

ANTIMICROBIAL ACTIVITY OF LEAF EXTRACT OF *MORUS*
INDICA (MULBERRY) FROM CHHATTISGARH

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ABSTRACT

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The Methanolic and Ethanolic extract of *Morus indica* was screened against five different pathogens and showed significant antimicrobial activity against *Staphylococcus aureus*, *Aspergillus niger* and *Penicillium*. It has been observed that ethanolic extract has maximum antibacterial activity against *S.aureus* (12mm). Among both solvents methanolic extracts has maximum antifungal. Activity against *Aspergillus* (30mm) followed by *Penicillium* (29mm). The present study shows that mulberry (*Morus indica*) leaf possess antimicrobial property as well as antifungal activity.

KEYWORDS: Antifungal activity, Antibacterial activity, Mulberry.

INTRODUCTION

Medicinal plants are the back bone of traditional systems of medicine in India. Pharmacological studies have acknowledged the value of medicinal plants as a potential source of bioactive compounds (Prusti *et al.*, 2008). Medicinal plants are of great importance to the health of individuals and communities. The medicinal value of plants lies in some chemical substances that produce a definite physiological action on the human body. According to World Health Organization, medicinal plants would be the best source to obtain a variety of drugs. About 80% of individuals from developed countries use traditional medicine, which has compounds derived from medicinal plants (Senthil kumar *et al.*, 2009). The use of plant extracts and

phytochemicals, both with known antimicrobial properties, can be of great significance in therapeutic treatment. Many plants have been used because of their antimicrobial traits, which are due to compounds synthesized in the secondary metabolism of the plant. These products are known by their active substance, for example, the phenolic compounds which are part of the essential oils, flavonoids, alkaloids, as well as tannin (Thenmozhi *et al.*, 2010).

Infectious diseases accounts for high proportion of health problems in the developing countries including India. Microorganisms have developed resistance to many antibiotics and as a result, immense clinical problem in the treatment of infectious diseases has been created. The resistance of the organisms increased due to indiscriminate use of commercial antimicrobial drugs, commonly used for the treatment of infectious disease. This situation forced the researchers to Search for new antimicrobial substance from various sources including medicinal plants. Keeping in view the importance of medicinal value, the current study was done on mulberry (*Morus spp.*). Black mulberry (*Morus indica*) is a fruit not known only for its nutritional qualities and its flavour, but also for its traditional use in natural medicine as it has a high content of active therapeutic compounds. The most important bioactive compounds of plants are alkaloids, flavonoids, tannins and phenolic compounds. The phytochemical research based on ethno-pharmacological information is generally considered as an effective approach in the discovery of new anti – infective agents from higher plants (Bauer *et al.*, 1966). Thanks to the richness of this active components the *Morus alba*, which belongs to the *Moraceae* family has become the most precious resource in varied branches of industry, especially to get bioactive substances useful in medicine and pharmacy as well as food and cosmetic production.

In the present investigation the antimicrobial study of *Morus indica* from both methanolic and ethanolic leaf extracts were evaluated as antimicrobial agents against the growth of bacterial strain *Staphylococcus aureus* ATCC 25923, *Escherichia coli* ATCC 25922, *Pseudomonas aeruginosa* ATCC 27853 and fungal strain *Aspergillus* and *Penicillium* species.

MATERIALS AND METHODS

Collection of Samples: Healthy mulberry leaves were collected from the plants present in campus of Devleela Biotech, Raipur, Chhattisgarh during the month of October- January, 2014.

Preparation of samples: *Morus indica* (mulberry) leaf samples collected were washed thoroughly with tap water followed by distilled water, then wiped and dried under shade followed by oven drying at 60° C - 65° C till constant weight was attained. Completely dried leaf samples were ground using an electric blender to obtain a fine powder. The powder was further passed through successive cycles using Soxhlet apparatus using the two solvent systems (Methanol and Ethanol), 200 ml of each solvent with 20 gm dried plant material was used for extraction through Soxhlet apparatus. The resulting extracts were filtered and concentrated in vacuum evaporator (Roteva:Medical Instrument Mfg.Co.). The concentrated extracts were then used to determine the antimicrobial activity.

Antimicrobial activity of methanolic and ethanolic leaf extract of *Morus indica* .:

Anti – bacterial Assay-

Antibacterial activity was carried out Using disc diffusion method. For the preparation of inoculation, the tested bacteria were cultured in nutrient broth at 37⁰ C for 24 hours. 20 µl of prepared culture was spread on the surface of nutrient agar media for bacterial pathogens. Then two Well were mad on the surface of nutrient agar with the help of borer. One well contains only solvent used as negative controls and another well contains 100 µl of leaf extract. Susceptibility test disc of two antibiotics Penicillin- G (10 units / disc) and Streptomycin (10 mcg / disc) (Himedia) were used as positive controls. Then the plates were incubated for 37⁰ C for 24 hours. The antibacterial activity was evaluated by measuring the diameter of inhibition zone to the nearest size in mm.

