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Research Article

Screening of antimicrobial activity of the medicinal plant extracts against skin infection in diabetic patient

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Abstract

India has a rich heritage of knowledge on plant based drugs both for use in preventive and curative medicine. A large number of these plants grow wild and exploited especially for use in indigenous pharmaceutical houses. Some of these plants produce valuable drugs which have high export potential. Antibacterial activity of herbal plant extract of *Coccinia grandis*, *Tridax procumbens* and *Phyllanthus niruri* showed that the methanol extract showed promising antimicrobial activity against bacterial and fungal human pathogens followed by chloroform extract and ethyl acetate extract. Among the three plants, maximum inhibition activity was exhibited by *Phyllanthus niruri* followed by *Tridax procumbens* and *Coccinia grandis*. The results also indicated that scientific studies carried out on medicinal plants having traditional claims of effectiveness might warrant fruitful results. These plants could serve as useful source of new antimicrobial agents.

Keywords: Medicinal plants, Antimicrobial activity, Diabetics and Human pathogens.

Introduction

India has a rich heritage of knowledge on plant based drugs both for use in preventive and curative medicine. A country like India is very much suited for development of drugs from medicinal plant. Because of its vase and wide variations in soil and climate, the Indian sub – continent is suitable for cultivation of large number of medicinal and aromatic plant which can be used as raw materials for pharmaceutical, perfumery, cosmetics, flavor, food and agrochemical industries. A large number of these plants grow wild and exploited especially for use in indigenous pharmaceutical houses. Some of these plants produce valuable drugs which have high export potential. [(2)]

Bacterial infections are common to all people, but can be especially troublesome for people with type 2 diabetes. Bacteria-laden infections of the skin are often painful and warm to the touch, with swelling and redness. They may increase in size, number, and frequency if your blood glucose is repeatedly high.

The most common bacteria that cause skin infections are *Staphylococcus* and *Streptococcus* [(7).] Serious bacterial infections can cause carbuncles (deep-tissue infections) that may need to be lanced by a physician and drained. If you suspect that you have a bacterial infection, notify your doctor immediately so you may be treated with antibiotics. Other common bacterial infections include boils, sties (infections around the eyes), folliculitis (infections of the hair follicles), and infections around the fingernails and toenails. [(6)]

Fungal infections, caused by the spread of the fungus *Candida albicans*, or yeast, are also common for type 2 diabetes patients, especially if their blood glucose is not well controlled. Yeast infections look like areas of red, itchy, swollen skin that are surrounded by blistering or dry scales. Yeast fungus thrives in the warm folds of the skin, under breasts, in the groin, in the armpits, in the corners of the mouth, and under the foreskin. Common skin irritations like athlete's foot,

jock itch, and ringworm are fungal infections. They can itch, spread, and worsen if not treated with prescription medication. [5]

Coccinia grandis, the ivy gourd, also known as *baby watermelon, little gourd, gentleman'stoes, tindora* in Hindi, *tondli* in Marathi,*dondekayi* in Kannada, *don dakaya* in Telugu, Kovaykka in Malayalam and Kovaikkai in Tamil or sometimes inaccurately identified as*gherkin*, is a tropical vine. It is also known as *Cephalandra indica* and *Coccinia indica*.Ivy gourd in India.

Tridax procumbens is known for several potential therapeutic activities like antiviral, anti oxidant antibiotic efficacies, wound healing activity, insecticidal and anti-inflammatory activity. Some reports from tribal areas in India state that the leaf juice can be used to cure fresh wounds, to stop bleeding, as a hair tonic.

