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INVITRO ANTI-OXIDANT ACTIVITY OF ALCOHOLIC EXTRACT OF THE OCIMUM SANCTUM

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ABSTRACT

India has a rich culture of medicinal herbs and spices, which includes about more than 2000 species and has a vast geographical area with high potential abilities for Ayurvedic, Unani, Siddha traditional medicines but only very few have been studied chemically and pharmacologically for their potential medicinal value.[1] In this present investigation In-vitro study of Aqueous Ethanolic extract of *Ocimum Sanctum* was carried out for the evaluation of Antioxidant activity by DPPH Assay method and found that the extract was acting as a powerful Antioxidant activity.

KEYWORDS: DPPH**,** Ocimum Sanctum, Ascorbic acid.

INTRODUCTION

Medicinal plants

Human beings have used plants for the treatment of diverse ailments for thousands of years.[2] Since the advent of modern allopathic medicine, the use of traditional medicine (inclusive of the use of medicinal plants for cure) declined to a considerable extent. However, in recent years, traditional medicine has made a comeback for a variety of reasons including side effects and toxicity of modern synthetic drugs, evolution of multi-drug resistance microorganisms, and the inability of modern medicine to find effective cures for a number of diseases. More than 70% of the developing world's population now depends on traditional medicinal system, otherwise known as complementary or

alternative systems of medicine.[3] It is a fact that plants used by indigenous peoples in their traditional medicinal systems are forming the sources of many important new pharmaceuticals.[4]

To cite a few instances of the influence of traditional medicines on the development of modern drugs and treatments, it has been reported that Native American traditional medicine provides a unique approach to the treatment of cardiovascular disease, which can complement modern medicine treatments.[5]

- Pharmacologically active chemicals present in plants (plants that have long been used in Traditional Chinese Medicine) e.g. artesunate, Hom harringtonine and cantharidin are now proving their potential for use in cancer therapy.[6]
- Herbal medicines are in great demand in both developed and developing countries as a source of primary health care owing to their attributes having wide biological and medicinal activities, high safety margins and lesser costs. Herbal molecules are safe and would overcome the resistance produced by the pathogens as they exist in a combined form or in a pooled form of more than one molecule in the protoplasm of the plant cell.[7]
- Traditional use of medicine is recognized as a way to learn about potential future medicines. Researchers have identified number of compounds used in mainstream medicine which were derived from "ethnomedical" plant sources.[8]
- Plants are used medicinally in different countries and are a source of many potent and powerful drugs.[9]

Plant Profile

Ocimum Sanctum is commonly known as **holy basil**, ***tulasi*** (***thulasi*** or ***tulsi)***, is an aromatic plant in the family *Lamiaceae* which is native to the Indian subcontinent and widespread as a cultivated plant throughout the Southeast Asian tropics.[25]

Tulasi is cultivated for religious and medicinal purposes, and for its essential oil. It is widely known across the Indian subcontinent as a medicinal plant and a herbal tea, commonly used in Ayurveda, and has an important role within the Vaishnava tradition of Hinduism, in which devotees perform worship involving holy basil plants or leaves. This plant is revered as an elixir of life.

The variety of *Ocimum tenuiflorum* used in Thai cuisine is referred to as "***Thai holy basil* (Thai: *kaphrao*)",** it is not to be confused with Thai basil, which is a variety of ***Ocimum basilicum***.[25]

MATERIALS AND METHODS

- The leaves of ***ocimum sanctum*** were collected and dried under shade, powdered and subjected to successive continuous maceration with aq.ethanol (37°C), the extracts thus obtained were concentrated by evaporation.
- Preliminary qualitative phytochemical analysis of the extracts of leaves.
- To evaluate antioxidant activity by DPPH Free radical Scavenging method.
- To Evaluate anti-oxidant activity of aq.ethanol extracts compared with standard values of ascorbic acid.

