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Research Article

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Evaluation of free radical scavenging potential of plant part extracts of medicinal plant- *Aglaia lawii*

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ABSTRACT

Phytochemical investigation of Aglaia lawii leaves showed presence of flavonoids, carbohydrates, phenolics, triterpenoids, tannins etc. An attempt made to perform free radical scavenging potential of A. lawii's different parts. Free radicals are well documented for playing role in our body. Excess production of free radicals leads to oxidative stress in body. It results in pathological manifestations as atherosclerosis, cancer, inflammatory condition, diabetes, alzheimer's disease Parkinson's disease etc. A variety of synthetic medicines employed in the treatment of different diseases that are capable to generate free radicals in body which may cause another disease. In Ayurveda, the plant sources are rich of antioxidants. Phyto-constituents are capable to terminate free radical reactions and prevent body from oxidative damage with less side effects and compatible to body physiology. Considering the above facts present work deals with evaluation of free radical scavenging potential of A. lawii plant. The extracts of leaves, bark, stem and seed were screened in- vitro, for their possible radical scavenging antioxidant activity by employing 2, 2-diphenyl-1- picryl-hydrazyl (DPPH) and nitric oxide (NO) reducing power with ascorbic acid as standard. The results revealed that ethanolic extract of leaves, bark, stem and seed can be a potential source of natural antioxidant, which can be applied as new source of Antioxidant Activity Index (AAI) which may complete demand of modern era to use such phytoconstituents or phytomedicines. The data obtained in these testing systems clearly established the antioxidant potency of the plant.

Key words: Antioxidants, *Aglaia lawii*, DPPH, NO, Ascorbic acid.

INTRODUCTION

There is an increasing interest in antioxidants, particularly in those intended to prevent the presumed deleterious effects of free radicals in the human body and to prevent the deterioration of fats and other constituents of foodstuffs. In both cases, there is a preference for antioxidants from natural rather than from synthetic source [1]. Natural products like fruits, vegetables and medicinal herbs are the richest sources of antioxidant compounds [2]. Natural antioxidants from edible medicinal plants, to replace synthetic antioxidants due to the long-term safety and negative consumer perception of synthetic antioxidants [3]. The natural antioxidants generally function as free radical scavengers and chain breakers, complexes of pro- oxidant metal ions and quenchers of singlet-oxygen formation [4].

There is therefore a parallel increase in the use of methods for estimating the efficiency of such substances as antioxidants [5, 6]. One such method that is currently popular is based upon the use of the stable free radical - diphenylpicrylhydrazyl DPPH. Oxidative stress causes serious cell damage leading to a variety of human diseases

like Alzheimer, Parkinson, atheroscleorosis, cancer, arthritis, immunological incompetence, neurodegenerative disorders etc. In these cases, there is a preference for antioxidants from natural rather than from synthetic sources [7,8]. Antioxidants are intimately involved in the prevention of cellular damage - the common pathway for cancer, aging and a variety of diseases. The body relies on obtaining its antioxidants from food and other supplements. Epidemiological studies and intervention trials on prevention of cancer and cardiovascular disease, antioxidant supplements are suggestive that dietary intake of antioxidants can help scavenge free radicals to protect the body against diseases [9]. In view of the immense medicinal importance for the antioxidants it is aimed to evaluate the antioxidant potential of plant extracts. There is an increasing interest in antioxidants, particularly in those intended to prevent the presumed deleterious effects of free radicals in the human body to prevent the deterioration of fats and other constituents of foodstuffs. Phytoconstituents are also important source of antioxidants and capable to terminate the free radical chain reactions [10].

Aglaia lawii is a woody big tree distributed in India, through Burma (Myanmar), Thailand, Indo-China and throughout Malaysia towards the Solomon Islands [11-13]. It is a traditional medicinal plant where, all parts of the plants having been used for the treatment of bacterial infection, liver, tumour diseases and headaches [14-15]. The pharmacological studies have shown that Aglaia species possess various notable biological activities such as anthelmintic, antimicrobial, analgesic, anti-inflammatory, immunimodulatory, antifungal etc [16**]**.

In this study attempts have been made to perform the antioxidant potency of arial parts of *Aglaia lawii* by scavenging activity using DPPH and NO. This work is carried out for the first time.

