonstrates antifungal activity against the selected pathogenic fungus; *C. albicans*, with average inhibition zones of 1, 6, 7.5, 7.5, and 11 mm at concentrations of 6.5, 12.5, 25, 50, and 100%, sequentially. The minimum inhibitory concentration (MIC) of the methanol fruit extract (Table 3) was 12.5%, 50%, and 12.5% against *E. coli*, *P. aeruginosa*, and *C. albicans*, with mean inhibition zones of 1 mm, 4 mm, and 4.5 mm, respectively. In comparison, the MIC of the aqueous fruit extract (Table 4) was 100%, 12.5%, 6.5%, and 6.5% against *S. aureus*, *E. coli*, *P. aeruginosa*, and *C. albicans*, with mean inhibition zones of 4 mm, 1 mm, 3 mm, and 1 mm, correspondingly. These results indicate that the aqueous extract was effective against *S. aureus* and showed more efficacies at lower concentrations against *P. aeruginosa* and *C. albicans*.

Table 4. Inhibition zone diameters (mm) of *S. incanum* aqueous fruit extract against tested Pathogens.

Tested Microorganism (Bacterial Pathogens)	**Type**	**Extraction Concentration %**					**-ve Control (DMSO)**	**+ve Control (Antibiotics)**		**MIC%**
		100%	50%	25%	12.5%	6.5%		OFOX	AMP	
		Inhibition Zone Diameter (mm)								
S. aureus	G +ve	4	-	-	-	-	-	24	20.5	100
E. coli	G -ve	13	11	6	1	-	-	17	27.5	12.5
P. aeruginosa	G -ve	8	12	10	7	3	-	-	16	6.5
Tested Microorganism (Fungal Pathogen)		**Extraction Concentration%**					**-ve Control (DMSO)**	**+ve Control (Antibiotics)**		**MIC%**
		100%	50%	25%	12.5%	6.5%		MIC	NS	KT
		Inhibition Zone Diameter (mm)								
C. albicans		11	7.5	7.5	6	1	-	3	25	-	6.5

G +ve = Gram positive; G-ve = Gram negative; AMP = Ampicllin; OFOX = Ofloxacin; MIC = Miconazol; NS= Nystation & KT= Ketoconazole

DISCUSSION

The results presented in Table 1 demonstrate that the phytochemical analysis of the *S. incanum* methanol fruit extract reveals the presence of several secondary metabolites with potential pharmacological applications. These compounds include alkaloids, phenols, flavonoids, tannins, saponins, and steroids. This finding aligns with previous investigations, which reported that alkaloids, flavonoids, and saponins (Kipngeno *et al.*, 2014; Jepkoech and Gakunga, 2017); steroids (Jepkoech and Gakunga, 2017); and phenols and tannins (Kipngeno *et al.*, 2014) are present in the methanol fruit extract of *S. incanum*, supporting the traditional medicinal uses of the fruit. On the other hand, Sahle and Okbatinsae (2017) reported that the aqueous fruit extract of *S. incanum* contains alkaloids, phenols, flavonoids, tannins, saponins, and steroids. These findings are closely aligned with the results in Table 1, with one exception—steroids were not detected in the aqueous extract, as indicated in Table 1. However, Umar et al. (2015) reported the absence of steroids in the aqueous extract of *S. incanum* fruit, which is consistent with our results mentioned in Table 1.

The FT-IR spectral analysis indicates that the methanol fruit extract of *S. incanum* contains distinct functional groups characteristic of various bioactive compounds that may contribute to the extract's pharmacological activity, such as hydroxyl (O–H), methylene (C–H), carbonyl (C=O), alkene, aromatic C=C, and carbon–oxygen single bonds (C–O). These findings are consistent with those of Karanja et al. (2021), where they reported that the FT-IR spectrum of *S. incanum* fruit ethanol extract exhibited similar functional groups, with corresponding absorption peaks at 3348, 2931, 1218, 1396, and 1110 cm^{-1}, respectively. In contrast, the FT-IR analysis of the aqueous fruit extract, as shown in Table 2 and Figure 3, revealed the presence of hydroxyl (O–H), methylene (C–H), carbonyl (C=O), amine (N–H), and carbon–oxygen single bonds (C–O). These results align with those of Rahman et al. (2024), where they observed that the aqueous fruit extract of *S. xanthocarpum* exhibited comparable functional groups, with absorption peaks at 3315, 2900, 1614, and 1200 cm^{-1}, accordingly.