METHODOLOGY

***Ocimum sanctium* leaves**

(family: Lamiaceae)

↓

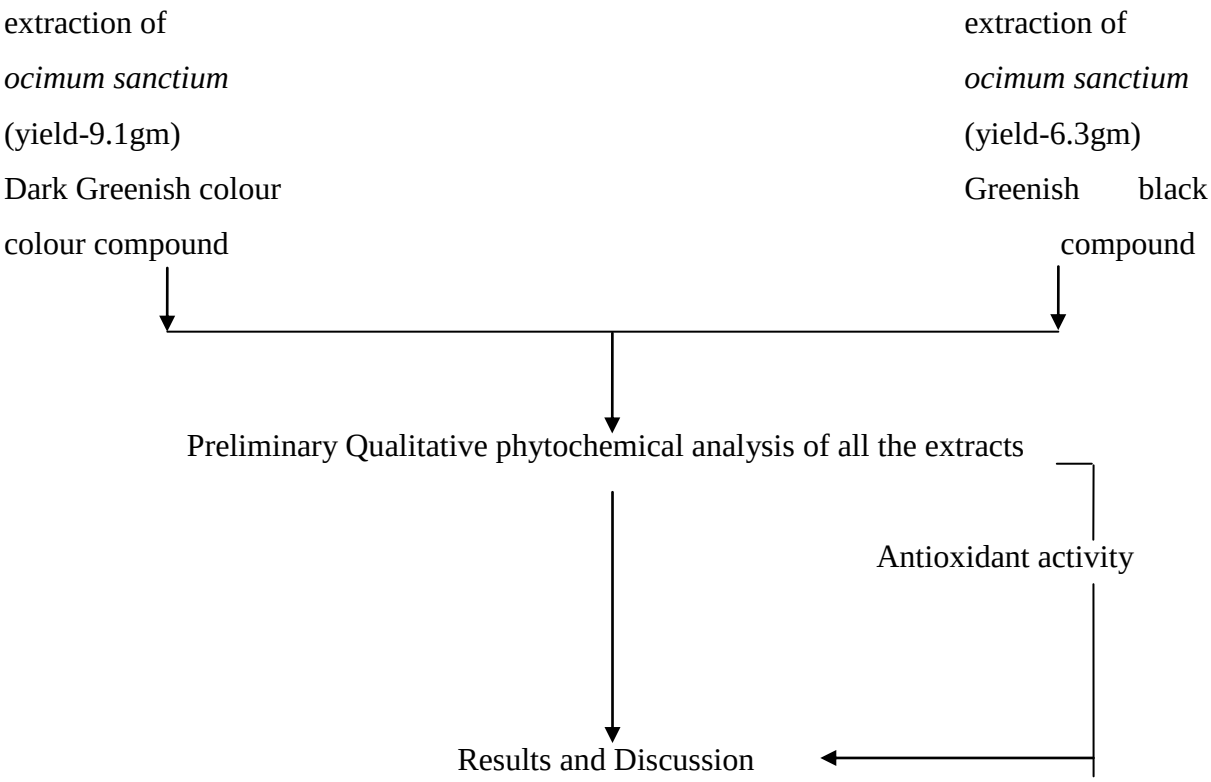
Around 0.5 kg

Continuous successive Maceration with

Different ratios of solvents (gradiant)

↓

Aq.ethanol(95%) Aq.ethanol(90%)



S. No.	Apparatus	Volume(mL)
1.	UV-Visible spectrophotometer (LabIndia^{R})	-
2.	Quartz cuvette (OPTIGLASS)	3.5
3	Electrical balance (INFRDADIGI^{TM})	-
4	Sonicator (CITIZEN)	-
5	Heating mantle (LAB TECH.CO)	-
6	Micro pipette	-
7	Heating water bath (SISCO)	-
8	Hot air oven (LAB TECH.CO)	-
9	Beaker	25,50,100
10	Volumetric flask	10,100
11	Test tube	10
12	Funnel	1.5-2mm
13	Glass rod	-
14	Spatula	-
15	Pipette	1,2,5,10
16	China dish	5,10,20
17	Thermometer	110^{o}C,360^{o}C
18	Measuring cylinder	10,25,50,100
19	Steam distillation apparatus	-
20	Whatmann filter paper	-
21	Tissue paper	-
22	Aluminium	-
23	Round bottom flask	250,500,1000
24	Labels	-
25	Dispensing balance	Upto100 gr
26	Melting point apparatus (LAB TECH.CO)	-

Note: Borosilicate glassware, stainless steel apparatus & other materials are imported from

"AMAN SCIENTIFIC CENTRE-VIJAYAWADA AND VIJAYA SCIENTIFIC CENTRE HYDERABAD"

Solvent used

Solvent	Boiling point (^{o}C)
Aq. Ethanol	72 – 73

Chemicals & Reagents

Laboratory grade (LR) Chemicals were used for routine work. Analytical grade (AR) reagents were used for analytical work.