EXPERIMENTAL SECTION

Sample collection and Identification of plant materials

The plant material was collected from Mulshi district of Pune, Maharashtra, India. It was authenticated at Botanical survey of India, Pune, Maharashtra, India. Its Authentication No. is BSI/WRC/Tech/2010/1028, Pune, India.

Materials

1,1-diphenyl-2-picrylhydrazyl (DPPH), nitric oxide (NO), sulphanilamide, naphthyl ethylenediamine dihydrochloride were obtained from Sigma Chemicals Co., USA. All other chemicals and reagents were of analytical grade.

UV Spectrophotometer (UV-VIS1700Pharma Spectrophotometer Schimadzu) was used to measure the absorbance at various concentrations of the extracts under study.

Preparation of extracts:

Air shade dried, finely pulverized and exactly weighed plant materials were utilized to prepare extracts with measured volumes of solvent like methanol. The freshly prepared extracts were analysed to prevent any degradation. Solvents were removed under reduced pressure to get the crude mass of extracts. Weighed amounts of dried extracts were dissolved in known volume of methanol. Various aliquots of each extract were prepared for the DPPH and Nitric Oxide assays.

DPPH radical scavenging activity [17-20]:

DPPH is converted to 1, 1-diphenyl -2-picryl hydrazine when it reacts with antioxidants. A change in colour from purple to yellow is observed. Aliquots of extracts were made to total volume of 3ml using methanol. 0.15ml of freshly prepared DPPH solution (98 µg/ml) was added, stirred and left to stand at room temperature (27 0C) for 30 minutes in dark. The control contains only DPPH solution in methanol while methanol served as the blank (negative control).The reduction capability of DPPH radical was determined by the decrease in its absorbance. Absorbance was noted at 517nm by using UV-VIS spectrophotometer.

Nitric Oxide scavenging activity [20-22]:

In this spectrophotometric method the absorbance of chromophore formed during the diazotization of the nitrile with sulphanilamide and the subsequent coupling with naphthyethylenediamine dihydrochloride was measured. Sodium nitroprusside (SNP-5mM) in phosphate-buffer saline was mixed with an equivalent amount of methanol to get the control. Methanol served as blank. Methanol was added to test solutions at different concentrations to make

up a volume of 3ml and incubated at room temperature (27 0C) for 90 minutes. This incubated solution (1.5 ml) was added to 1.5 ml of Greiss Reagent. Absorbance at 546 nm was noted using UV–VIS spectrophotometer.

In both methods the capacity of scavenging free radicals was calculated as follows:

Scavenging activity (%) = {(Control Abs.—Sample Abs.)/Control Abs} × 100

Each experiment was carried out in triplicates and results were recorded as mean % antiradical activity ± SD.

IC 50 values were calculated from the plotted graph of scavenging activity against the concentrations of the samples. IC 50 is defined as the total antioxidant necessary to decrease the initial DPPH radical by 50%. IC 50 was calculated for all the extracts based on the percentage of DPPH radicals scavenged. Ascorbic acid was used as the reference compound (positive control) with concentrations 50 to 500 µg/ml for both the above spectroscopic methods.

RESULTS AND DISCUSSION

Freshly prepared extracts of the dried plant materials were subjected to screen for their possible antioxidant activities. DPPH free radical scavenging activity and Nitric Oxide scavenging methods using UV- VIS spectrophotometer were employed. DPPH radical scavenging test is based on the exchange of hydrogen atoms between the antioxidant and the stable DPPH free radical. It is a stable free radical at room temperature which, accepts an electron or hydrogen radical to form a stable diamagnetic molecule. This radical is reduced to the corresponding hydrazine, a colour change of the solution from violet to yellow is observed and that is monitored spectrophotometrically. More reduction of DPPH radical is related to the high scavenging activity of the particular extract [23].

The reduction capability of DPPH radicals was determined by the decrease in its absorbance at 517 nm, which is induced by antioxidants. The significant decrease in the concentration of the DPPH radical is due to the scavenging ability of *A. lawii.*

It can be seen that the radical scavenging activity for bark and leaves is found to be more significant than stem and seed extracts. The experiment established the following order: **bark > leaves > seed > stem.** The minimum and maximum values observed for stem and bark are 2.017 & 0.903 mg respectively where ascorbic acid is employed as a positive control. The findings are recorded. (**Fig. 1** & **2**, **Table 1**).The results demonstrate that the free radical scavenging activity of leaves and bark is nearly the same and it is 2 times more than stem and 1.5 times than seeds.

