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# Antimicrobial activity of aqueous and methanolic extracts of *Aegle marmelos*

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# ABSTRACT

Methanolic and aqueous extract of *Aegle marmelos* leaves were evaluated for antimicrobial activity against common human pathogens. The findings have proved that aqueous extract of *Aegle marmelos* was more effective in inhibiting all the five test pathogens with zone of inhibition ranging between 31 mm and 51 mm as compared to methanolic leaf extract (14 to 39 mm). The results indicated that the methanolic and aqueous leaf extracts of *Aegle marmelos* might be exploited as natural drug for the treatment of several infectious diseases caused by these organisms. The present study justifies the antimicrobial uses of *A. marmelos* in the traditional system of medicine to treat various infectious diseases.

**Key words:** *Aegle marmelos*, Methanolic Extract, Aqueous extract, Antimicrobial activity.

# INTRODUCTION

Historically, medicinal plants have provided a source of inspiration for novel drug compounds, as plant derived medicines have made large contributions to human health and well being. *Aegle marmelos* is an important medicinal plant of India and are reported to have various medicinal properties in traditional medicinal systems. The medicinal properties of *Aegle marmelos* plant have been described in the Ayurveda and in a great Vedic literature. Ancient literature such as Rigveda, Yajurveda, Atharvaveda, Charak Samhita and Sushrut Samhita also describes the use of this plants for the treatment of various health problems (Meena *et al.*2016).

*Aegle marmelos* belonging to Family Rutaceae is commonly called as Bael or Belphal (Sanskrit – Bilwa), possess various medicinal properties like anti-diarrhoeal, immune-stimulant and antiinflammatory agent. Leaf extracts are helpful to cure inflammation. *Aegle marmelos* leaf juice with honey can prove useful for treating fever. It can be used to treat tuberculosis. It is rich in anti-oxidants which helps in insulin secretion which leads to low blood sugar levels. The extracts of the various parts of the plant is used for controlling diarrhoea, dysentery, dyspepsia, stomach pain, seminal weakness, uropathy, vomiting, diabetes, snake bite, fish poisoning and some antiviral activities (Krithikar and Basu,1999).

During present investigation, the aqueous and methanolic extracts of Aegle marmelos were evaluated for antimicrobial activity against common human pathogens. Many plant species have been evaluated for the antimicrobial activity. Therefore, it is necessary to establish the scientific basis for the therapeutic action of these plants. More than hundreds of phytochemical compounds have been isolated from various parts of the plant, namely phenols, flavonoids, alkaloids, cardiac glycosides, saponins, terpenoids, steroids, and tannins (Mujeeb et al.2014). Leaves are considered to be one of the highest accumulatory parts of the plant containing bioactive compounds which are synthesized as secondary metabolites (Cowan, 1999). Considering the uses of A. marmelos in traditional system of medicine, an attempt is made to assess the antibacterial activity of leaf extracts.

The present study is an attempt to evaluate the antibacterial activity of leaves of *A. marmelos.* Similar study of plant has been undertaken by Poonkothai and Saravanan (2008) and Suresh et. al. (2009) from Tamil Nadu, Pandey and Mishra (2011) from Lucknow, Rathod et. al. (2013) from Gujrat, Mujeeb et. al. (2014) from Faizabad, Meena et. al. (2016) from Jaipur and Gupta et. al. (2018) from Bhopal.

In the past, many researchers have evaluated the pharmaceutical importance of different parts of *Aegle marmelos* plant and found that the plant possesses natural healing capacity in curing many ailments along with antiinflammatory, antipyretic, anti-diarrhoeal, anti-diabetic, analgesic, antimicrobial, radio protective, anticancer and antiviral properties (Gupta *et al.*2018).

#### **MATERIAL AND METHODS**

Methanolic and aqueous extract of *Aegle marmelos* leaves were evaluated for antimicrobial activity. The plant leaves were collected from different localities in Hingoli district during January 2018 to December 2019. The leaves were washed thoroughly with water, dried in Oven and crushed in to a powder. 25 gm plant

powder was soaked in 100 ml of each of methanol and water, and allowed to stand for 72 hrs followed by filtration. The methanolic and aqueous extracts were obtained after filtration and then were kept for water bath at 40 and 80 °C respectively till evaporation and weighed after it. These extracts were dissolved in DMSO (Dimethyl Sulphoxide) at the concentration of 1 ml DMSO/ mg extracts, and stored at  $4^{\circ}$ C in a refrigerator (Satish and *et al.*2008).

