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Antioxidant and Antibacterial activity of *Canna indica* seeds

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ABSTRACT: *Canna indica* vernacularly known with numerous names such as Indian shot, Indian canna, African arrowroot, purple arrowroot, dev ked, bajarbattu etc. the plant thrives well in various tropical and sub-tropical atmosphere encompasses to family Cannaceae. The plant has been endowed with multitudinous chemical constituents. *Canna indica* demonstrates multifarious activity includes antioxidant, anti-inflammatory, hepatoprotective, antiviral, anti-diarrheal, anthelmintic, antibacterial, molluscicidal, cytotoxic, analgesic. hemostatic and immunomodulatory etc. The seeds of the plant are small, ovoid, globular, stiff and dense enough to sink in water. The seed of the plant would be used to treat variety of human condition. The ethanolic extract of seeds exhibited good antioxidant potential with IC_{50} of 18.80 compared to standard IC_{50} of 16.45. The antibacterial potential has been tested with gram positive *S. aureus* and gram-negative *E. coli.* The ethanolic extract herald a good antibacterial effect at 2% concentration as compared to 1%, the study also steered that the extract of seeds had greater antibacterial against gram negative *E. coli.*

KEYWORDS: *Canna indica*, seed, antioxidant, antibacterial, hydroalcoholic

INTRODUCTION

Canna indica profoundly known as numerous names such as Indian shot, Indian canna, African arrowroot, purple arrowroot, dev ked *etc.* the plant thrives well in various tropical and sub-tropical atmosphere encompasses to family Cannaceae. The plant has been bestowed with multitudinous chemical constituents like cardiac glycosides, alkaloids, flavonoids, steroids, terpenoids, carbohydrates, proteins, saponins, tannins and pigments. *Canna indica* demonstrates multifarious activity included as antioxidant, anti-inflammatory, hepatoprotective, antiviral, anti-diarrheal, anthelmintic, antibacterial, molluscicidal, cytotoxic, analgesic. hemostatic and immunomodulatory etc. The seeds of the plant are small, ovoid, globular, hard and dense enough to sink in water. Presence of vital constituent could offer a wide variety of protection against various skin ailments. The seed extract would be used in various skin disorders and restore the normal appearance and function. Due to presence of potential active metabolites, antioxidant and antibacterial activity of hydroalcoholic extract of seeds of *Canna indica* has been chosen for the study. Antioxidants are compounds which primarily counterbalance the deleterious effect of free radical in our body and protect the cell and tissue from inimical oxidative stress.[1-5]

MATERIALS AND METHODS

Analytical grade chemicals and reagents had 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 seeds of *Canna indica* have been collected in the month of September from the medicinal herbal garden, Madhav university campus, abu road, Pindwara India. The seeds were primarily washed with tap water then shade dried till complete drying. The dried seeds were used to made coarse powder. The powder is then shifted to get powder of uniform size, The powdered seeds was subjected to extraction with appropriate solvents

Extraction of Plant Material

The hydro alcoholic extract has been prepared by soaking the powdered sample in 70% ethanol. 250g of coarsely powdered plant sample was macerated with 70% ethanol for seven consecutive days in closed flask with occasional 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 potential of *Canna indica* seeds extract had been evaluated by DPPH radical scavenging method. The comparison of sample data was made with ascorbic acid as standard antioxidant compound. 0.1mM solution of 2,2-Diphenyl-1-picrylhydrazyl (DPPH) was prepared in methanol. [6-8]

Preparation of Sample/Standard

One mg of ascorbic acid and *Canna indica* 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.[6-9]

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.[6-10]

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.[11-15]

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°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 *Canna indica*

Concentration (µg/ml)	Percentage inhibition
20	54.228
40	63.847
60	69.556
80	78.646
100	80.126
Control	0
IC50	18.80

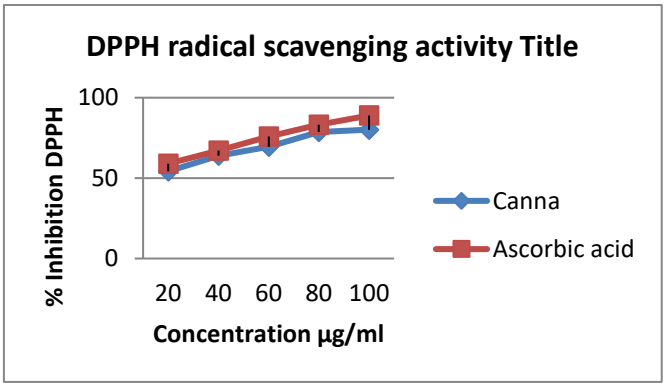

Fig.1 Graph represents the percentage inhibition *vs* concentration of sample extracts

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

Extract	Plate 1	Plate 2	Plate 3	Mean±SD
1%	6 mm	5 mm	6mm	5.66±0.533
2%	9 mm	11 mm	11 mm	10.33±1.307
Control	0mm	0mm	0mm	0±00
Ofloxacin (1mg/ml)	22 mm	21 mm	25 mm	22.66±2.356

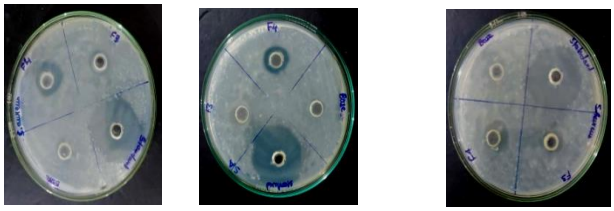

Fig.2 Antimicrobial activity of extract against *S. aureus*

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

Extract	Plate 1	Plate 2	Plate 3	Mean±SD
1%	12 mm	11 mm	9 mm	10.66±1.729
2%	14 mm	13 mm	15 mm	14.00±1.132
Control	0mm	0mm	0mm	0±00
Gentamycin (1mg/ml)	25 mm	26 mm	25 mm	25.33±0.653

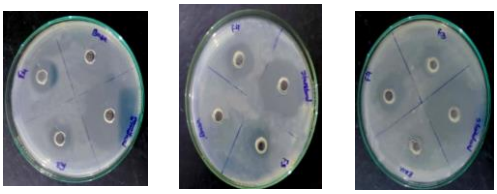

Fig.3 Antimicrobial activity of extract against *E. coli*

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 seeds had good antioxidant potential with IC_{50} of 18.80 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 seeds displayed greater potential of antibacterial against *S. aureus* at 2% concentration with the inhibition of 10.33±1.307 as compared to 1% with the inhibition of 5.66±0.533. the antibacterial activity of seed extract against *E. coli.* has been acclivitous at 2% concentration with the zone of inhibition of 14.00±1.132 as compared to 1% with inhibition of 10.66±1.729, while compared both strain it was found that the extract had greater antibacterial against gram negative *E. coli.* 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 used in various formulation. In this study we have tested the efficiency and potential of newer effects of *Canna indica* seeds extracts that might be useful to explore the plant in the newer area of herbal formulation. The results indicated that the *Canna indica* extracts had positive result on the parameter tested and could be used as anti-ageing, antibacterial and antioxidant potential. Further study needed to refine the extract by using isolated components and some more pharmacological evaluation needed that could broader the ambit of drug.

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