Antimicrobial property of different parts of *Citrus limon*

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Abstract: Lemon (Citrus) are acidic fruits of family Rutaceae. Lemon is called the tree of life because of its various medicinal properties. Almost all parts of the fruit can be used against different diseases due to its wide range of medicinal property. Other than nutritional content they also have antioxidant, antimicrobial, insect repellent, medicinal and many other properties. Different biocompounds of the fruit can replace the function of

synthetic drugs which are available in market.

In the present study, the antimicrobial extent of crude juice, different extracts of leaf (matured and young), peel and standards streptomycin and tetracycline (1mg/mL) was determined against E. coli by agar diffusion method. The highest and minimum inhibition zone was found in crude juice (more than the standard) and matured leaf respectively. MIC for the crude juice was recorded as $0.01\mu L/mL$ (v/v) and for streptomycin and tetracycline was recorded as $0.1\mu g/mL$ and $0.01\mu g/mL$ respectively. The study clearly points out the antimicrobial potential emphasizing the importance of incorporating lemon as a regular component in diet. **Keywords -** inhibition zone, MIC, streptomycin, tetracycline, therapeutic treatments.

I. Introduction

Even though pharmacological industries have produced a number of new antibiotics in the last three decades, resistance to these drugs by microorganisms has increased. In general, bacteria have the genetic ability to transmit and acquire resistance to drugs, which are utilized as therapeutic agents (Gislene *et al.*, 2000). For a long period of time, plants have been a valuable source of natural products for maintaining human health. The use of plant extracts and photochemical, both with known antimicrobial properties can be of great significance in therapeutic treatments (Seenivasan *et al.*, 2006). 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 substances e.g. the phenolic compounds which are part of the essential oils, as well as tannin (Tyagi and Malik, 2010). Essential oils are more effective in controlling biofilm cultures due to their better diffusibility and mode of contact (Al-Shuneigat *et al.*, 2005). Hence the essential oils and other extracts of plants have evoked interest as sources of natural products. They have been screened for their potential uses as alternative remedies for the treatment of many infectious diseases (Tepe *et al.*, 2004).

Antimicrobial compound study is in great demand because microorganisms become resistant to most antibiotics and spread rapidly. Microorganisms are basically used for antibiotic production but plants also represent an important source of antimicrobial compounds. Different bioactive compounds are there, which contribute for the antimicrobial activity. Hammers *et al.* (1999) studied the effects of essential oils from *C. aurantium*, *C. limon*, *C. paradise* and many other plant oils and extracts and found that minimum inhibitory concentrations (MIC) were between 5-2% v/v.

II. Methodology

2.1 Collection of Samples: Fresh ripen samples of *Citrus limon* were collected from local market of Guwahati, Assam, India.

2.2 Extraction of Juice: The fruits were washed thoroughly in water and dried. The juices were extracted by manual juicer. The collected juice was centrifuged for 10,000 rpm to get a clear supernatant which was used for the present study.

2.3 Sample Preparation: A quantity (50 gm) of each fruit part was weighed separately and mixed with 250 ml of methanol described by Lopornik *et. al.*, 2005.

2.4 Microbial culture: Microbe from the stock was sub cultured in Nutrient broth. Present experiment was performed using 24 hours grown bacterial cultures at 37°C.

2.5 Evaluation of antimicrobial property: Antimicrobial study was performed to detect antimicrobial property of the different extracts of leaf (matured and young), peel in methanol and standards streptomycin and tetracycline (1mg/mL) was determined against *E. coli* by agar plate diffusion method. Nutrient agar plates (diameter 15cm) were prepared. 1mL of the bacterial culture was spread followed by boring 1.0cm well. Dilution of the juices was made to prepare the extracts for the present study. The concentrations of the juices used for the present study are crude juice (100% pure juice), 0.1μ L/mL, 0.01μ L/mL, 0.001μ L/mL, and 0.0001μ L/mL respectively. Along with the crude extract in distilled water, methanol extracts of peel, young

leaf, and mature leaf were also added in the plate in the same manner. Streptomycin sulphate and tetracycline (1mg/mL) were used as standard. Plates were incubated at 37°C for 24 hours.

2.6 Determination of minimum inhibitory concentration (MIC): The antimicrobial effect of the juice and different extracts was determined in terms of MIC by measuring the zone of inhibition (cm). The values were compared with the standards.

| Table1: MIC of different parts of Citrus limon in cm. | | | | | | | | | |
|---|-------------|-------------|----------|-----------|------------|-------------|--|--|--|
| Sl. No. | Extract | Crude juice | 0.1µL/mL | 0.01µL/mL | 0.001µL/mL | 0.0001µL/mL | | | |
| 1 | Fruit juice | 3.6 | 1.7 | 1.2 | х | Х | | | |
| 2 | Mature leaf | 1.3 | Х | Х | Х | Х | | | |
| 3 | Young leaf | 1.5 | Х | Х | Х | Х | | | |
| 4 | Peel powder | 1.4 | Х | Х | Х | Х | | | |

III. **Results And Discussion**

| 2 | Mature leaf | 1.3 | Х | Х | Х | Х | | | | |
|--------------------------------------|-------------|-----|---|---|---|---|--|--|--|--|
| 3 | Young leaf | 1.5 | Х | Х | Х | Х | | | | |
| 4 | Peel powder | 1.4 | Х | Х | Х | Х | | | | |
| Table2: MIC of standards used in cm. | | | | | | | | | | |

MIC (µg/mL) Sl. No. Standard 0.001 Tetracycline Streptomycin 0.010



Fig 1: MIC of different parts of Citrus limon in cm

Fig 2: Plate showing effect of juice on E. coli (Streptomycin at center)

The antimicrobial effect was tested against *E.coli* for the crude juice as well as for different methanol extracts of peel, leaf (mature and young). Among all the extracts tested crude juice at concentration in distilled water was found to be maximum. The MIC was recorded as $0.01 \mu L/mL(v/v)$ for the crude juice as shown in Table 1. The MIC for crude juice is found to be more than two standards used. However, on dilution the zone of inhibition for the crude juice was seen only upto the concentration of 0.01μ L/mL but for the standards, the MIC is very strong and is found to be $0.01(\mu g/mL)$ and $0.001(\mu g/mL)$ of streptomycin and tetracycline respectively as shown in Table 2. The MIC of different parts of Citrus limon (cm) against E.coli is represented in Fig 1 and Fig 2 represents effect of Juice extracts on E. coli . Streptomycin used as standard is shown at the center of plate

The antimicrobial activity of different methanol extracts was not much significant and after dilution of the crude methanol extracts inhibition zone was not recorded.

Conclusion IV.

The present study has proved the antibacterial property of lemon. Microorganisms can cause harmful diseases which may be life threatening. In place of synthetic drugs if lemon is used in diet it will be more beneficial since it is available in market easily and almost consumed by all class of people. The requirement is to explore the lemon effect at global level for the betterment of heath and economy of any state or country.

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