Antibacterial Activity and Phytochemical Screening of Different Parts of *Moringa oleifera* Against Selected Gram Positive and Gram Negative Bacteria

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**ABSTRACT**

The antibacterial activity and phytochemical screening of different extracts of *Moringa oleifera* Leaves, Flower, Pulp & Seed was investigated against two test bacteria *Escherichia coli* and *Staphylococcus aureus*. The antibacterial activity was performed by Agar well diffusion method. Four solvents 70% Ethanol, 80% Methanol, Petroleum ether & Aqueous extracts were used for extraction of the active ingredients present in plant. 80% Methanol extract showed maximum zone of inhibition against *E.coli* (28 mm) & *S.aureus* (26mm) in Leaves. 70% Ethanol extract showed maximum zone of inhibition against *E.coli* (23 mm) & *S.aureus* (17mm) in Flower. In pulp, 80% Methanol extract showed maximum zone of inhibition against *E.coli* (15.33 mm), while in *S.aureus*, Aqueous extracts showed maximum zone of inhibition (18.33 mm). In Seed, 80% Methanol extract showed maximum zone of inhibition against *E.coli* (18.33 mm) & in *S.aureus*, 70% Ethanol extracts showed maximum zone of inhibition (15.66 mm). The Qualitative phytochemical screening indicated the maximum presence of phytochemical constituents Tannin, Saponin, Phenol & Flavonoid with different extracts except petroleum ether in Leaves compared to Flower, Pulp & Seed. Therefore, present study confirms that 70% Ethanol & 80% Methanol had higher antibacterial activity compared to Petroleum ether & Aqueous extracts of Leaves, Flower, Pulp & Seed.

Keyword: *Moringa oleifera*; antibacterial activity; phytochemical screening; solvent extracts; agar well diffusion method

**INTRODUCTION**

*Moringa Oleifera* (*lam*) is considered a complete food as it has an impressive range of medicinal uses with high nutritional value. Its multiple pharmaceutical effects are capitalized as therapeutic remedy for various diseases in traditional medicinal system, the extracts of the
leaves are known to have biological properties and these are usually found to vary with the type of solvent used to extract the active components. The plant has been reported to possess antimicrobial properties and this explains the reason for its wide use in the treatment of human respiratory tract diseases [1, 2].

The increasingly high numbers of bacteria that are developing resistance to classical antibiotics drive much of the current interest on natural antimicrobial molecules in hope that they may provide useful leads into anti-infective drug candidates. Several antimicrobial agents were isolated from plant including secondary metabolites as essential oil and terenoides, amongst which can be cited xanthones, benzophenones, coumarins and flavonoids. These new chemical substances can also serve as templates for producing more effective drugs through semi-synthetic and total synthetic procedure. About 74% of 119 plant-derived pharmaceutical medicines or biotechnology medicines are used in modern medicine in ways that correlate directly with their traditional uses [3].

*Moringa oleifera* leaf ethanolic extract had the broadest spectrum of activity on the test bacteria [4]. Antimicrobial activity from the ethanolic extract of leaves, seeds and flowers of *Moringa oleifera* against microorganisms like Escherichia coli, Klebsiella pneumoniae, Enterobacter spp, Proteus mirabilis, Pseudomonas aeroginosa, Salmonella typhi A, Staphylococcus aureus, Streptococcus and Candida albicans [5].

In traditional Indian medicine various parts of the tree are used therapeutically for treatment of venomous bites, ascites and rheumatism and helps in lowering blood pressure. The root and bark of young trees are considered rubefacient, stomachic carminative, vesicant and abortifacient. The flowers and roots contain an antibiotic that is highly effective in the treatment of cholera. Moringa leaves have been reported to contain more vitamin A than carrots, more calcium than milk, more iron than spinach, more vitamin C than oranges, and more potassium than bananas, and that the protein quality of Moringa leaves rivals that of milk and eggs. The nutritional properties of Moringa are now so well known that there seems to be little doubt of the substantial wealth benefit to be realized by consumption of Moringa leaf powder in situation where starvation is imminent [6].

The present study is an attempt to explore the Antibacterial activity & Phytochemical screening of 70% Ethanol, 80% Methanol, Petroleum ether & Aqueous extracts of Leaves, Flower, Pulp & Seed of the plant. The Antibacterial activity of above extracts was evaluated by using strains of gram positive & gram negative bacteria.

**MATERIALS AND METHODS**

**Plant materials**

The plant material of *Moringa oleifera* (Fresh leaves, flower, seed and pulp) used in study were collected from “National Research Centre on Medicinal and Aromatic plants”, Boriavi, Anand, Gujarat, India, December 2014.

**Preparation of plant extracts**

Fresh material of the plant samples were collected, cleaned, washed, air dried and homogenized to a fine powdered using mechanical stirrer and stored in airtight bottle. 10gm of collected powdered form of leaves, flower, seed and pulp weighed and extracted with 70% Ethanol, 80% Methanol, Petroleum ether & Water extracts by soaking the powdered materials in 30 ml of solvents & kept for 24 hours on a rotary shaker. After that, it was filtered through Whatman no.1 filter paper. The filtrate was allowed to evaporate until dry. The extracts were kept in sterile bottles under refrigerated condition until use.

**Maintenance and identification of test bacteria**
Bacterial cultures of Gram negative Bacteria *Escherichia coli*, Gram positive Bacteria *Staphylococcus aureus* were obtained from Microbial Type Culture Collection Center (MTCC), Chandigarh, India. These bacteria were maintained on nutrient broth at 37°C and were cultured on nutrient agar to perform Antibacterial activity.