The *Phyllanthus* genus of the family Euphorbiaceae was first identified in Central and Southern India in 18th century. It is commonly called carry me seed, stone-breaker, windbreaker,gulf leaf flower or gala of. There are over 300 genera with over 5000 species in the Euphorbiaceae worldwide. [4]

Several compounds including alkaloids, flavonoids, lignans, phenols and terpenes were isolated from this plant and some of them interact with most key enzymes. In traditional medicine, it is used for its hepatoprotective, anti-diabetic, antihypertensive, analgesic, anti-inflammatory and antimicrobial properties. *Phyllanthus niruri* leaf extract as a hepatoprotective agent .[3]

The beneficial medicinal effects of plant materials typically result from the secondary products present in the plant although, it is usually not attributed to a single compound but a combination of the metabolites. The medicinal actions of plants are unique to a particular plant species or group, consistent with the concept that the combination of secondary products in a particular plant is taxonomically distinct. [1]

2.Materials and Methods

2.1 General methods

2.1.1. Sterilization

All the media were sterilized in autoclave at 15 lbs pressure for 20 minutes. The glasswares were sterilized at 180°C for 3 hours in hot air oven.

2.1.2. Chemicals

All the chemicals used in the experiments were of analytical reagents (AR) grade and distilled water was used throughout the study.

2.1.3. Collection of Plant Material

Healthy leaves of *Coccinia grandis, Tridax procumbens* and *Phyllanthus niruri* were collected from Gingee hills.

The plant materials like leaves were washed thoroughly with tap water and then with sterilized distilled water for the removal of dust and sand particles. The leaves were shade dried and powdered by hand crushing. The powdered samples were hermetically sealed in separate polythene bags until the time of the extraction. This was used as the raw material for the extraction of antimicrobial compounds against the microbes used.

2.2. Isolation and identification of microbes

2.2.1. Sample

The skin scraping samples was collected from diabetes patient in government medical college in Tiruvannamalai, Tamilnadu.

2.2.2. Collection of Sample

The skin scrap swab was introduced deeply enough to obtain a moist specimen the swab was replaced in the transport media. The sampled scrap swab was taken immediately the laboratory for processing of the sample. If delayed inoculated in the nutrient broth.

2.2.3. Processing of Samples

The sampled swab was streaked in nutrient agar medium. It was incubated at37°Cfor 24 hrs. The developed colonies were differentiated based on their colony morphology.

2.3. Preliminary Test

2.3.1. Gram's staining

Gram staining technique was first discovered by Danish physician named Hans Christiangram in 1884. This technique divides bacterial cells into two major groups, gram positive an gram negative bacteria, this

makes it an essential tool for clarification and differentiation of microorganisms

2.3.2. Motility

The hanging drop technique was followed to observe the motility of the bacteria. It is done to check whether the organism is motile or non motile. A clean cover slip was place with petroleum jelly at the four corners. A microbial suspension was placed over it and inverted with the slide on top producing a hanging drop.

2.3.3. Inoculation on to medium

The culture was streaked on the nutrient agar medium with help of an inoculation loop. The medium was incubated at 37 °C for 24 hrs.

2.3.4. Inoculation into Selective Medium

After overnight incubation, the nutrient agar medium was checked for growth. Colony morphology was noted down and the gram‟s staining and motility was performed again. Based on the colony morphology and the grams staining and motility, culture was inoculated in selective media like Macconkey agar, Blood agar and Eosin methylene blue agar. It was incubated at 37 °C for 24 hrs.

2.4. Biochemical Characterstics

Indole Production Test, Methyl Red Test, Voges Proskauer Test, Citrate Utilization Test, Triple Sugar Iron Agar Test, Urease Test, Nitrate Test, Catalase Test, Oxidase Test

2.5. Identification of the Pathogen

Based on the results obtained in the biochemical tests and the colony morphology on the selective media, the pathogen was identified. Pure cultures of the particular pathogen were then maintained in either nutrient agar slants or nutrient agar plates for antimicrobial testing. The bacterial isolates were confirmed using Gram staining, motility test, platting on selective medium, catalase test, oxidase test and other biochemical test and also inoculating them on specific media.

