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Existence of Antimicrobial Activities in *Mentha arvensis* Against Some Pathogenic Bacteria

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ABSTRACT

Mentha plant is erect, perennial branched herb which is used to treat various types of human disorders. Ethanolic and aqueous extracts of *M. arvensis* (L.) were tested to their actions against two selected bacteria namely *Citrobacter freundii (gram-)* and *Micrococcus luteus (gram+)*. The antibacterial effects was assessed by using method of disc diffusion at various concentrations. The findings showed that ethanol was finest solvent used to extraction as compared to aqueous for testing the antibacterial properties of leaf and stem extracts. *Mentha arvensis (L.)* extract showed maximum zone of inhibition against both bacterial strains. Highest zone of inhibition 19.0mm was found in *Citrobacter freundii* at highest concentration, while maximum inhibition by 12.5mg/ml ethanolic stem extract was 11mm against *Citrobacter freundii and* maximum inhibition of *Micrococcus luteus was 9 mm* due to an ethanolic leaf extract of *Mentha arvensis* (L.), and 7.3mm by 12.5 mg/ml ethanolic stem extract. The results also revealed that ethanolic leaf extract of *Mentha arvensis* (L.) inhibited the population of tested pathogens considerably as compared to other ethanolic extract. The findings suggested that more research should be done on these plants and support their application in traditional medicine.

Key Words: *Mentha arvensis* (L.), Antimicrobial properties, Medicinal plant, Disc diffusion method, Ethanolic extract.

INTRODUCTION

In tropical and subtropical areas, infectious illnesses brought on by bacterial and fungal infections are the main causes of death. (Zhang et al,2015). India is one of the wealthiest nations in the world when it comes to the genetic resources of medicinal plants. Its topography and climate vary greatly, which has an impact on the vegetation and floristic makeup of the region. Furthermore, the agroclimatic conditions are favourable for bringing in and domesticating new exotic plant types. (Martins *et al,* 2001). Medical plants are living, irreversible resources that may be depleted if overused but can be sustained if used carefully and wisely. In the past, people have neglected the value of medicinal herbs. Nevertheless, nowadays, medicinal plants are valued as both a source of economical health treatment and a source of money (Purohit and Vyas, 2004). Secondary metabolites are responsible for the plant therapeutic effects. These metabolites provide plants used in the food and pharmaceutical sectors with colour, taste, and scent (Vivek *et al,* 2009 ; Nascimento *et al,* 2010; Alami *et al,* 2022; Wei *et al*, 2023). *Mentha arvensis* often called menthol mint, is a perennial herb with upright branches that can reach a height of 75 cm. It has flowing rootstocks and inflexible branching stems. Asthma, jaundice, and illnesses of the liver and spleen are all treated with this herb. Natural menthol is produced from menthol mint, an essential oil-bearing crop, and is used extensively in the pharmaceutical, cosmetic, and flavouring sectors. (Chand *et al,* 2004; Vivek *et al,* 2009; Nascimento etal,2010; Thawkar *et al,* 2016; Nazim *et al*,2020). Consequently, the purpose of the current study was to examine the possible antibacterial activity of Mentha arvensis (L.) against two harmful microorganisms, *Citrobacter freundii* and *Micrococcus luteus*.

MATERIALS AND METHODS

Collection of *Mentha Arvensis*

Mentha arvensis (L.) aerial parts were collected in the Agra area of Uttar Pradesh. The

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plants individual sections were meticulously chopped with a cutter to remove any contaminating bits. The harvested plant components were then placed in plastic bags and sealed. A refrigerator was used to retain the specimens that were collected and delivered to the lab. In accordance with Srinivasan *et al* (2004), plant material were carefully transferred to H_2O , sterilised in chemicals includes 0.1% HgCl2, dried, pulverised, and packaged (Fig. 1).

Fig 1: *Mentha arvensis* (L.)

Extraction of Active Constituents

Aqueous Extract

Using mechanical method to prepare aqueous extracts. Different plant components, such as the leaf and stem, were individually homogenized before being filtered through muslin fabric. Further straining was done using Whattman No. 1 filter paper to produce the filtrate. At room temperature, the extraction was done. (Zore *et al,* 2004).

Organic Extract (Soxhlet Extraction)

The shade dried plant material was evenly transferred to thimble and placed in an chamber situated beneath a condenser & above the flask containing ethanol. The powdered material weighed about 100g in this extraction chamber. Once the flask reached 65 °C, the ethanol evaporated, changed into a liquid in the condenser and then pours in chamber containing the plant material. Every time solvent level surrounding the sample exceeded a predetermined threshold, the extraction chamber was designed to overflow and trickle back into the boiling flask. After the extraction process was complete, the extract flask taken out & remaining traces of ethanol was removed by rotary evaporator. Then, the extract was stored at 4°C in the refrigerator.(Okeke *et al*, 2001).