**Antibacterial activity**

The antibacterial assay of 70% Ethanol, 80% Methanol, Petroleum ether & Aqueous extracts was performed by Agar well diffusion method using nutrient agar medium [7]. The media (Nutrient Agar- Hi media) was poured into the petriplates and inoculated with bacterial cultures. For the agar well diffusion method, a well was prepared in the plates with a cup borer (1.2 cm) and 100μl of the extracts was inoculated directly into the well. For control, two different antibiotics (Ampicillin 0.2 mg/ ml & Tetracycline 0.2 mg/ml) were filled in the wells with the help of micropipette.

**Phytochemical screening**

Phytochemical analysis was done to screen the extracts for the presence of the following active ingredient: Tannins, saponins, phenol, flavonoid, anthraquinone [8, 9].

**RESULTS**

The present study was on the determining antibacterial activity using agar well diffusion method by measuring the inhibition zone in mm against two bacterial strain *E.coli* & *S.aureus* and phytochemical screening in different parts like Leaves, Flower, Pulp, Seed of *Moringa oleifera* with different solvents like water, 70% ethanol, 80% methanol and petroleum ether.

The result reveals that solvent extracts of the different parts of *Moringa oleifera* were active against the test organisms.

Fig. 1 shows that in *Moringa oleifera* leaves, all extracts had higher antibacterial activity against *E.coli* compared to *S.aureus*. 70% Ethanol showed highest antibacterial activity against *E.coli* while 80% Methanol showed highest antibacterial activity against *S.aureus*. In *Moringa oleifera* Flower, 70% Ethanol & 80% Methanol had higher antibacterial activity against *E.coli* & *S.aureus* both. 70% Ethanol showed highest antibacterial activity against *E.coli* whereas 70% Ethanol & 80% Methanol both have antibacterial activity against *S.aureus*. In *Moringa oleifera* Pulp, Water & 70 % Ethanol had highest antibacterial activity against *S.aureus* compared to *E.coli*. 80% Methanol had higher antibacterial activity against *E.coli* & Water extract showed highest antibacterial activity against *S.aureus*. In *Moringa oleifera* Seed, 80% Methanol both had higher antibacterial activity against *E.coli* but 70% Ethanol showed highest antibacterial activity against *S. aureus*.

**PHYTOCHEMICAL SCREENING**

Table 1 shows the results of the phytochemical screening of the different *Moringa oleifera* parts (Leaves, Seed, Pulp and Flower). The result reveals the presence of tannins, saponins, flavonoid & phenol in all extracts except petroleum ether whereas Anthraquinone is absent in all extracts of *Moringa oleifera* leaves. In *Moringa oleifera* flower, tannin & phenol were present in 70% ethanol & 80% Methanol. Flavonoid was present in water extract while Saponin was present in all extracts except 80% methanol. Anthraquinone was absent in all extracts. In *Moringa oleifera* Pulp, tannins, phenol & saponins were present in all extracts. Flavonoid & Anthraquinone were absent in all extracts. There were no phytochemical constituent presents in seed.
Fig. 1: Zone of inhibition of extracts of *Moringa oleifera* Leaves, Flower, Pulp & Seed

Table 1: Screening of phytochemicals in *Moringa oleifera* Leaves, flower, Pulp, Seed in different extracts.

<table>
<thead>
<tr>
<th>Phytochemical Constituents</th>
<th>Leaves</th>
<th>Flower</th>
<th>Pulp</th>
<th>Seed</th>
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<tr>
<td></td>
<td>W</td>
<td>E</td>
<td>M</td>
<td>P</td>
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<tr>
<td>TANNIN</td>
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<td>PHENOL</td>
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<td>FLAVONOID</td>
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<td>SAPONINS</td>
<td>+</td>
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<tr>
<td>ANTHRAQUINONE</td>
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W: Water, E: 70% Ethanol, M: 80% Methanol, P: Petroleum ether

**DISCUSSION**

Various researchers reported antimicrobial activity of *Moringa oleifera* against variety of pathogens including *S. aureus*, *S. albus*, *S. pyogenes*, *P. aeruginosa*, *Salmonella gallinarum*, *B. subtilis* and *E. coli* [10, 11, 12]. Antimicrobial activity from the seeds of *Moringa oleifera* were assayed for the evaluation of antimicrobial activity against bacterial strains (*Pasteurella multocida*, *Escherichia coli*, *Bacillus subtilis* and *Staphylococcus aureus*), which was evaluated by Amer Jamil et al. (2008) [13].

Antimicrobial activity from the ethanolic extract of leaves, seeds and flowers of *Moringa oleifera* against microorganisms like *Escherichia coli*, *Klebsiella pneumoniae*, *Enterobacter* spp, *Proteus mirabilis*, *Pseudomonas aeruginosa*, *Salmonella typhi A*, *Staphylococcus aureus*, *Streptococcus* and *Candida albicans* was reported by R.E. Renitta et al. (2009) [14]. Phytochemicals are present in virtually all plant tissues of *Moringa oleifera* e.g. leaves, roots, stem and fruits [15]. Presence of phytoconstituents like alkaloids, flavonoids, tannins, saponins are responsible for antibacterial activity [16].

**CONCLUSION**

This study revealed that *Moringa oleifera* leaves have greater antibacterial activity and phytochemical constituents. *Moringa oleifera* parts could be good source of drugs that may be used against bacterial infection if it is found...
effective and nontoxic in animal trial. Detailed study is needed to investigate the active compounds present in these plant parts having antibacterial activity that may help us to design more effective chemotherapeutic agent to heal bacterial infections.

CONFLICT OF INTEREST STATEMENT
The authors declare that they have no competing interests.

REFERENCES