# Evaluation of Antimicrobial Activities of Medicinal Plant Extracts on Common Human Pathogenic Bacteria

**Renuka M. R**
Department of Zoology, N.S.S. Hindu College, Changanacherry, 686102, Kerala renujayan2000@gmail.com; 9446124601.
Corresponding author: Renuka M.R

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**Abstract** - Present study aimed at the analysis of sensitivity of *E. Coli* . to *Zingiber officinale*, *Azadirachta indica*, *Ocimum sanctum* and *Allium sativum.* The ethanolic extract of these four medicinal plants traditionally used in medicine, were studied for their antibacterial activity against the gram negative, *E.coli*. All the medicinal plant extracts showed inhibitory activity against *E. coli*. Among the four medicinal plants, *Zingiber officinale* showed maximum activity against the test microbe. Minimum activity was shown by and *Azadirachta indica* and *Ocimum sativum* the lowest inhibition was exerted by *Allium sativum*. The study comes to the conclusion that medicinal plants which are traditionally used in Ayurveda or in other herbal medical practices have scientific basics and can be modified to produce specific medicines against each bacterium.

**Key Words** : *Zingiber officinale, Azadirachta indica, Ocimum sanctum, Allium sativum E. Coli*

## I. INTRODUCTION

*Escherichia coli* also known as *E. coli* is a Gram-negative, facultative, anaerobic rod-shaped bacterium of the genus *Escherichia*, that is commonly found in the lower intestine of warm blooded organisms. Most *E.coli* strains are harmless, but some serotypes can cause serious food poisoning in their hosts, and are occasionally responsible for product recalls due to food contamination. An alarming increase in bacterial strains resistant to existing antimicrobial agents demands a renewed effort to seek agents effective against pathogenic bacteria resistant to current antimicrobials. Spices are some of the most commonly used natural antimicrobial agents in foods. Natural antimicrobial compounds in spices were found to possess antimicrobial activity. Present study aimed at the analysis of antimicrobial properties of four indigenous herbs on the bacteria *Escherichia coli.*

## II. MATERIAL AND METHODS

Before screening of antimicrobial activity all glassware's, prepared media and other materials required to use in the test are autoclaved at 121°C and 15 lbs pressure for 20 min.

**Preparation of plant extracts**

50 grams of garlic cloves, ginger rhizomes and Neem and Thulsi leaves were dried in shade and ground to powder. This powder was used in ethanolic extraction using a soxhlet evaporator for 48 h. The filtrates were dried to powder form by keeping in pre -sterilized oven at $60^0$C. 10 gm of the ethanol extract powder was further dissolved and diluted with 100 ml DMSO to get 10% solution. Similarly 10% solution was prepared for Ampicillin also. From these 10% solutions of plant extracts and ampicillin,1ml was taken to impregnate the filter paper discs.

**Medium used** - Mueller Hinton Agar, MHA (Himedia Laboratories Ltd., India) was used in the study. 3.8g of Muller Hinton Agar is added to 100 ml distilled water, mixed well and dissolved by heating with continuous stirring. Boiled for 1 to 2 minutes and autoclaved at 121° C for 15 minutes at 15 lbs. Cooled to 45° C and poured in sterile Petri plates up to a uniform thickness of approximately 4mm and the agar is allowed to set at ambient temperature.

**Preparation of inoculums** -The test microorganisms were maintained at 4°C on nutrient agar slants. Active cultures for *E. coli* was prepared by transferring a loopful of cells from the stock cultures to test tubes of 30 ml

nutrient broth. The inoculated tubes were incubated without agitation for 24 h at 37°. The cultures were diluted with 20 ml fresh nutrient broth before using it for plate culture.