The previous results (Figures 4& 5) indicate that the methanolic extract of *S. incanum* fruits exhibits greater antioxidant activity compared to the aqueous extract. This observation aligns with the results reported by Ramamurthy et al. (2012), who cited that the methanolic fruit extract of *S. torvum* showed a higher antioxidant activity (20.48%, 35.43%, 54.97%, 85.99%, and 87.48% at concentrations of 10, 50, 100, 250, and 500 µg/mL, respectively) compared to the aqueous fruit extract, which demonstrated lower activity at the same concentrations, 25.20%,

14.11%, 51.44%, 74.69%, and 78.24%). The high antioxidant activity of the methanolic extracts can be attributed to the efficient extraction properties of organic solvents such as methanol, which are more effective than water in isolating a broader range of antioxidant compounds, particularly phenolics and flavonoids, due to their greater solubility in organic solvents (Kabra et *al.*, 2019; Irnameria and Okfrianti, 2023; Güçlü, 2024).

According to the results presented in Table 3, the methanolic fruit extract of *S. incanum* exhibited antimicrobial activity against *E. coli*, *P. aeruginosa*, and *C. albicans*. These findings are consistent with previous studies by Indhumathi *et al.* (2014), Sahle and Okbatinsae (2017), and Mawia *et al.* (2020). However, the absence of activity against *S. aureus* observed in this study may be attributed to resistance mechanisms present in the tested strain of *S. aureus*, as *S. aureus* is known to resist plant-derived antimicrobials through various mechanisms, including efflux pumps, enzymatic degradation of bioactive compounds, and reduced cell envelope permeability (Upadhyay *et al.*, 2014).

Moreover, the results in Table 4 indicate that the aqueous extract of *S. incanum* fruit demonstrated antimicrobial activity against all tested pathogenic microorganisms. This aligns with findings by Sahle and Okbatinsae (2017) and Chilala *et al.* (2023), who reported the antimicrobial effectiveness of aqueous *S. incanum* fruit extracts against *S. aureus*, *E. coli*, and *C. albicans*. However, no prior study has specifically observed the effect of aqueous *S. incanum* fruit extract against *P. aeruginosa*. In addition, the highest concentration (100%) of *S. incanum* fruit methanol extract exhibited lower antifungal activity against *C. albicans* compared to the 50% concentration, as shown in Table 3. An alleged explanation for this inverse result lies in the physical properties of the concentrated extract, which slowed its diffusion through the agar matrix. As noted by Balouiri *et al.* (2016); the extract at high concentrations, may have become more viscous or precipitated, which would interrupt its radial diffusion in the growth media. This would lead to a high local concentration of bioactive compounds near the well, leading to a misleadingly small zone of inhibition. Furthermore, the aqueous fruit extract of *S. incanum* exhibited higher antimicrobial activity against the tested pathogenic microorganisms compared to the methanolic fruit extract. This enhanced activity may be attributed to the presence of various bioactive antimicrobial peptides (AMPs), which are key components of the natural defense systems in most living organisms against invading pathogens (Ryan and Pearce, 2003; Meneguetti *et al.*, 2017; Afroz *et al.*, 2020). These peptides contain both N–H and C=O functional groups in their structure, which aligns with the findings shown in Table 2, where these groups were identified in the aqueous extract of *S. incanum* fruit. Numerous AMPs have been isolated from a wide range of plant species, including members of the genus *Solanum*, where they have been found in different plant parts such as seeds, leaves, fruits, and tubers (Afroz *et al.*, 2020). These peptides have demonstrated significant antibacterial, antifungal, and antiviral activities against both phytopathogenic and human pathogenic strains (Afroz *et al.*, 2020). Their antibacterial mechanism is believed to involve disruption of membrane permeability (Muhammad *et al.*, 2019; Afroz *et al.*, 2020), while their antifungal effects are associated with inhibition of fungal growth and hyphal development (Maracahipes *et al.*, 2019; Afroz *et al.*, 2020).