2.6. Evaluation of medicinal plant leaf extracts for its antimicrobial activity

2.6.1. Preparation of leaf extract

The preparations of different leaves extract was done through modified method [8]

2.6.2. Methanol extraction method

The shade dried leaf materials were used for the methanol extraction procedure; about 5 gm of leaf powder were weighed and mixed with ethanol (1:3 w/v) which was incubated for two days. After the incubation period, the slurry was filtered through Whattman No.1 filter paper in a beaker and allowed it for evaporation. The residue was dissolved with Dimethyl sulfoxide (DMSO) with different concentrations and checked it for its Antimicrobial activity.

2.6.3. Ethyl acetate extraction method

The shade dried leaf materials were used for the ethyl acetate extraction procedure; about 5 gm of leaf powder were weighed and mixed with ethyl acetate (1:3 w/v) which was incubated for two days. After the incubation period, the slurry was filtered through Whattman No.1 filter paper in a beaker and allowed it for evaporation. The residue was dissolved with Dimethyl sulfoxide (DMSO) with different concentrations and checked it for its antimicrobial activity.

2.6.4. Chloroform extraction method

The shade dried leaf materials were used for the chloroform extraction procedure; about 5 gm of leaf powder were weighed and mixed with chloroform (1:3 w/v) which was incubated for two days. After the incubation period, the slurry was filtered through Whattman No.1 filter paper in a beaker and allowed it for evaporation. The residue was dissolved with Dimethyl sulfoxide (DMSO) with different concentrations and checked it for its antimicrobial activity.

2.7. Evaluation of Antimicrobial Activity of Medicinal Plants

Antimicrobial activity of medicinal plant was tested through several methods like Tube dilution method ,well plate method and Disc diffusion method. Disc diffusion method is most commonly employed method to evaluate the antimicrobial activity. In the present study, Disc diffusion method was used to test the antimicrobial activity of leaves of Coccinia grandis, *Tridax procumbens* and *Phyllanthus niruri*. The Disc diffusion technique was introduced by Kirby – Bauer.

2.8. Antimicrobial susceptibility test

Disc diffusion method was adopted for the evaluation of antimicrobial activity of five different medicinal leaves. Muller Hinton agar was prepared and autoclaved at 15 lbs pressure for 20 minutes and cooled at 45°C. The cooled media was poured on to sterile petriplates and allowed for solidification. The plates with media were seeded with the respective microbial suspension using sterile swab. The disc impregnated with respective leaf extracts at different concentration (100 - 300 mg/ml) individually were placed on the four corners of each petridishes, control disc was also placed. The petridishes were then incubated at 37°C for 24 hours. After incubation period, the diameter of the zone formed around the paper disc were measured and expressed in mm.

3.Results and Discussion

The results of the experiment describes the antimicrobial effect of crude extract of *Coccinia grandis*, *Tridax procumbens* and *Phyllanthus niruri* with three different solvents *viz*., Methanol, Chloroform and Ethyl acetate against bacteria and fungi. The antimicrobial activity was studied by Disc diffusion method and the results were described below.

3.1. Antimicrobial Activity of Medicinal Plants

The antimicrobial activity of methanol, chloroform and ethyl acetate leaf extract of *Coccinia grandis*, *Tridax procumbens* and *Phyllanthus niruri* were studied in different concentrations (100 mg/ml, 200 mg/ml, 300 mg/ml). Antibacterial potential of leaf extract was assessed in terms of zone of inhibition of bacterial and fungal growth.

3.2. Antimicrobial activity of *Coccinia grandis*

3.2.1. Methanol extract

The antimicrobial activity of methanol extract of *Coccinia grandis* was analyzed in the present study and the results were furnished in Table – 1. Among the three concentrations (100 mg/ml, 200, 300 mg/ml) used, maximum inhibitory zone was observed at 300 mg/ml followed by 200 mg/ml and 100 mg/ml. Maximum antibacterial activity was observed in the bacteria *Bacillus subtilis* (24 mm, 28 mm and 34 mm) followed by *Pseudomonas aeruginosa* (25 mm, 30 mm and 31 mm), *Staphylococcus aureus* (21 mm, 23 mm and 25 mm) and *Streptococcus pyogens* (8 mm, 13 mm and 15 mm). The fungi *Penicillium* sp. (13 mm, 15 mm and 16 mm) showed more inhibitory activity when compared to *Aspergillus niger* (No zone, 10 mm and 12 mm). No zone of inhibition was observed in the negative DMSO control.

