

International
Research Journal of
**BIOLOGICAL
SCIENCES**



Volume 01 | Issue 01 | 2019



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***In vitro* Antioxidant Activity of Leaf and Bark Extracts of *Barringtonia acutangula* Linn.**

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ARTICLE INFORMATION

Received: November 27, 2018

Accepted: December 19, 2018

Published: January 31, 2019

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ABSTRACT

Barringtonia acutangula Linn. (Family *Lecythydaceae*) is a traditional medicinal plant used to cure many diseases in India. *Barringtonia acutangula* Linn. popularly known as *Samudraphal* in Hindi, *Kadambu* in Tamil and Indian Oak in English. The aim of the current study was evaluate the *in vitro* antioxidant activity of ethanolic leaf and bark extracts of *Barringtonia acutangula*. The antioxidant activity of plant material was assayed by DPPH radical scavenging assay and data was statistically analyzed using student "t" test. Results showed that IC₅₀ value of bark extract and leaf extract were 48.10±4.50 µg mL⁻¹ and 38.16±2.59 µg mL⁻¹, respectively; comparable with IC₅₀ value of standard (Vitamin C) 26.84±4.04 µg mL⁻¹. Therefore, it was concluded that *Barringtonia acutangula* improving the human health by participating in the antioxidant defence system against free radical generation.

Key words: *Barringtonia acutangula*, antioxidant, DPPH, IC₅₀ value, free radical

INTRODUCTION

Free radicals arising from metabolism or environmental sources interact continuously in biological systems and their uncontrolled generation correlates directly with molecular level of many diseases¹. Lots of research has clearly showed that free radicals would damage nearby structures including DNA, proteins or lipids. Radical scavenging antioxidants are particularly important in antioxidative-defence in protecting cells from the injury of free-radical². It is known that free radicals are the major cause of various chronic and degenerative diseases, such as coronary heart disease, inflammation, stroke, diabetes mellitus and cancer³. Therefore, it was important to assess antioxidant activity of the plants used in the herbal medicine either to elucidate the mechanism of their pharmacological action or to provide information on antioxidant activity of these herbal plants⁴. *Barringtonia acutangula* Linn. (Family *Lecythydaceae*) is an important medicinal plant of India. It is one of the useful traditional medicinal plant used for the cure and treatment of many ailments like hemolytic disease, joint disorder, spleen disorder, skin disease, diarrhoea, inflammation, syphilis, malarial and diabetes⁵. In the present study evaluated the antioxidant properties of the leaf and bark of *B. acutangula*. The ethanolic extracts of both parts of *B. acutangula* were used to investigate the antioxidant activity in terms of free radical scavenging activity (DPPH).

MATERIALS AND METHODS

Chemicals: DPPH (1,1-diphenyl-2-picrylhydrazyl) and L-ascorbic acid were purchased from Sisco Research Laboratories Pvt. Ltd., India. All other chemicals and solvents used were of analytical grade available commercially.

Collection and identification of plant material: The plant material was collected from Madurai, Tamil Nadu, India and authenticated by Dr. John Britto, Rapinat Herbarium, St. Joseph's College, Tiruchirappalli, Tamil Nadu.

Preparation of plant extracts: Fresh plant material was washed under running tap water, air dried and powdered. About 100 g of coarsely powdered plant materials (100 g/500 mL) were extracted in a soxhlet extractor for 8 h, with ethanol. The extracts obtained were then concentrated and finally dried to a constant weight. Dried extracts were kept at 4°C until further test were carried out.

Preparation of sample: The different concentrations of leaves and bark extract (20, 40, 60 and 80 µg mL⁻¹) were prepared for *in vitro* antioxidant activity. L-Ascorbic acid (Vitamin C) was used as the standard.

DPPH radical-scavenging activity: DPPH radical-scavenging activity was determined by the method of Shimada *et al.*⁶. A 2 mL aliquot of DPPH solution (25 µg mL⁻¹) was added to 0.5 mL sample solution at different concentrations (20, 40, 60 and 80 µg mL⁻¹). The mixture was shaken vigorously and allowed to stand at room temperature in the dark for 30 min. Then the absorbance was measured at 517 nm in a spectrophotometer.

$$\text{Radical scavenging activity (\%)} = 100 - \left(\frac{A_c - A_s}{A_c} \right) \times 100 \quad (1)$$

where, A_c = Absorbance of the control and A_s = Absorbance of the reaction mixture (in the presence of sample).

Statistical analysis: The results were presented as Mean ± SD. Data was statistically analyzed using student "t" test. For the calculation of IC₅₀, Linear regression analysis was done using Graph Pad prism statistical software.