Based on the previous results in Tables 4, the high concentrations of *S. incanum* fruit aqueous extract exhibited strong antimicrobial activity against *S. aureus*, *E. coli*, and *C. albicans*. However, the same high concentration showed a reduced effectiveness against *P. aeruginosa*. In contrast, the lower concentrations of the extract were more effective against *P. aeruginosa*, suggesting a non-linear relationship between extract concentration and antimicrobial activity. These findings are consistent with previous reports indicating that the relationship between extract concentration and antibacterial efficacy is not always linear. While higher concentrations

typically enhance antibacterial activity, there are cases where high concentrations may actually reduce effectiveness against certain bacterial types or strains (King *et al*., 2008; Eloff, 2019). This reduction in activity may be attributed to the increased presence of antagonistic compounds in the crude extract mixture at higher concentrations, which can interfere with the antibacterial potential against specific bacteria (Ejaz *et al*., 2023).

CONCLUSION

Phytochemical screening confirmed the presence of various bioactive compounds in both methanolic and aqueous extracts of *S. incanum* fruit, with steroids present only in the methanol extract, FT-IR analysis further supported the presence of these compounds by revealing their characteristic functional groups. The methanol extract exhibited higher antioxidant activity, while the aqueous extract showed high antimicrobial effects against all tested pathogens. This antimicrobial activity of the aqueous extract is probably attributed to the presence of bioactive antimicrobial peptides (AMPs). These findings suggest that *S. incanum* fruit extracts could serve as natural sources of antioxidant and antimicrobial agents.

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CONFLICT OF INTEREST

There is no conflict of interest.

REFERENCES

Afroz, M., Akter, S., Ahmed, A., Rouf, R., Shilpi, J.A., Tiralongo, E., Sarker, S.D., Göransson, U., Uddin, S.J., 2020. Ethnobotany and Antimicrobial Peptides From Plants of the Solanaceae Family: An Update and Future Prospects. Front. Pharmacol., 11: 565.

Al Dubaie, A.S., Al Khulaidi, A.A., 2005. Medical and Aromatic Plants of Yemen, ed. 2 (in Arabic). Obadi center for studies and publisher, Sana'a, Yemen.

Al Khulaidi, A.A., 2013. Flora of Yemen. The Sustainable Natural Resource Management Project (SNRMP II), EPA and UNDP, Republic of Yemen.

Al-Hubaishi, A., Müller-Hohenstein, K., 1984. An Introduction to the Vegetation of Yemen. GTZ. Deutsche Gesellschaft Für Technische Zusammenarbeit (GTZ) GmbH, Eschborn, Germany.

Almoulah, N.F., Voynikov, Y., Gevrenova, R., Schohn, H., Tzanova, T., Yagi, S.M., Thomas, J., Mignard, B., Ahmed, A.A., Siddig, M.A., Spina, R., Laurain-Mattar, D., 2017. Antibacterial, antiproliferative and antioxidant activity of leaf extracts of selected *Solanaceae* species. South Afr. J. Bot., 112: 368-374.

Al-Naqeb, G., 2015. Antioxidant and Antibacterial Activities of Some Yemeni Medicinal Plants. Int. J. Herbal Med., 3: 06-11.

Balouiri, M., Sadiki, M., Ibnsouda, S.K., 2016. Methods for in vitro evaluating antimicrobial activity: A review. J. Pharm. Analysis., 6(2): 71-79.