3.2.2. Chloroform extract

The antimicrobial activity of chloroform extract of *Coccinia grandis* was determined in the present investigation and the results were given in Table – 2. Among the three concentrations (100 mg/ml, 200, 300 mg/ml) used, maximum inhibitory zone was noticed at 300 mg/ml followed by 200 mg/ml and 100 mg/ml. Maximum antibacterial activity was observed in the bacteria *Bacillus subtilis* (19 mm, 23 mm and 29 mm) followed by *Pseudomonas aeruginosa* (20 mm, 25 mm and 26 mm), *Staphylococcus aureus* (16 mm, 18 mm and 20 mm) and *Streptococcus pyogens* (No zone, 8 mm and 10 mm). The fungi *Penicillium* sp. (8 mm, 10 mm and 11 mm) showed more inhibitory activity than *Aspergillus niger*. No zone of inhibition was observed against *Aspergillus niger* and negative DMSO control.

3.2.3. Ethyl acetate extract

The antimicrobial activity of ethyl acetate extract of *Coccinia grandis* was evaluated in the present research and the results were presented in Table – 3. Among the three concentrations (100 mg/ml, 200, 300 mg/ml) used, maximum inhibitory zone was recorded at 300 mg/ml followed by 200 mg/ml and 100 mg/ml. Maximum antibacterial activity was observed in the bacteria *Bacillus subtilis* (16 mm, 20 mm and 26 mm) followed by *Pseudomonas aeruginosa* (17 mm,

22 mm and 23 mm), *Staphylococcus aureus* (13 mm, 15 mm and 17 mm) and *Streptococcus pyogens* (No zone, No zone and 8 mm). The ethyl acetate extract of *Ocimum sanctum* showed resistance against *Penicillium* sp. and *Aspergillus niger*. No zone of inhibition was observed against *Penicillium* sp., *Aspergillus niger* and negative DMSO control.

Table - 1: Antimicrobial activity of methanol extract of *Coccinia grandis*

S.No	Organisms	Concentration of the extract (mg/ml) and zone of inhibition (mm in dm)			
		DMSO Control	100 mg/ml	200 mg/ml	300 mg/ml
1	*Staphylococcus aureus*	NZ	21 mm	23 mm	25 mm
2	*Streptococcus pyogens*	NZ	8 mm	13 mm	15 mm
3	*Pseudomonas aeruginosa*	NZ	25 mm	30 mm	31 mm
4	*Bacillus subtilis*	NZ	24 mm	28 mm	34 mm
5	*Aspergillus niger*	NZ	NZ	10 mm	12 mm
6	*Penicillium* sp.	NZ	13 mm	15 mm	16 mm

NZ – No Zone

Table - 2: Antimicrobial activity of chloroform extract of *Coccinia grandis*

S.No	Organisms	Concentration of the extract (mg/ml) and zone of inhibition (mm in dm)			
		DMSO Control	100 mg/ml	200 mg/ml	300 mg/ml
1	*Staphylococcus aureus*	NZ	16 mm	18 mm	20 mm
2	*Streptococcus pyogens*	NZ	NZ	8 mm	10 mm
3	*Pseudomonas aeruginosa*	NZ	20 mm	25 mm	26 mm
4	*Bacillus subtilis*	NZ	19 mm	23 mm	29 mm
5	*Aspergillus niger*	NZ	NZ	NZ	NZ
6	*Penicillium* sp.	NZ	8 mm	10 mm	11 mm