Test Micro-Organisms

Two selected pathogenic bacterial strains: *Citrobacter freundii* and *Micrococcus luteus* were taken from Institute of Microbial Technology, Chandigarh. A nutrient agar slant was used to maintain the typed bacterial culture, which was then kept at 40^{o} C until it was needed.

Antibacterial Activity Screening

Disc diffusion procedure was used to assess the in vitro antibacterial activity of a chosen plant extract. (Mukherjee *et al*, 1995). Plant extract solutions with varied concentrations (12.5 mg/ml, 6.25 mg/ml, 3.12 mg/ml, and 1.56 mg/ml) were generated by serial dilution to test susceptibility. Plant extract was diluted in preferred solvent. 6 mm-sized sterile discs were pouring in 25 µl of each successively diluted extract solution. A nutrient broth was combined with a few colonies from the pure culture. A cotton swab dipped in culture was used to inoculate this broth throughout the whole surface of the nutrient agar plate. With the use of sterile forceps, plant extracts containing discs were applied to the agar plate's infected surface. 24 hours at 37°C were spent incubating these plates. The measurement of the antibacterial activity was assessed by calculating the diameter of the zones of inhibition surrounding disc in petri plates. Mean value of zone of inhibition was determined in millimeter.

RESULTS AND DISCUSSION

In present study, disc diffusion method was used to evaluate the antibacterial effects of *Mentha arvensis* (L.) at various concentrations. Table 1–3 shows the effects of various doses of crude leaf and stem extracts, demonstrate that both aqueous and ethanolic extracts of leaves and stems were effective at preventing bacterial growth.

Table 1. Antibacterial activity of the aqueous extracts of *Mentha arvensis* (L.) against *Citrobacter freundii and Micrococcus luteus.*

Plant name	Aqueous extract against *Citrobacter freundii*	
Mentha arvensis (L.)	Leaf (zone of inhibition)	Stem (zone of inhibition)
	7.6 mm	6.3 mm
	Aqueous extract against *Micrococcus luteus*	
	Leaf (zone of inhibition)	Stem (zone of inhibition)
	7.5 mm	6.4 mm

Table 2. Antibacterial activity of the ethanolic extracts of *Mentha arvensis* (L.) against *Citrobacter freundii.*

Plant name	Different concentrations of Ethanolic extracts			
Mentha arvensis (L.)	Leaf extract against *Citrobacter freundii*			
	12.5 mg/ml	6.25mg /ml	3.12mg /ml	1.56 mg /ml
	19 .3	9.0	7.0	6.5
	Stem extract against *Citrobacter freundii*			
	12.5 mg /ml	6.25mg /ml	3.12mg /ml	1.56 mg /ml
	11	9.0	8.0	6.0

Table 3. Antibacterial activity of the ethanolic extracts of *Mentha arvensis* (L.) against *Micrococcus luteus*

Plant name	Different concentrations of Ethanolic extracts			
Mentha +arvensis (L.)	Leaf extract against *Micrococcus luteus*			
	12.5 mg/ml	6.25mg /ml	3.12mg /ml	1.56 mg /ml
	9.0	8.0	7.0	6.5
	Stem extract against *Micrococcus luteus*			
	12.5 mg	6.25mg /ml	3.12mg /ml	1.56 mg /ml
	7.3	6.8	6.2	6.0

As compared to aqueous extracts, *Mentha arvensis* (L.) shown considerable inhibitory action in ethanol extracts .Because (1) certain active chemicals were found in water extracts, albeit in small amounts, and (2) bio-active were soluble in organic solvents and hence absent from water extract. The plant extracts in the current investigation were shown to function in a dose-dependent way, exhibiting their highest level of activity at a dosage of 12.5 mg/ml.The above findings of our investigation showed that leaf extract from the test plant significantly inhibits the development of both test bacteria when compared to stem extract. Because most bioactive compounds are abundant in leaves that have been shown to have therapeutic effects as well as physiological and antibacterial properties. The above results was also showed that *Mentha arvensis* (L.) extracts significantly inhibited the growth of tested pathogens.

CONCLUSION

Plant extracts have a lot of promise as antimicrobial agents against bacteria and can be utilised to treat infectious diseases brought on by hard-to-treat germs.Several natural organic compounds had to be screened in order to find the

active agents because effective result –molecules estimation & identification of drugs will be beneficial in the drug development process.

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