Chen, Y., Xie, M.-Y., Yan, Y., Zhu, S.-B., Nie, S.-P., Li, C., Wang, Y.-X, Gong, X.-F., 2008. Discrimination of *Ganoderma lucidum* according to geographical origin with near-infrared diffuse reflectance spectroscopy and pattern recognition techniques. Anal. Chim. Acta., 618(2): 121–130.

Chilala, P., Ndubi, F., Mayoka, G., Kampamba, M., Kabuka, R., Chabalenge, B., Mudenda, S., 2023. Antimicrobial

activities of Solanum aculeastrum fruit extract against *Escherichia coli*, *Staphylococcus aureus* and *Candida albicans*: significance of african traditional medicine in combating infections and attaining universal health coverage. Pharmacol. Pharm., 14(5): 176-188.

Chirchir, D.K., Mshimba, S.M., Cheruiyot, G., Adongo, J.O., 2014. Antimicrobial activity and phytochemical investigation of crude extracts of the fruits of *Solanum incanum* (*Solananceae*) and *Dovyalis abbysinica* (*Flacourtiaceae*). doi:10.7237/sjmb/193

Chowanski, S., Adamski, Z., Marciniak, P., Rosiński, G., Büyükgüzel, E., Büyükgüzel, K., Falabella, P., Scrano, L., Ventrella, E., Lelario, F., Bufo, S.A., 2016. A Review of Bioinsecticidal Activity of Solanaceae Alkaloids. Toxins, (Basel) 8, 1–28. doi: 10.3390/toxins8030060.

Ebad, F.A, Ismaeil, S.M., Gabr, M.M., Alhadi, F.A., 2006. Influence of Altitudinal Gradient on Some Physiological and Ecological Aspects of *Solanum incanum* L. Growing Naturally in Yemen. Egyptian J. Desert Res., 56(2): 243-258.

Ejaz, S., Abdullah, I., Rashid, S., Ashraf, M., 2023. Analysis of antibacterial and cytotoxic potential of medicinal plants from Cholistan desert, Pakistan. Saudi J. Biol. Sci., 30(9): 103750.

Eloff, J.N., 2019. Avoiding pitfalls in determining antimicrobial activity of plant extracts and publishing the results. BMC Complement. Alternative Med., 19(1): 106.

Ghatak, A., Chaturvedi, P., Paul, P., Agrawal, G.K., Rakwal, R., Kim, S.T., Weckwerth, W., Gupta, R., 2017. Proteomics survey of Solanaceae family: Current status and challenges ahead. J. Proteomics, 169: 41-57.

Güçlü, G., 2024. In Vitro Antioxidant Assessment of Methanol and Water Extracts of *Anthemis kotschyana* Boiss. var. kotschyana. Cumhuriyet Sci. J., 45(3): 449-453.

Hiregoudar, M., Suubramaniam, R., Mittal, S., Muralikrishna, K. S., Sakeenabi, B., Prashant, G. M., Chandru, G. N., 2011. Antibacterial activity of some spice extracts on *Streptococcus* mutans: An: invitro: study. J. Ind. Assoc. Public Health Dent., 9 (Suppl 1): S347-S351.

Hussain, F., Kalim, M., Ali, H., Ali, T., Khan, M., Xiao, S., Iqbal, M.N., Ashraf, A., 2016. Antibacterial Activities of Methanolic Extracts of *Datura innoxia*. PSM Microbiol., 1(1): 33-35.

Ibrahim, H.M., Abdullah, Q.Y.M., Humaid, A.A., Tabit, A.A.M., Al-alawi, S.M., Al-ramsi, A.T., 2022. Phytochemical screening and anti-microbial activities of *Aloe fleurentinorum* lavranos & Newton. J. Chem. Biol. Phys. Sci., 12: 335-348.

Ibrahim, H., Humaid, A.A., Thabit, A.A.M., Rizq, E.A., Al-awadhi, B.H., 2023a. Phytochemical screening, antioxidant and antimicrobial activities of *Acacia origena* hunde. J. Chem. Biol. Phys. Sci, 13(3): 308-321.