RESULTS AND DISCUSSION

Free radicals produced by radiation, chemical reactions and several redox reactions of various compounds may contribute to protein oxidation, DNA damage, lipid peroxidation in living

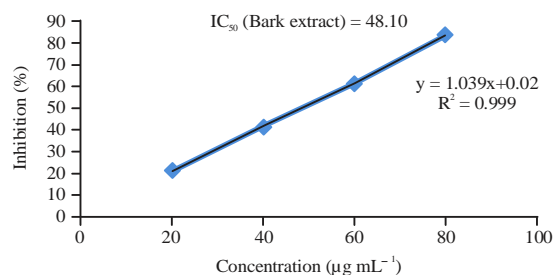


Fig. 1: DPPH Radical scavenging activity of *Barringtonia acutangula* bark extract at different concentrations

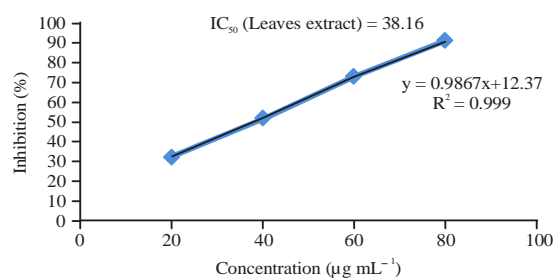


Fig. 2: DPPH Radical scavenging activity of *Barringtonia acutangula* leaves extract at different concentrations

tissues and cells⁷. This oxidative stress may be related to many disorders, such as cancer, atherosclerosis, diabetes and liver cirrhosis⁸. Free radical scavenging is one of the known mechanisms by which antioxidants inhibit lipid oxidation. DPPH, a stable free radical with a characteristic absorption at 517 nm, was used to study the radical scavenging effects of extract. The extract contained antioxidant components, which could react rapidly with DPPH radicals and reduce most DPPH radicals. This result reveals that the extracts are a free radical inhibitor or scavenger, acting possibly as primary antioxidants. Various extracts might react with free radicals, particularly the peroxy radicals, which are the major propagators of the autoxidation chain of fat, thereby terminating the chain reaction⁹⁻¹¹. Antioxidant activity of natural antioxidants has been shown to be involved in termination of free radical reaction¹². The results indicated that ethanolic extract of *B. acutangula* have a noticeable effect on scavenging free radical. The IC₅₀ of bark extract, leaves extract and standard were 48.10 ± 4.50, 38.16 ± 2.59 and 26.84 ± 4.04 µg mL⁻¹, respectively. The antioxidant activity and IC₅₀ values of bark, leaves extract and standard are shown in Fig. 1-3. The combined DPPH radical scavenging activity of *B. acutangula* leaf and bark extracts at different concentrations shown in Fig. 4. The inhibition concentration at 50% inhibition (IC₅₀) was the parameter used to compare the radical scavenging

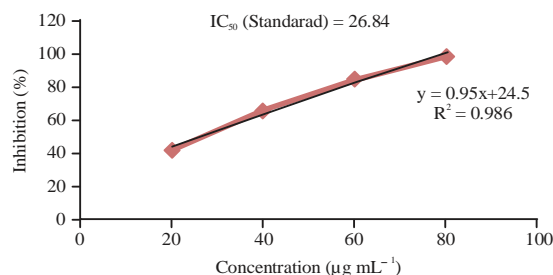


Fig. 3: DPPH Radical scavenging activity of Standard (Vitamin C) at different concentrations

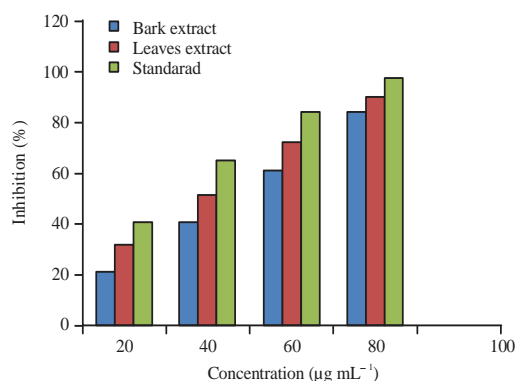


Fig. 4: Comparative study of DPPH radical scavenging activity of bark and leaves extract

activity. A lower IC_{50} meant better radical scavenging activity. DPPH \cdot is one of the free radicals widely used for testing preliminary radical scavenging activity of the plant extract¹³. Scavenging of DPPH \cdot radical is related to the inhibition of lipid peroxidation¹⁴. DPPH \cdot is usually used as a substance to evaluate the antioxidant activity¹⁵. Antioxidants either transfer an electron or a hydrogen atom to DPPH \cdot , thus neutralizing its free radical character¹⁶. DPPH \cdot test, which is based on the ability of DPPH \cdot , a stable free radical, to decolorize in the presence of antioxidants, is a direct and reliable method for determining radical scavenging action¹⁷. The DPPH \cdot assay has been largely used as a quick, reliable and reproducible parameter to search the *in vitro* general antioxidant activity of pure compounds as well as plant extracts¹⁸. The reducing capacity of compounds could serve as indicator of potential antioxidant property¹⁹. In the present study, the percentage of scavenging effect on the DPPH \cdot radical was concomitantly increased with the increase in the concentration of both leaf and bark ethanolic extracts from 20 to 80 $\mu\text{g mL}^{-1}$. The percentage of inhibition was existing from 31.89 ± 0.64 at 20 $\mu\text{g mL}^{-1}$ to 90.6 ± 4.12 at 80 $\mu\text{g mL}^{-1}$ for leaf extract and for bark extract, they were 21.35 ± 1.18 at 20 $\mu\text{g mL}^{-1}$ and 83.89 ± 6.34 at 80 $\mu\text{g mL}^{-1}$. From the results it is known that

B. acutangula extracts scavenging free radicals. Furthermore, it was noticed that the leaf extract has more pronounced scavenging activity than that of the bark.

CONCLUSION

The findings of the present study suggested that *B. acutangula* could be a potential source of natural antioxidant that would have great importance as therapeutic agents in preventing or slowing the progress of reactive oxygen species and associated oxidative stress related degenerative diseases.

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