Experimental Procedure

Anti –Oxidant Activity

DPPH radical scavenging activity

The ability of the plant extract to scavenge 1,1-diphenyl-2-picryhydrazyl (DPPH) free radicals was assessed by the standard method. The stock solution of extracts were prepared in methanol to achieve the concentration of 1 mg/ml. Dilutions were made to obtain concentrations of 250,500,750,1000,2500 µg/ml. Diluted solutions (1 ml each) were mixed with 2 ml of methanolic solution of DPPH (DPPH, 0.004%). After 30 min of incubation at room temperature the reduction of the DPPH free radical was measured by reading the absorbance at 517nm using UV-Visible Spectrophotometer. Initially, absorption of blank sample containing the same amount of methanol and DPPH solution was prepared and measured as control. Ascorbic acid was used as standard. The experiment was carried out in triplicate. Percentage inhibition was calculated using equation (1), whilst IC50 values were estimated from the % inhibition versus concentration plot, using a non-linear regression algorithm. The data were presented as mean values ± standard deviation (n = 3).

$$\% \text{ inhibition} = \frac{\text{(Absorbance of control- absorbance of sample)}}{\text{Absorbance of control}} \times 100 \quad \text{equation (1)}$$

RESULTS AND DISCUSSION

Results of DPPH free radical scavenging activity

The DPPH radical scavenging activity of Aq. ethanol extracts (95% & 90%) of ***Ocimum sanctum*** (AEPL) *leaves* were detected and compared with Ascorbic acid. The percentage inhibition (% inhibition) at various concentration (250-2500 µg/ml) of AEPL (95%) and

AEPL (90%) as well as standard Ascorbic acid (12.5 -100 µg/ml) were calculated and plotted in Figure-1 using Microsoft Office Excel 2007. The IC50 values are calculated from graph and were found Ascorbic acid.

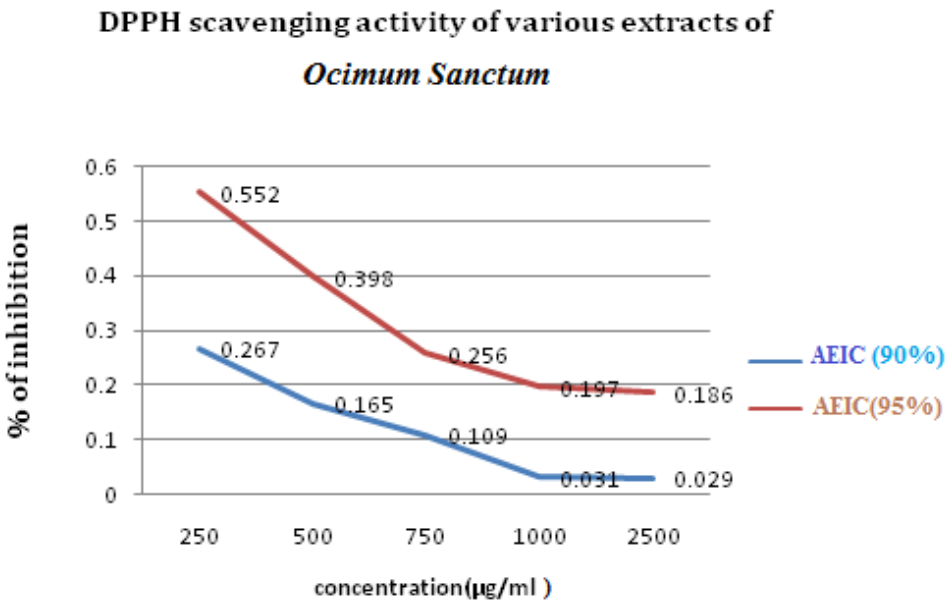


Figure-1

SUMMARY AND CONCLUSION

The shade dried leaves of ***Ocimum Sanctum*** were collected and dried under shade, powdered and subjected to successive continuous maceration with aq. ethanol (95% & 90%).

CONCLUSION

By performing the above work, it can be concluded that ***Ocimum Sanctum*** extracts possess anti-oxidant activity and the potency of anti-oxidant activity depends on the type of extract. The aq. ethanolic extract (95%) of ***Ocimum Sanctum*** leaves possess highest ***in vitro*** anti-oxidant activity compared to the aq. ethanolic extract (90%) extract.

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