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ANTIOXIDANT AND ANTIBACTERIAL ACTIVITY OF MURRAYA KOENIGII LEAVES

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ABSTRACT

Murraya koenigii (L.) Spreng, colloquially known as meethi neem, kari patta or curry leaves belongs to the Rutaceae. The plant is renowned in various traditional system of medicine for its peculiar aroma and therapeutic significance. The plants prosper well in tropical and subtropical climates *M. koenigii* is significantly used in Indian culinary and complementary system of medicine to mitigate various disorder. The ethanolic extract of leaves manifested fair antioxidant potential with IC_{50} of 17.83 compared to standard IC_{50} of 16.45. The ethanolic extract of leaves displayed fair potential of antibacterial against *S. aureus* at 2% concentration with the inhibition of 18 ±1.132 as compared to 1% with the inhibition of 12.33 ±0.653. the antibacterial activity of leaves extract against *E. coli.* has been progressively increased at 2% concentration with the zone of inhibition of 15 ±1.132 as compared to 1% with inhibition of 10.66 ±0.653, while compared both strain it was found that the extract had greater antibacterial against gram positive *S. aureus.*

KEYWORD:- *Murraya koenigii* (L.), leaves, Antioxidant, antibacterial, Hydroalcoholic.

INTRODUCTION

Plants are vital for human existence in the earth. Plants recognized as a preferential source of medicine and used to alleviate, recuperate and treat variety of ailments from ancient times. *Murraya koenigii* (L.) Spreng, colloquially known as meethi neem, kari patta or curry leaves belongs to the Rutaceae. The plant is renowned in various traditional system of medicine for its peculiar aroma and therapeutic significance. The plants prosper well in tropical and subtropical climates *M. koenigii* is significantly used in Indian culinary and complementary system of medicine to mitigate various disorder. Leaves, bark and roots are used in skin eruptions, stimulant, tonic, stomachic, dysentery, diarrhea, anthelmintic, inflammation, analgesic, curing piles, itching, leukoderma, vomiting and blood disorders. The vital constituents of plant are O-phellandrene, P-caryophyllene, P-elemene and P-gurjunene, α-pinene, β-phellandrene, β-caryophyllene carbazole alkaloids, coumarins, acridine, carbazole alkaloids, essential oil, carotenoids, saponins, proteins, steroids, tannins, flavonoids, phenylpropanoids. glycosides, phenolics, nicotinic acid, vitamin C, sesquiterpenes etc. The leaves of plant are green, lanceolate, exstipulate, bipinnately compound with reticulate venation. The plant has been asserted to possess array of therapeutic properties includes anti-oxidant, antimicrobial, antibacterial, immunomodulation, hepatoprotective, antidiabetic, wound healing, antipyretic, cytotoxic, antiulcer, cholesterol lowering, anti-obesity, neuroprotection and antitrichomonal effects.[1-6]

MATERIALS AND METHODS

Analytical grade reagents and chemicals has been used for the purpose of study all the chemicals were procured from Central Drug House (P) LTD. New Delhi, the glassware used in the study was borosilicate and ASGI mark. Pharmaspec Shimadzu UV-VIS Spectrophotometer model UV-1700, Japan has been used.

Collection and Processing of Plant Material

The leaves of *Murraya koenigii* have been collected in the month of march from Madhav University campus Rajasthan india. The collected plant leaves were thoroughly washed with tap water then shade dried till crumpled. The dried leaves were coarsely powdered, the powder is screened to obtain uniform size of particle, The powdered drug was extracted with selected solvent.

Extraction of plant material

The hydro alcoholic extract has been prepared by macerating the drug in 70% ethanol. 250g of coarsely powdered plant sample was steeped in 70% ethanol for seven consecutive days in closed container with intermittent stirring. The extract was collected and

filtered using whatman filter paper, the filtrate was evaporated and concentrated to remove excess solvent under reduced pressure at 35°C in rotary evaporator. The concentrated extract was then placed in the desiccators to expunge residual solvent.