NZ – No Zone

Table - 3: Antimicrobial activity of ethyl acetate extract of *Coccinia grandis*

S.No	Organisms	Concentration of the extract (mg/ml) and zone of inhibition (mm)			
		DMSO Control	100 mg/ml	200 mg/ml	300 mg/ml
1	*Staphylococcus aureus*	NZ	13 mm	15 mm	17 mm
2	*Streptococcus pyogens*	NZ	NZ	NZ	8 mm
3	*Pseudomonas aeruginosa*	NZ	17 mm	22 mm	23 mm
4	*Bacillus subtilis*	NZ	16 mm	20 mm	26 mm
5	*Aspergillus niger*	NZ	NZ	NZ	NZ
6	*Penicillium* sp.	NZ	NZ	NZ	NZ

NZ – No Zone

3.3. Antimicrobial activity of *Tridax procumbens*

3.3.1. Methanol extract

The antimicrobial activity of methanol extract of *Tridax procumbens* was studied and the results were showed in Table – 4. Among the three concentrations (100 mg/ml, 200, 300 mg/ml) used, maximum inhibitory zone was observed at 300 mg/ml followed by 200 mg/ml and 100 mg/ml. Maximum antibacterial activity was observed in the bacteria *Bacillus subtilis* (28 mm, 32 mm and 35 mm) followed by *Staphylococcus aureus* (25 mm, 26 mm and 28 mm), *Pseudomonas aeruginosa* (19 mm, 22 mm and 25 mm) and *Streptococcus pyogens* (17 mm, 20 mm and 23 mm). The fungi *Penicillium* sp. (15 mm, 17 mm and 20 mm) showed more inhibitory activity when compared to *Aspergillus niger* (12 mm, 14 mm and 18 mm). No zone of inhibition was observed in the negative DMSO control.

3.3.2. Chloroform extract

The antimicrobial activity of chloroform extract of *Tridax procumbens* was tested and the results were tabulated in Table – 5. Among the three concentrations (100 mg/ml, 200, 300 mg/ml) used, maximum inhibitory zone was observed at 300 mg/ml followed by 200 mg/ml and 100 mg/ml. Maximum antibacterial activity was noticed in the bacteria *Bacillus subtilis* (23 mm, 27 mm and 33 mm) followed by *Staphylococcus aureus* (20 mm, 21 mm and 23 mm), *Pseudomonas aeruginosa* (14 mm, 17 mm and 20 mm) and *Streptococcus pyogens* (12 mm, 15 mm and 18 mm). The fungi *Penicillium* sp. (10 mm, 12 mm and 15 mm) showed more inhibitory activity when compared to *Aspergillus niger* (7 mm, 9 mm and 11 mm). No zone of inhibition was observed in the negative DMSO control.

3.3.3. Ethyl acetate extract

The antimicrobial activity of ethyl acetate extract of *Tridax procumbens* was investigated and the results were furnished in Table – 6. Among the three concentrations (100 mg/ml, 200, 300 mg/ml) used, maximum inhibitory zone was observed at 300 mg/ml. Maximum antibacterial activity was recorded in the bacteria *Bacillus subtilis* (20 mm, 24 mm and 30 mm) followed by *Staphylococcus aureus* (17 mm, 18 mm and 20 mm), *Pseudomonas aeruginosa* (11 mm, 14 mm and 17 mm) and *Streptococcus pyogens* (9 mm, 12 mm and 15 mm). The fungi *Penicillium* sp. (No zone, 9 mm and 12 mm) showed more inhibitory activity when compared to *Aspergillus niger* (No zone, No zone and 8 mm). No zone of inhibition was observed in the negative DMSO control.

