Comparison of Antibacterial Activities of Leaf extracts of *Eclipta alba* against *Klebsiella pneumoniae*

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Abstract

This study was focused on the comparison of biological activities of the leaf extracts of *Eclipta alba* L., against *Klebsiella pneumoniae* (MTCC-4032). The extracts were prepared from the leaves of the aforementioned plant species using Rotavapour R-300. The antibacterial activity of the extracted essential oils was evaluated against *Klebsiella pneumoniae* (MTCC-4032) using broth micro-dilution method recommended by Clinical Laboratory Standards Institute (CLSI). The Inhibition Concentration *i.e.* IC50 and Minimum Inhibition concentrations (MIC) using SpectramaxPlus384 Molecular Devices were recorded. The IC50 value of acetone leaf extract, ethanol leaf extract and petroleum ether extract were showed 0.020, 0.080 and 0.039 mg/ml respectively. The acetone leaf extract was found to be most effective with their MIC 0.020 mg/m1 whereas ethanol leaf extract was found to be least effective with their MIC 1.030 mg/ml against *K. pneumoniae*. Hence, leaf extracts of aforementioned plant exhibit great potential for the development of eco-friendly, non-toxic, cost effective, anti-bacterial formulations.

Key words: Antibacterial activity, *E. alba*, Broth Micro-dilution, etc.

Introduction:

Emerging and re-emerging infections and unfold of deadly, drug-resistant strains of organisms create a challenge to the worldwide public health for his or her treatment. Production of new antibiotics can be lowered down significantly due to a notable increase in resistant strains of bacteria [1]. Thus, the rummage around for novel antimicrobial agents is of the utmost importance within the current world [2]. Global attention has been shifted towards finding new chemicals, especially herbals, for the development of new drugs. These natural product will give distinctive parts of molecular diversity and biological practicality, that is indispensable for novel drug discovery [3]. Natural product or natural product-derived medicine comprise about 28% of all new chemical entities launched onto the market [4]. A large proportion of natural product in drug discovery has stemmed from the varied molecular structures and therefore the convoluted carbon skeletons of natural product [5]. Plants have tried to be vital natural

resources for medications; documentation of their use in medicine originates from history. Ethnobotanical and present plants give an upscale resource for natural drug analysis and development [6]. Medicinal plant primarily based drugs or medicine have the else advantage of being easy, effective and providing a broad spectrum of activity with larger stress on preventive action [7]. Autochthonic systems of medication and plant primarily based drugs might give each ideas of medical aid similarly as therapeutic agents to enrich trendy medicine, particularly in management of mode and communicable diseases. Medicinal plant product might conjointly prove helpful in minimizing the adverse effects of assorted therapy agents similarly as in prolonging longevity and attaining positive general health [8]. The worldwide interest within the medicinal potential of plants throughout the previous few decades is so quite logical.

Material and method

Extraction of essential oil - The plant materials of *E. alba* were collected from Phaphamau, Prayagraj in the month of September. Plants were identified at Department of Botany, University of Allahabad. Leaves were dried in shade. Dry leaves were crushed using a rotavapour (BUCHI Rotavapour R-300). Extracts were prepared accordingly. Extracts was stored at 4°C until analysis (9-11).

Preparation of Mueller-Hinton broth (MHB) – Take 1000 ml of DDW in a beaker. Add 21 gms of MHB powder. Shake well and boil up to 100 ºC. Close the mouth with cotton plug. Place the solution inside autoclave. After this, MHB is ready to use.