Ibrahim, H.M., Al- Ghani, A.M.S., Humaid, A.A., Abdullah, Q.Y., Thabit, A.A., AL-Awadhi, B.H., 2023b. Phytochemical Screening, Antioxidant and Antimicrobial Activities of *Aloe rubroviolacea* Schweinf. PSM Biol. Res., 8(3): 92-108.

Indhumathi, T., Mohandass, S., Arts, N., 2014. Efficacy of Ethanolic extract of Solanum incanum fruit extract for its antimicrobial activity.International Journal of Current Microbiology and Applied Sciences, 3(6):939-949.

Iqbal, M.N., Anjum, A.A., Ali, M.A., Hussain, F., Ali, S., Muhammad, A., Irfan, M., Ahmad, A., Shabbir, A., 2015. Assessment of Microbial Load of Un-pasteurizedFruit Juices and in vitro Antibacterial Potential of Honey against Bacterial Isolates. The Open Microbiol. J., 9:26-32.

Irnameria, D., Okfrianti, Y., 2023. Differences In the Antioxidant Activity of Sungkai Leaf Extract Using Methanol And Aquades Solvents. AGRITEPA: Jurnal Ilmu dan Teknologi Pertanian, 10(1), 219-228.

Jepkoech, K.E., Gakunga, N.J., 2017. Antimicrobial activity and phytochemical screening of *Solanum incanum* fruit extract against clinical samples of

Staphylococcus aureus collecting from Nakuru Provincial General Hospital Laboratory, Kenya. Int. Res. J. Med. Biomed. Sci., 2(1): 1-8.

Kabra, A., Sharma, R., Hano, C., Kabra, R., Martins, N., Baghel, U.S., 2019. Phytochemical composition, antioxidant, and antimicrobial attributes of different solvent extracts from myrica esculenta buch.-Ham. ex. D. Don Leaves. Biomol., 9(8): 357.

Kalim, M., Hussain, F., Ali, H., Iqbal, M.N., 2016. Antifungal activities of Methanolic Extracts of *Datura innoxia*. PSM Biol. Res., 01(2): 70-73.

Karanja, L.N., K'Owino, I.O., Wangila, P.T., Ramkat, R.C., 2021. Phytochemical Composition and Antibacterial Activity of Fruit Extract of *Solanum incanum* L. against *Ralstonia solanacearum*. Asian J. Appl. Chem. Res., 9(4): 1-16.

King, T., Dykes, G., Kristianti, R., 2008. Comparative evaluation of methods commonly used to determine antimicrobial susceptibility to plant extracts and phenolic compounds. J. AOAC Int., 91(6): 1423-1429.

Kipngeno, C.D., Mshimba, S.M., Gilbert, C., John, O., 2014. Antimicrobial activity and phytochemical investigation of crude extracts of the fruits of *Solanum incanum* (Solananceae) and *Dovyalis abbysinica* (Flacourtiaceae). Sci. J. Microbiol., doi:10.7237/sjmb/193.

Mahesh, H., Suubramaniam, R., Simpy, M., Muralikrishna, K.S., Akeenabi, S.B., Prashant, G.M., Chandru, G.N., 2011. Antibacterial activity of some spice extracts on *Streptococcus mutans*: An invitro study. JIAPHD, 2011(17): 347- 351.

Maracahipes, Á.C., Taveira, G.B., Mello, E.O., Carvalho, A.O., Rodrigues, R., Perales, J., Teixeira-Ferreira, A., Silva, M.S., Rocha, G.L., Fernandes, K.V., Gomes, V.M., 2019. Biochemical analysis of antimicrobial peptides in two different Capsicum genotypes after fruit infection by Colletotrichum gloeosporioides. Biosci. Rep. 39(4): BSR20181889. doi: 10.1042/BSR20181889.

Mawia, K.J., Muthuka, J.K., Wambura, F.M., Muthui, J., 2020. Antibacterial Activity of *Solanum incanum* Roots and Fruits Methanol Extracts against Gastrointestinal Bacteria Causing Food Poisoning. IOSR J. Pharm. Biol. Sci., 15(2): 53-59.