Table - 4: Antimicrobial activity of methanol extract of *Tridax procumbens*

S.No	Organisms	Concentration of the extract (mg/ml) and zone of inhibition (mm)			
		DMSO Control	100 mg/ml	200 mg/ml	300mg/ml
1	*Staphylococcus aureus*	NZ	25 mm	26 mm	28 mm
2	*Streptococcus pyogens*	NZ	17 mm	20 mm	23 mm
3	*Pseudomonas aeruginosa*	NZ	19 mm	22 mm	25 mm
4	*Bacillus subtilis*	NZ	28 mm	32 mm	35 mm
5	*Aspergillus niger*	NZ	15 mm	17 mm	20 mm
6	*Penicillium* sp.	NZ	12 mm	14 mm	18 mm

NZ – No Zone

Table - 5: Antimicrobial activity of chloroform extract of *Tridax procumbens*

S.No	Organisms	Concentration of the extract (mg/ml) and zone of inhibition (mm)			
		DMSO Control	100 mg/ml	200 mg/ml	300 mg/ml
1	*Staphylococcus aureus*	NZ	20 mm	21 mm	23 mm
2	*Streptococcus pyogens*	NZ	12 mm	15 mm	18 mm
3	*Pseudomonas aeruginosa*	NZ	14 mm	17 mm	20 mm
4	*Bacillus subtilis*	NZ	23 mm	27 mm	33 mm
5	*Aspergillus niger*	NZ	10 mm	12 mm	15 mm
6	*Penicillium* sp.	NZ	7mm	9mm	10mm

NZ – No Zone

Table-6: Antimicrobial activity of ethyl acetate extract of *Tridax procumbens*

S.No	Organisms	Concentration of the extract (mg/ml) and zone of inhibition (mm)			
		DMSO Control	100 mg/ml	200 mg/ml	300 mg/ml
1	*Staphylococcus aureus*	NZ	17 mm	18 mm	20 mm
2	*Streptococcus pyogens*	NZ	9 mm	12 mm	15 mm
3	*Pseudomonas aeruginosa*	NZ	11 mm	14 mm	17 mm
4	*Bacillus subtilis*	NZ	20 mm	24 mm	30 mm
5	*Aspergillus niger*	NZ	NZ	9 mm	12 mm
6	*Penicillium* sp.	NZ	NZ	NZ	8 mm

NZ – No Zone

3.4. Antimicrobial activity of *Phyllanthus niruri*

3.4.1. Methanol extract

The antimicrobial activity of methanol extract of *Phyllanthus niruri* was determined and the results were given in Table – 7. Among the three concentrations (100 mg/ml, 200, 300 mg/ml) used, maximum inhibitory zone was observed at 300 mg/ml followed by 200 mg/ml and 100 mg/ml. Maximum antibacterial activity was noticed in the bacteria *Bacillus subtilis* (27 mm, 30 mm and 35 mm) followed by *Pseudomonas aeruginosa* (25 mm, 29 mm and 32 mm), *Staphylococcus aureus* (22 mm, 28 mm and 31 mm) and *Streptococcus pyogenes* (17 mm, 20 mm and 24 mm). The fungi *Penicillium* sp. (15 mm, 18 mm and 21 mm) showed more inhibitory activity when compared to *Aspergillus niger* (12 mm, 15 mm and 19 mm). No zone of inhibition was observed in the negative DMSO control.

3.4.2. Chloroform extract

The antimicrobial activity of chloroform extract of *Phyllanthus niruri* was evaluated and the results were presented in Table – 8. Among the three concentrations (100 mg/ml, 200, 300 mg/ml) used, maximum inhibitory zone was observed at 300 mg/ml followed by 200 mg/ml and 100 mg/ml. Maximum antibacterial activity was observed in the bacteria *Bacillus subtilis* (22 mm, 27 mm and 31 mm) followed by *Pseudomonas aeruginosa* (20 mm, 25 mm and 28 mm), *Staphylococcus aureus* (17 mm, 23 mm and 26 mm) and *Streptococcus pyogens* (12 mm, 15 mm and 19 mm). The fungi *Penicillium* sp. (10 mm, 13 mm and 16 mm) showed more inhibitory activity when compared to *Aspergillus niger* (No zone, 10 mm and 14 mm). No zone of inhibition was observed in the negative DMSO control.