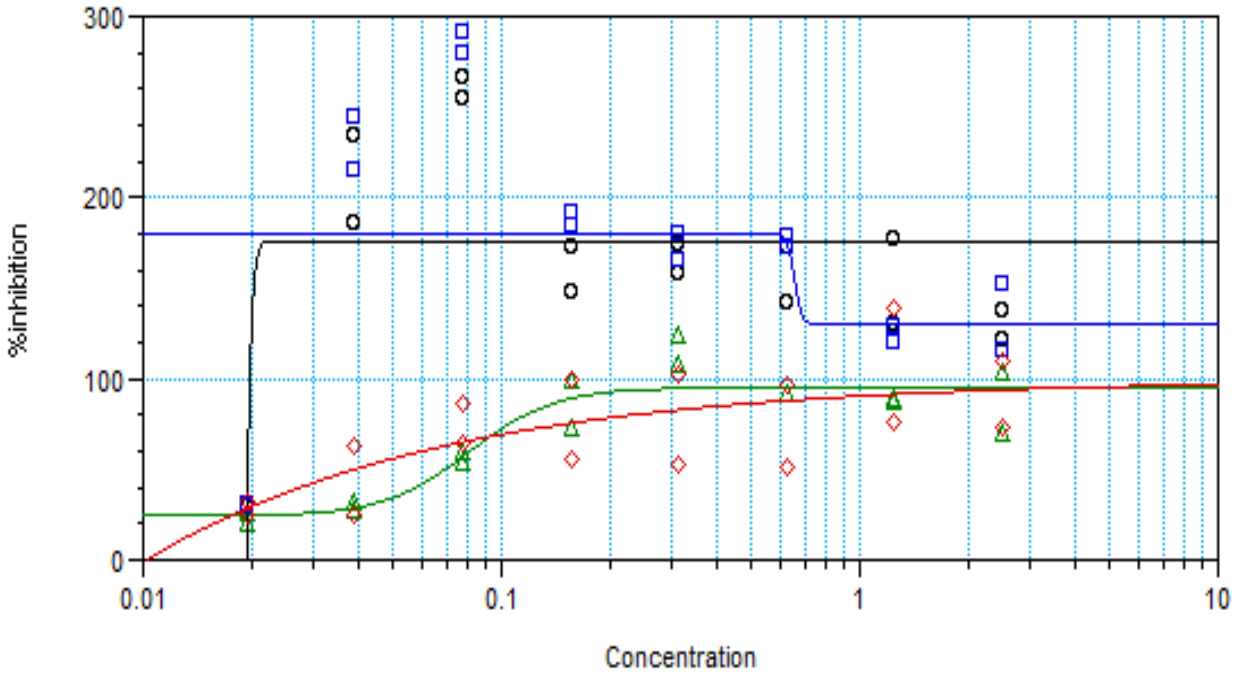
Preparation of inocula- Inocula of procured culture of pathogenic bacteria (48 hours old) was prepared comparing 0.5 McFarland Standard Solution by using Spectrophotometer at 480nm.

Antibacterial Screening- Leaf extracts were screened for antibacterial activity against *K. pneumoniae* Minimum Inhibitory Concentrations (MIC) were determined using Broth Micro-dilution method recommended by Clinical Laboratory Standard Institute (CLSI). 96 well plate was used for micro dilution. The Broth Micro-dilution protocol was used as recommended by CLSI- Antibacterial assay. Column-1 contains formaldehyde and is known as negative control. Column-2 contains MHB as broth control. Column-3 and Column-4 contains drug in each row. Row A and B of streptomycin. Row C and D contain acetone leaf extracts. Row E and F contain ethanol leaf extracts whereas row G and H contains petroleum ether extracts. Column-3 is known as column of drug control. Now dilute the drugs horizontally from column-4 to column-11 by using micropipette. Column-12 was filled with bacterial inoculum as positive control. The extract solutions over horizontally diluted 1:1 in MHB in a 96 well plates were incubated at 37 °C for 24 hours (9-11). Inhibitory concentration and it was determined as the lowest concentration without turbidity. Streptomycin used as Drug (Standard) control. Formaldehyde was used as a negative control.

Results:

The results were recorded in terms oil Inhibition Concentrations (IC50) and Minimum Inhibition Concentrations (MICs) via Spectramax Plus384, Molecular Devices Corporation, USA (graph 1). IC50

value of acetone extract, ethanol extract and petroleum ether extract were showed 0.020, 0.080 and 0.039 mg/ml respectively (table 1). The minimum inhibition concentrations (MIC) of acetone extract, ethanol extract and petroleum ether extract were recorded 0.020, 1.030 and 0.323 mg/ml respectively (table 2). Acetone leaf extract was found to be most effective with their MIC 0.020 mg/m1 whereas ethanol leaf extract was found to be least effective with their MIC 1.030 mg/ml against *K. pneumoniae*.



Graph 1. Graph obtained from Spectramax Plus384 for concentration Vs %inhibition.

S.N.	IC-50 Values
IC50-1	0.070
IC50-2	0.020
IC50-3	0.080
IC50-4	0.039

Table 1. IC50 values of extracts.

S.N.	MICs Values
MIC-1	0.117
MIC-2	0.020
MIC-3	1.030
MIC-4	0.232

Table 2. MICs of extracts.

Conclusion:

It has been concluded from the present study that all the three leaf extracts possess antibacterial activity against *K. pneumoniae*. Acetone leaf extract shows remarkable efficiency over ethanol and petroleum ether extracts against bacteria. Leaf extracts from the aforementioned plant exhibit great potential eco-friendly, non-toxic, cost-efficient and antibacterial herbal formulations.

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