***In-vitro* Anti-oxidant Activity**

Antioxidant efficacy of *Murraya koenigii* leaves extract has been evaluated by DPPH radical scavenging method. Ascorbic acid was used as standard antioxidant compound to compare sample. 0.1mM solution of 2,2-Diphenyl-1-picrylhydrazyl (DPPH) was prepared in methanol.[7-13]

Preparation of Sample/Standard

One mg of ascorbic acid and *Murraya koenigii* dried powdered extract were dissolved individually in 1ml of methanol to get 1mg/ml standard and sample stock solution. Dilutions were made to get the viable concentration of 20,40,60,80,100 µg/ml for both standard and sample in methanol. 2 ml of 0.1mM DPPH reagent was added and mixed thoroughly to each test tubes of sample and standard. The mixture is then incubated for 30 minutes in dark condition away from light then absorbance of standard and sample were recorded at the wavelength 517 nm.[7-13]

Preparation of control

Three milliliters of 0.1mM DPPH solution was prepared. The solution was incubated for 30 minutes at room temperature in dark condition. Absorbance of the control solution has been recorded against methanol as blank at 517 nm. The antioxidant activity of sample/ standard was reckoned by using formula.[7-13]

Percentage Inhibition = [(Abs of control- Abs of sample/ Abs of control x 100]

Antibacterial activity

Antimicrobial efficiency of the sample extract has been tested through well diffusion assay against gram positive bacteria *S. aureus* MTCC 10787 and gram-negative bacteria *E. coli* MTCC 42. The nutrient culture media was prepared by addition of twenty-eight-gram nutrient agar in one litre of distilled water. The media pH was checked after formulation and recorded for future reference. The media was sterilized through autoclave at 121^{o}C at 15 lbs pressure for 15 minutes, sterilized media was stand to cool and poured into plates before it gets solidified the process was carried out in laminar air flow.[11-16]

Well diffusion assay

The sample solution was prepared by mixing 1% and 2% of test extract discretely with distilled water. The culture of specific bacterial strain was spread on prepared media. Standard solution for comparison with test was prepared by dissolving one mg of ofloxacin and gentamycin in 1ml of distilled water to get 1mg/1ml of standard solution. The inoculum of *E. coli* MTCC42 and *S. aureus* MTCC 10787 were prepared, preliminary test organisms were inoculated in 10 mL of nutrient broth. The bacterial suspension was optimized to get 10^{8} CFU/ml. 100 µl of the inoculum was taken and transferred in to clear and sterile solidified agar media. Three wells of 6 mm were made by sterile cork-borer. The initial two wells were filled with test sample with concentration of 1% and 2% furthermore, third well were filled with 50µl of standard drug. The standard and sample were vault in sterile condition and allowed to diffuse for 30 minutes at room temperature. All samples were incubated for 24 hours at 37^{o}C. The incubated plates were inspected for effect of test sample and standard. The clearing zone observed around the well portend antimicrobial efficiency of tested compounds. The zone of inhibition was measured and calculated in mm with ruler to the back of the inverted petri plate.[11-16]

Table 1: DPPH radical scavenging activity of Ascorbic acid.

Concentration (µg/ml)	Percentage inhibition
20	58.985
40	67.124
60	75.792
80	83.298
100	88.900
Control	0
IC50	16.45

Table 2: DPPH radical scavenging activity of *Murraya koenigii*.

Concentration (µg/ml)	Percentage inhibition
20	55.073
40	64.482
60	70.190
80	76.744
100	80.443

Control	0
IC50	17.83

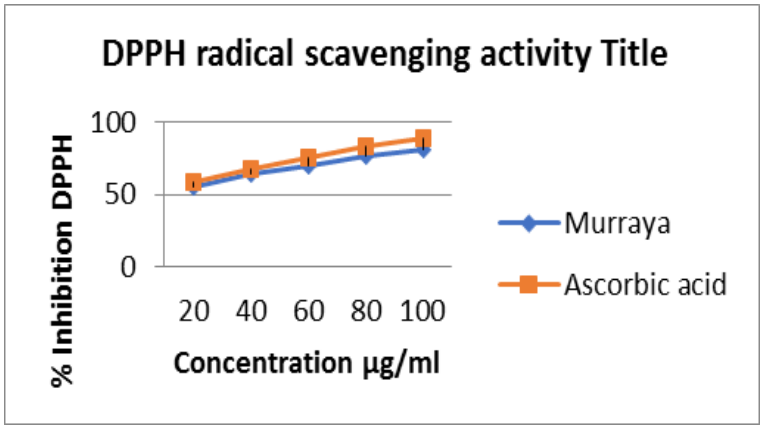

Fig. 1: Graph represents the percentage inhibition *vs* concentration of sample extracts.