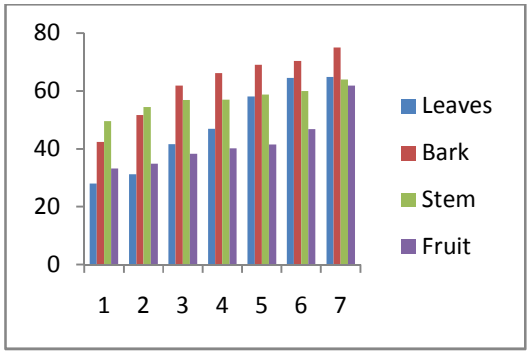


Fig 1 DPPH assay of plant parts

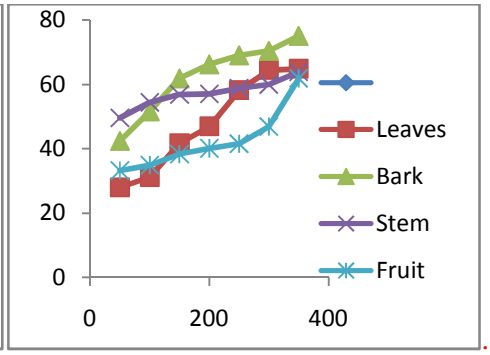
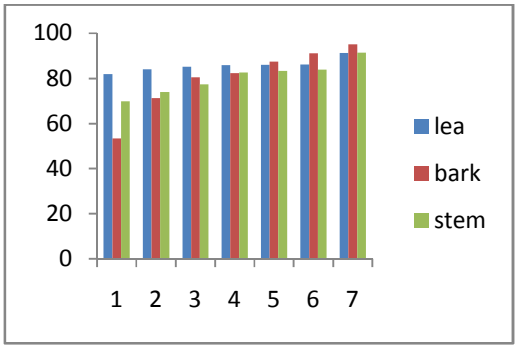


Fig 2 DPPH assay of plant parts



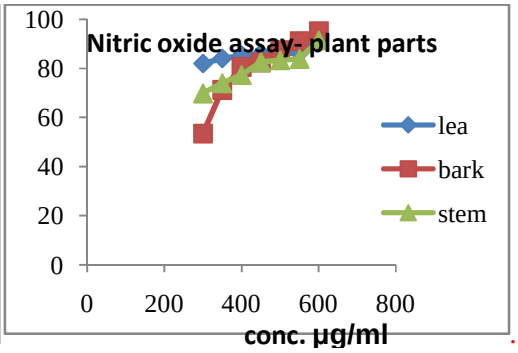


Fig 3 NO assay of plant parts **Fig 4 NO assay of plant parts**

NO is a molecular mediator of many physiological processes, Nitric oxide scavenging activity for ethanol extract of plant parts of *A. lawii* leaves with ascorbic acid as a standard are recorded **(Fig 3** & **4)**. IC_{50} values for the plant parts are mentioned. **(Table 1).** It can be seen that the radical scavenging activity for bark and stem is found to be more significant than leaves extracts. The order of activity is acknowledged: **bark > stem > leaves.** The minimum and maximum values observed for bark and leaves 0.611 & 1.048 mg respectively where ascorbic acid is used as a control. The results demonstrate that the free radical scavenging activity for leaves is 1.7 times and for stem 1.2 times less than bark.

Table 1: IC_{50} Values of plant parts

Plant parts	DPPH assay	NO assay
Leaves	0.915	1.048
Stem	2.017	0.754
Bark	0.903	0.611
Seed	1.462	-

Statistical analysis

Results are expressed as the standard error mean of three independent experiments. Student's *t* test was used for statistical analysis; P values < 0.05 were considered to be significant.

CONCLUSION

Comparative assay for the plant parts shows that the bark and leaves exhibit higher radical scavenging activity than seed and stem by DPPH assay. All plant part extracts exhibit higher range of the radical scavenging activity. It means these extracts are rich in flavonoids as well as phenolic compounds which, along with other polyphenolics in the plant material may be responsible for the antioxidant activities of these extracts.

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