**Screening of antimicrobial properties by Agar disc diffusion method**

This method (Kirby Bauer et al, 1966) is suitable for organism that grows rapidly over night at 35-37°C. A sterile cotton swab was inserted into the bacterial suspension and then rotated and compressed against the wall of the test tube so as to express the excess fluid. The surface of Muller Hinton Agar plate was inoculated with the swab. To ensure that the growth is uniform and confluent the swab is passed three times over the entire surface, by repeating the procedure, taking care the second and third time to turn the plate through 60°. 7 mm filter paper discs were impregnated with 1 ml of each of the different plant extracts. The discs were kept under refrigeration until ready to be used. Discs loaded with natural products as well as ampicillin were placed onto the surface of the agar. Discs were dispensed to the agar surface with a sterile forceps. After application, ensured that the disc has made complete contact with the agar surface by touching the top of the disc with forceps. Commercial antibiotic ampicillin and paper discs impregnated with 1ml of DMSO diluents used to dilute natural products were used as control. Tests were repeated for four times and the results were statistically analysed. Results are expressed as mean value ±SD of diameters of zone of inhibition for four replications.

## III RESULTS AND DISCUSSION

After incubating for 24 hours at $37^0$C, the bacteria produced uniform colonies on the surface of the Muller Hinton Agar plate. Following results were obtained from the antimicrobial property assay using disc diffusion method. Table 1 depicts the statistical analysis of different plant materials and ampicillin used for anti microbial property assay in the present study.

**Ampicilin** -Antibiotic Ampicillin produced IZDs of 18 mm, 18 mm, 16mm, 18 mm in four different treatments.

***Zingiber officinale*** - The analysis of the inhibitory activity of extracts from *Zingiber officinale* (ginger) in different treatments was 11mm, 11mm, 10mm and 10mm. The maximum inhibition zone produced is of 11mm diameter.

***Allium sativum*** - ***Allium sativum*** produced the lowest IZDs in the treatment. The four treatments of the plant extracts gave a maximum value of 7mm and a minimum value of 5mm for IZD.

***Ocimum sanctum*** - The average zone diameter is 7 mm with a standard deviation of 1.

***Azadirachta indica*** - *Neem* produced IZDs of 9.5 mm,8 mm, 9 mm and 7 mm. The average IZD is 8.38 mm with a standard deviation 1.10868.

All the medicinal plant extracts showed inhibitory activity against *E. coli*. Ampicillin produced zone of inhibition diameter up to 18 mm. Among the four medicinal plants, *Zingiber officinale* showed maximum activity against the test microbe. Zingiber obtaine maximum IZD of 11 mm. Minimum activity was shown by and *Azadirachta indica* and *Ocimum sativum* the lowest inhibition was exerted by *Allium sativum* in which the IZD was 6 mm. The inhibition zones were clearly visible on 24 hour incubation at $37^0$C. Significant difference between the inhibition zone diameters (IZDs) produced by different plant extracts were observed. This shows the difference in the antibacterial potency of these plants.

**Table 1 Statistical analysis of the significance of data obtained.**

| Treatment | Inhibition zone diameter (Mean±SD) | t value |
|---|---|---|
| Ampicilin | 17.5 ± 0.5 | |
| *Zingiber officinale* | 10.5 ± 0.5 | t= 12.12435, P= 1.9 |
| *Azadirachta indica* | 8.38± 1.1087 | t= 7.40656, P= 0.000311 |
| *Ocimum sanctum* | 7 ± 1 | t= 13.74772 , P= 0.00001 |
| *Allium sativum* | 6 ± 0.707 | t= 17.81572 , P= 0.00001 |

The indigenous medicinal system of India widely uses *Azadirachta indica* as a remedy for skin diseases. The mean IZD obtained from the treatment of the leaf extracts of *A. indica* on the tested bacteria was 9.5 mm. This indicates the strong efficiency of the leaf contents to resist the growth of bacteria.

Srinivasan, et. al., (2001) showed that about 22 Indian medicinal plants used on folkloric medicine were active against major gram negative bacteria. The results

obtained from the present study was also supporting to this .All the four medicinal plants tested, showed significant activity against the tested gram negative human pathogen , *E. coli*. All these results supported the traditional use of these medicines.

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## AUTHOR

Dr. M R Renuka, M.Sc., Ph.D.,
Associate Professor and Head, Post Graduate and Research Department of Zoology, N S S Hindu College, Changanacherry renujayan2000@gmail.com. , Ph. 9446124601