3.4.3. Ethyl acetate extract

The antimicrobial activity of ethyl acetate extract of *Phyllanthus niruri* was investigated and the results were given in Table – 9. Maximum antibacterial activity was observed in the bacteria *Bacillus subtilis* (19 mm, 24 mm and 28 mm) followed by *Pseudomonas aeruginosa* (17 mm, 22 mm and 25 mm), *Staphylococcus aureus* (14 mm, 20 mm and 23 mm) and *Streptococcus pyogens* (9 mm, 12 mm and 16 mm). The fungi *Penicillium* sp. (No zone, 9 mm and 11 mm) showed more inhibitory activity when compared to *Aspergillus niger* (No zone, 10 mm and 13 mm). No zone of inhibition was observed in the negative DMSO control.

Table - 7: Antimicrobial activities of methanol extract of *Phyllanthus niruri*

S.No	Organisms	Concentration of the extract (mg/ml) and zone of inhibition(mm)			
		DMSO Control	100 mg/ml	200 mg/ml	300 mg/ml
1	*Staphylococcus saureus*	NZ	22 mm	28 mm	31 mm
2	*Streptococcus pyogens*	NZ	17 mm	20 mm	24 mm
3	*Pseudomonas aeruginosa*	NZ	25 mm	29 mm	32 mm
4	*Bacillus subtilis*	NZ	27 mm	30 mm	35 mm
5	*Candida albicans*	NZ	12 mm	15 mm	19 mm
6	*Penicillium* sp.	NZ	15 mm	18 mm	21 mm

NZ – No Zone

Table - 8: Antimicrobial activities of chloroform extract of *Phyllanthus niruri*

S.No	Organisms	Concentration of the extract (mg/ml) and zone of inhibition (mm)			
		DMSO Control	100 mg/ml	200 mg/ml	300 mg/ml
1	*Staphylococcus aureus*	NZ	17 mm	23 mm	26 mm
2	*Streptococcus pyogens*	NZ	12 mm	15 mm	19 mm
3	*Pseudomonas aeruginosa*	NZ	20 mm	25 mm	28 mm
4	*Bacillus subtilis*	NZ	22 mm	27 mm	31 mm
5	*Aspergillus niger*	NZ	NZ	10 mm	14 mm
6	*Penicillium* sp.	NZ	10 mm	13 mm	16 mm

NZ – No Zone

Table - 9: Antimicrobial activity of ethyl acetate extract of *Phyllanthus niruri*

S.No	Organisms	Concentration of the extract (mg/ml) and zone of inhibition (mm)			
		DMSO Control	100 mg/ml	200 mg/ml	300 mg/ml
1	*Staphylococcus aureus*	NZ	14 mm	20 mm	23 mm
2	*Streptococcus pyogens*	NZ	9 mm	12 mm	16 mm
3	*Pseudomonas aeruginosa*	NZ	17 mm	22 mm	25 mm
4	*Bacillus subtilis*	NZ	19 mm	24 mm	28 mm
5	*Aspergillus niger*	NZ	NZ	9 mm	11 mm
6	*Penicillium* sp.	NZ	NZ	10 mm	13 mm

NZ – No Zone

4. Conclusion

The study of antibacterial activity of herbal plant extract of *Coccinia grandis*, *Tridax procumbens* and *Phyllanthus niruri* showed that the methanol extract showed promising antimicrobial activity against bacterial and fungal human pathogens followed by chloroform extract and ethyl acetate extract. Among the three plants, maximum inhibition activity was exhibited by *Phyllanthus niruri* followed by *Tridax procumbens* and *Coccinia grandis*. The results also indicated that scientific studies carried out on medicinal plants having traditional claims of effectiveness might warrant fruitful results. These plants could serve as useful source of new antimicrobial agents

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