Anti – fungal Assay –

Anti - fungal activity was carried out using agar - diffusion method. For the preparation of the inoculation, the tested fungal cultures were cultured in Sabouraud dextrose broth at 28⁰ C for 48 – 72 hours. After that 20 µl of prepared culture was spread on the surface of Sabouraud dextrose agar for fungal pathogens. Then four wells were

made on the surface of Sabouraud dextrose agar plate with the help of borer. One well contain only solvent, and was used as negative control. Second well contains 100 μ l of leaf extract. Third well contains antibiotic fluconazole (Tablet IP 150mg, Ecogen) (1mg / ml) used as positive control. The plates were allowed to stand for at least one hour for diffusion to take place and then incubated at 37⁰ C for 24 hrs. The antifungal activity was evaluated by measuring the diameter of inhibition zone to the nearest size in mm.

RESULT AND DISCUSSION

The Ethanolic extract of *Morus indica* (mulberry) was the most potent against *Staphylococcus aureus* with largest diameter of inhibition zone i.e. 15 mm whereas, methanolic extract showed smallest zone of inhibition i.e. 12 mm. Ethanolic and methanolic none of the extract showed any resistance against *E.coli*. The methanolic and ethanolic extract of mulberry and both the antibiotic (*Penicillin* and *Streptomycin*) does not show any zone of inhibition against the pathogen *Escherichia coli* and *Pseudomonas aeruginosa*.

The methanolic leaf extract of *Morus Indica* (Mulberry) was the most potent against the two fungus *Aspergillus* and *Penicillium* species with largest diameter of zone of inhibition 30mm and 20mm, respectively while ethanolic extract showed the smallest zone of inhibition 20mm against the two fungal colonies.

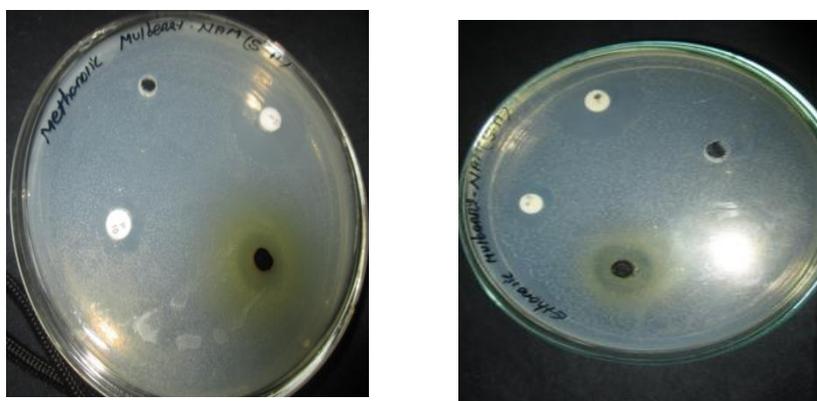


Figure 1: Antibacterial activity against S.aureus

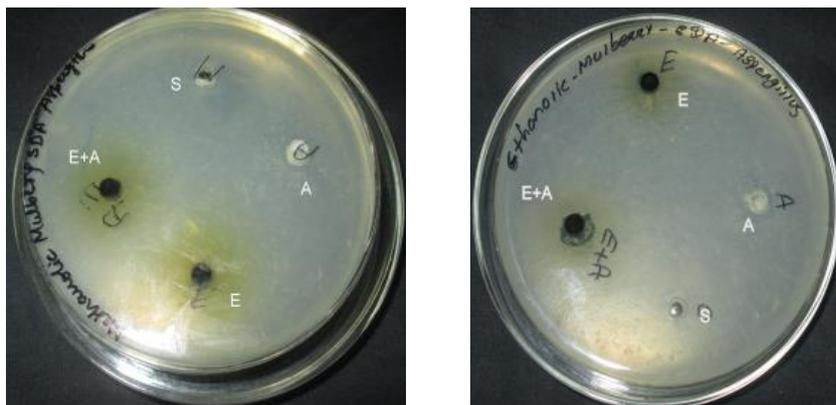


Figure 2: Antifungal activity against *Aspergillus niger*



Figure 3: Antifungal activity against *Penicillium*

Table 1: Antibacterial activity of *Morus indica* (Mulberry) extracts by measuring zone of inhibition diameter (mm).

Test pathogen	Solvent	Extract (100µl)	Penicillin-G (10mcg/disc)	Streptomycin-S10 (10unit/disc)
<i>Escherichia coli</i>	Methanol	—	—	—
	Ethanol	—	—	—
<i>Pseudomonas aeruginosa</i>	Methanol	2	—	—
	Ethanol	2	—	—
<i>Staphylococcus aureus</i>	Methanol	12	10	18
	Ethanol	15	12	20

Table 2: Antifungal activity of *Morus indica* (Mulberry) extract by measuring zone of inhibition diameter (mm).

Test pathogen	Solvent	Extract(100µl)	Fluconazole
<i>Aspergillus</i> sp.	Methanol	30	11
	Ethanol	20	10
<i>Penicillium</i> sp.	Methanol	29	10
	Ethanol	20	8

CONCLUSION

The present study and data revealed that the antimicrobial activity of leaf extract of *Morus indica* was found to be the best against bacterial and fungal cultures, and also proved that the factors which are found in the form of secondary metabolites were responsible for antimicrobial activity. The future prospect for this study was to analyze the purified compound for drug validation. However further research is needed *in-vitro* as well as *in - vivo* and the extracts against other different types of microorganism to reach a better conclusion.

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