Meneguetti, B.T., Machado, L.S., Oshiro, K.G.N., Nogueira, M.L., Carvalho, C.M. E., Franco, O.L., 2017. Antimicrobial peptides from fruits and their potential use as biotechnological tools—A Review and Outlook. Front. Microbiol., 7(2136): 1–13. doi: 10.3389/fmicb.2016.02136

Muhammad, S.M., Sabo, I.A., Gumel, A.M., Fatima, I., 2019. Extraction and purification of antimicrobial proteins from *Datura stramonium* Seed. J. Adv. Biotechnol., 18: 1073–1077. doi: 10.24297/jbt.v8i0.8221

Nath, P., Yadav, A.K., Soren, A.D., 2017. Sub-acute toxicity and genotoxicity assessement of the rhizome extract of *Acorus calamus* L., A medicinal plant of India. Eur. J. Pharm. Med. Res., 4: 392–399.

Rahman, G., Fazal, H., Ullah, A., Ahmad, S., Nadeem, T., Ahmad, M., Ahmad, I., Mishra, N., Ashique, S., Zengin, G., Farid, A., 2024. Empowering silver and copper nanoparticles through aqueous fruit extract of *Solanum xanthocarpum* for sustainable advancements. Biomass Convers. Bior., 15(4): 5155-5169.

Ramamurthy, C., Kumar, M.S., Suyavaran, V., Mareeswaran, R., Thirunavukkarasu, C., 2012. Evaluation of antioxidant, radical scavenging activity and polyphenolics profile in *Solanum torvum* L. fruits. J. Food Sci., 778: C907-13.

Ryan, C.A., Pearce, G., 2003. Systemins: a functionally defined family of peptide signals that regulate defensive genes in Solanaceae species. Proc. Natl. Acad. Sci. U. S. A., 100 (Suppl. 2): 14577–14580. doi: 10.1073/ pnas.193478810.

Sahle, T., Okbatinsae, G., 2017. Phytochemical investigation and antimicrobial activity of the fruit extract of Solanum incanum grown in Eritrea. Ornam. Med. Plants., 1(1): 15-25.

Séré, M., Konaté, K., Santara, B., Sanou, D.O., Belem, A.M., 2022. Study of the antibacterial capacity of extracts of *Solanum incanum* L., (Solanaceae) traditionally used for the management of avian cholera in rural areas of Burkina Faso. World J. Adv. Res. Rev., 13: 047-57.

Shah, V.V., Shah, N.D., Patrekar, P.V., 2013. Medicinal Plants from Solanaceae Family. Res. Pharm. Technol., 6: 143–151.

Shahzad, M.I., Ashraf, H., Iqbal, M.N., Khanum, A., 2017. Medicinal Evaluation of Common Plants against Mouth Microflora. PSM Microbiol., 2(2): 34-40.

Umar, A., Piero, N.M., Mgutu, A.J., Ann, N.W., Maina, G.S., Maina, M.B., Njagi, J.M., Mworia, J.K., Ngure, J.K., Mwonjoria, J.K., 2015. Bio efficacy of aqueous crude fruit sap extract of *Solanum incanum* against green peach aphids *Myzus persicae* Sulzer (Homoptera: Aphididae). Entomol. Ornithol. Herpetol., 5(169): 2161-0983.

Upadhyay, A., Upadhyaya, I., Kollanoor-Johny, A., Venkitanarayanan, K., 2014. Combating pathogenic microorganisms using plant-derived antimicrobials: a mini-review of the mechanistic basis. BioMed Res. Int., 2014(1): 761741.

Venkatesan, D., Karrunakaran, C.M., Kumar, S.S., 2009. Studies on phytochemical constituents, functional group identification and antimicrobial activity of *Solanum nigrum* (Solanaceae). Ethnobotanical Leaflets, 13: 1485-1503.

Wood, J.R.I., 1997. Handbook of the Yemen Flora. Royal Botanic Gardens, Kew, UK.