The various human pathogenic microorganisms were procured from National Center for Microbial Resource (NCMR), Pune which included Gram positive bacteria, *Streptococcus mutans, Pseudomonas aeruginosa, Staphylococcus aureus* and Gram-negative bacterium *Escherichia coli* and a yeast *Candida albicans*. All the slants were kept at 25°C in the refrigerator for further studies.

The microbial inoculum was standardized at 0.5 McFarland (Andrews, 2001). Antimicrobial activities of aqueous and methanolic leaf extracts were tested using agar well diffusion method (Khan, *et al.*1988) and the inhibition zones were measured using a Himedia zone scale.

#### **RESULTS AND DISCUSSION**

The results obtained are presented in Table 1, which clearly indicates anti-microbial activity of methanol and aqueous extract of *Aegle marmelos*. The aqueous extract showed maximum antimicrobial activity against the test microbes with zone of inhibition lying within the range of 31 mm to 51 mm. Methanolic extract also showed significant antimicrobial activity with the zone of inhibition lying in the range of 14 mm to 39 mm. Ciprofloxacin was used as positive control, which produced the inhibition zones ranging between 10 and 35 mm whereas *C. albicans* showed zone of inhibition of 21 mm to Ketoconazole which was used as positive antifungal antibiotic.

The results thus indicated that the aqueous and methanolic leaf extracts of *Aegle marmelos* might be exploited as a natural drug for the treatment of several infectious diseases caused by these pathogens. The findings of the present investigation offer a scientific support to the ethnomedicinal use of the plant by the traditional healers.

Sample	Solvent	Zone of inhibition (mm)				
		Escherichia	Pseudomonas	Staphylococcus	Streptococcus	Candida
		coli	aeruginosa	aureus	mutans	albicans
Aegle marmelos	Methanol	39	35	14	37	31
Aegle marmelos	Aqueous	51	41	31	40	41
Positive control	Antibiotic	14	10	16	35	21
Negative control	DMSO	-ve	-ve	-ve	-ve	-ve

Table 1. Antimicrobial activity of aqueous and methanolic extracts of Aegle marmelos.

The ability of the plant extracts of *Aegle marmelos* to inhibit growth of bacteria and fungi is an indication of its broad-spectrum antimicrobial activity which could be a potential source for development of novel antimicrobial agents (Mujeeb, et. al., 2014).

The findings are similar to the results of leaf extracts of *A. marmelos* which shows varying degree of antibacterial activity has reported by Rastogi and Mehrotra (1993), Pandey and Mishra (2011) and Gupta et. al. (2018). It is reported that the presence of compounds Cuminaldehyde and Eugenol present in the leaves may be responsible for antibacterial activity. It was reported earlier by Mohan et. al. (2005). that aqueous, acetone and petroleum ether extracts of *A. marmelos* were more effective against *B. coagulans, B. subtilis, B. thuringiensis, P. aeruginosa* and *S. aureus*.

*Escherichia coli* was the most susceptible bacterium, on observation that may be attributed to the presence of Tannins alkaloids, inhibit the growth of microorganisms. The *Aegle marmelos* leaves extract, show highly active against the microorganisms. *E. coli* followed by *Salmonella typhi, Staphylococcus aureus, Proteus mirabilis, Pseudomonas aeruginosa* as earlier reported by Suresh et. al. (2009).

The screening of antibacterial activity was carried out by disc diffusion method against the microorganisms such as *Escherichia coli, Salmonella typhi, Pseudomonas aeruginosa, Proteus vulgaris* and *Staphylococcus aureus* by Rathod et. al. (2013). Similar human pathogenic bacteria are considered for the present investigation.

The antibacterial assay done by Meena et. al. (2016) for screening purpose of selected gram positive and gram negative microorganisms showed zone of inhibition against test plant extract. Among these test micro-organisms are the most susceptible to methanol extract of *Aegle marmelos* as reported by Meena et. al. (2016). similar findings are seen in present work.

On the basis of the result obtained in this present investigation, it can revealed that, the methanol extracts of *Aegle marmelos* leaves had significant *in vitro* antimicrobial activity and the most active extracts can be farther subjected to isolation and identify therapeutic antimicrobials and undergo further pharmacological evaluation.

## CONCLUSION

From the research findings, it can be concluded that *Aegle marmelos* plant has immense potential to be used in pharmacology and as a prospective source of valuable drugs. Due to the presence of various compounds that are essential for good health, it can also be used to improve the health problems. The extracts showed a significantly high antibacterial activity against the microorganisms. The data clearly depicts the presence of compounds used for treating various bacterial diseases, indicating its use in the traditional system of medicine since ancient times. Further, the broad-spectrum activity of aqueous and methanol extracts proves to be encouraging in the development of novel antimicrobial formulation in future.

**Conflicts of Interest**: The authors declare no conflict of interest.

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