Table 3: Antimicrobial activity of extract against *S. aureus.*

Extract	Plate 1	Plate 2	Plate 3	Mean±SD
1%	12 mm	13 mm	12 mm	12.33 ±0.653
2%	17 mm	18 mm	19 mm	18 ±1.132
Control	0mm	0mm	0mm	0±00
Ofloxacin (1mg/ml)	22 mm	21 mm	25 mm	22.66±2.356

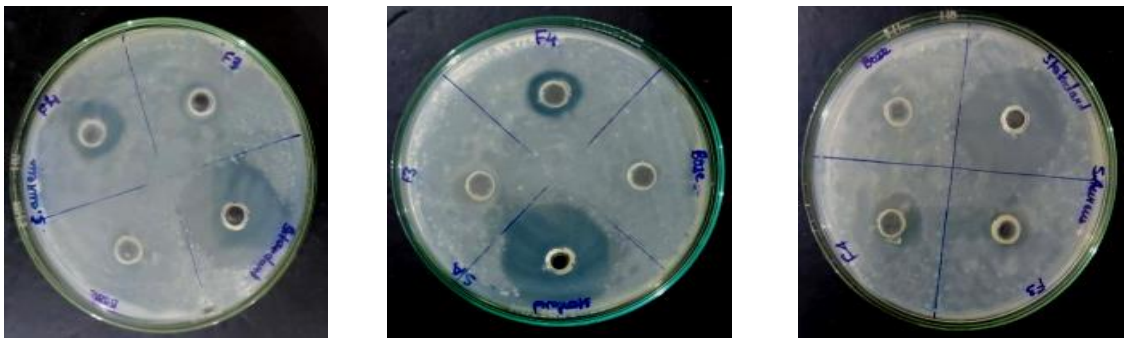

Fig. 2: Antimicrobial activity of extract against *S. aureus.*

Table 4: Antimicrobial activity of extract against *E.coli.*

Extract	Plate 1	Plate 2	Plate 3	Mean±SD
1%	11 mm	10 mm	11 mm	10.66 ±0.653
2%	14 mm	15 mm	16 mm	15 ±1.132
Control	0mm	0mm	0mm	0±00
Gentamycin (1mg/ml)	25 mm	26 mm	25 mm	25.33±0.653

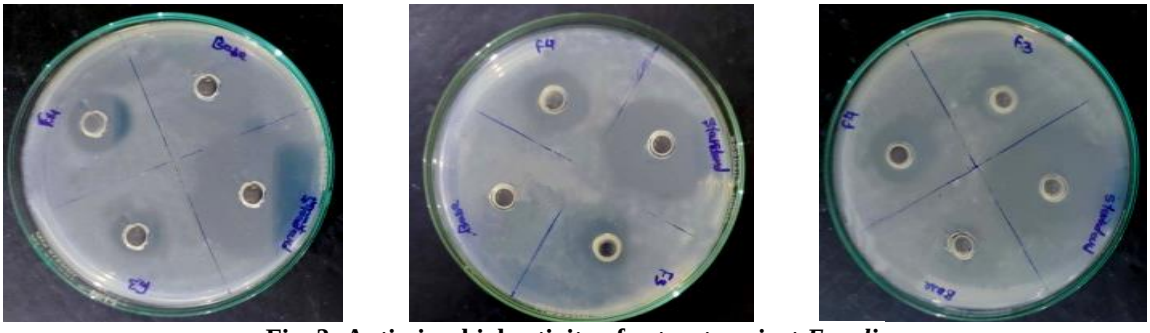

Fig. 3: Antimicrobial activity of extract against *E. coli.*

RESULT AND DISCUSSION

The DPPH radical scavenging potential of standard and extract has been compared to access the antioxidant potential of ethanolic extract the result indicated that the ethanolic extract of leaves had good antioxidant potential with IC_{50} of 17.83 compared to standard IC_{50} of 16.45. The antibacterial efficacy was tested against gram positive *S. aureus* and gram-negative *E. coli.* bacteria. The ethanolic extract of leaves displayed greater potential of antibacterial against *S. aureus* at 2% concentration with the inhibition of 18 ±1.132 as compared to 1% with the inhibition of 12.33 ±0.653. the antibacterial activity of leaves extract against *E. coli.* has been progressively increased at 2% concentration with the zone of inhibition of 15 ±1.132 as compared to 1% with inhibition of 10.66 ±0.653. while compared both strain it was found that the extract had greater antibacterial against gram positive *S. aureus.* The results supported that the plant had good antioxidant and antibacterial action that could be used for their rewarding effect in different formulations.

CONCLUSION

Increasing demand of herbal products inclined to explore the new potential substance from natural origin that could give better and safer alternative and efficiently. *Murraya koenigii* is a plant of therapeutic significance used to treat array of human ailments. The plant prospers well in tropical and subtropical climates. The antioxidant and antibacterial potential of *Murraya koenigii* leaves has been evaluated and. the results signified that the *Murraya koenigii* extract has promising outcome with fair antibacterial and antioxidant potential. Further study needed to refine the extract by using isolated components and some more pharmacological evaluation needed expand the realm of drug.

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