RESEARCH ARTICLE

In vitro free radical scavenging activity of leaf extracts from *Mimusops elengi*

Akash Bardhan[1], Rishov Mukhopadhyay[1], Sanjib Bhattacharya[2*], Moulisha Biswas[1]

[1]Bengal Institute of Pharmaceutical Sciences, Kalyani, Nadia 741235, West Bengal, India
[2]Edward Food Research and Analysis Centre Ltd., Kolkata 7000121, West Bengal, India

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ABSTRACT

Mimusops elengi L. (Sapotaceae), commonly known as *Bakul* in Bengali and Sanskrit is a medium-sized evergreen tree Indigenous to tropical regions including India and found throughout the Indian subcontinent. The present investigation assessed the different solvent extracts of *M. elengi* leaf for their *in vitro* free radical scavenging potential by 1,1-diphenyl-2-picryl-hydrazyl (DPPH) radical scavenging assay. All of the extracts exhibited potent *in vitro* free radical scavenging activity that increased with extract concentrations. The methanol extract was found to be the most potent in this regard, followed by the ethyl acetate and benzene extracts respectively. Therefore, the present study confirms marked *in vitro* free radical scavenging activity of *Mimusops elengi* leaf.

Keywords: Free radical scavenging, leaf, antioxidant, *Mimusops elengi*.

INTRODUCTION

Antioxidants protect living organisms from damage caused by uncontrolled production of reactive oxygen species (ROS) and the concomitant lipid peroxidation, protein damage and DNA strand breaking. Current interest is focused on the potential role of antioxidants and antioxidant enzymes in the treatment and prevention of atherosclerosis, heart failure, neurodegenerative disorders, aging, cancer, diabetes mellitus and several others diseases. [1]

Antioxidants are added to a variety of foods to prevent free radical induced lipid peroxidation, which is responsible for the development of off-flavors and the undesirable chemical compounds in food. [2] These ROS cause destructive and irreversible damage to the components of a cell, such as lipids, proteins DNA and other macromolecules. Although normal cells possess antioxidant defense systems against ROS in the cells induces diseases such as cancer and aging. [3]

ROS are formed and degraded by all aerobic organisms. ROS can readily react with most biomolecules including proteins, lipids, lipoproteins and DNA. Exogenous chemical and endogenous metabolic processes in the human body or in the food system might produce highly reactive oxygen species, which are capable of oxidizing biological molecules, resulting in tissue damage and cell death. When the mechanism of antioxidant protection becomes unbalanced by exogenous and endogenous factors, it results in inflammation, diabetes, genotoxicity, cancer and accelerating aging. [4]

Antioxidant supplements or foods containing antioxidants may be used to help the human body reduce oxidative damage. The most commonly used antioxidants are BHA, BHT, propyl gallate and *tert*-butyl-hydroquinone. [5] However, they have been suspected of being responsible for liver damage and carcinogenesis in laboratory animals. Therefore, the development and use of more effective antioxidants is desired.

Traditional medicine worldwide is being reevaluated by extensive research on different plant species and their therapeutic principles. Plants produce antioxidants to control the oxidative stress caused by sunbeams and oxygen, they can represent a source of new compounds with antioxidant activity.

Mimusops elengi L. (Sapotaceae), commonly known as *Bakul* in Sanskrit and Bengali is a medium-sized

Address for correspondence

Dr. Sanjib Bhattacharya
Drugs and Pharmaceutical Laboratory, Edward Food Research and Analysis Centre Ltd., Kolkata 7000121, West Bengal, India.
E-mail: sakkwai@yahoo.com

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evergreen tree indigenous to tropical regions and found in tropical forests in South Asia, Southeast Asia and Northern Australia. It is grown throughout the Indian subcontinent. Its English common names include Spanish cherry, medlar, and bullet wood. Traditionally, all different part of this plant, namely leaf, root, fruit, seed, bark and flower are used to cure various kinds of disorders. [6] However, reports on the experimental studies on this plant especially on its leaf part are comparatively scanty. In the present study, we have aimed to evaluate *in vitro* free radical scavenging activity of different solvent extracts from *M. elengi* leaf against 1, 1-diphenyl-2-picryl-hydrazil radicals to assess the antioxidant effect.

MATERIALS AND METHODS

Plant material

The mature leaves of *Mimusops elengi* L. (Sapotaceae), were collected during the month of October 2013 from Sodepur, North 24 Parghanas, West Bengal, India. The plant species was taxonomically identified at the Central National Herbarium, Botanical Survey of India, Howrah, West Bengal, India. The voucher specimen [CNH/17A/2014/Tech. II/76] was maintained in our research laboratory for future reference. Just after collection the fresh leaves were cleaned thoroughly with running tap water and shade-dried at room temperature (24-26°C) with occasional shifting and then powdered with mechanical grinder, passing through sieve no. 40, and stored in an air-tight container for use in the study.

Preparation of plant extracts

The dried powdered material was successively extracted with benzene, ethyl acetate and methanol for 72 h at room temperature (24-26°C) in a cone shaped percolator. Then the solvent was distilled off in reduced pressure and resulting semisolid mass was vacuum dried using rotary vacuum evaporator to yield the solid extracts and stored in refrigerator for use in the present study. Preliminary phytochemical studies were performed on the three extracts. [7]

Chemicals

L ascorbic acid (vitamin C) and 1,1-diphenyl-2-picryl-hydrazyl (DPPH) from Sigma Chemical Co. Ltd. (St. Louis, MO, USA). All the other chemicals and reagents were of analytical grade obtained commercially form Merck.

Free radical scavenging activity assessed by 1, 1-diphenyl-2-picryl-hydrazil

The free radical scavenging activity of all of three extracts were measured by 1,1-diphenyl-2-picryl-hydrazil (DPPH) using the reported method. [8] Briefly, an 0.1 mM solution of DPPH in methanol was prepared, and 1 ml of this solution was added to 3 ml of the all of the extracts solution respectively in benzene, ethyl acetete and methanol at different concentrations (2, 4, 6, 8, 10, 15 µg/ml). The mixture were shaken vigorously and allowed to stand at room temperature for 30 min. Then the absorbance was measured at 517 nm using a UV-Vis spectrophotometer (Genesys 10 UV: Thermo Electron Corporation). Lower absorbance values of reaction mixture indicate higher free radical scavenging activity. Ascorbic acid at same concentrations was used as reference agent. The capability to scavenge the DPPH radical was calculated by using the following equation:

DPPH scavenging effect (%) = $[(A_0 - A_1) / A_0) \times 100]$

Where, A_0 is the absorbance of the control reaction, and A_1 is the absorbance of presence of all of the extract samples or reference. The results are stated in Table 1.

RESULTS AND DISCUSSION

Preliminary phytochemical studies indicated the presence of triterpenoids and steroids in benzene extract; triterpenoids, steroids, glycosides and alkaloids in ethyl acetate extract; carbohydrates and polyphenols in methanol extract.

The stable DPPH radical model is a widely used, relatively quick and precise method for the evaluation of free radical scavenging activity. The effects of antioxidants on DPPH radical scavenging is thought to be due to their hydrogen-donating ability. DPPH is a

stable free radical that accepts an electron or hydrogen radical to become a stable diamagnetic molecule. The absorption maximum of a stable DPPH radical in methanol was 517 nm. The decrease in absorbance of DPPH radical caused by antioxidants, because of the reaction between antioxidant molecules and radical progressed, results in the scavenging of the radicals by hydrogen donation. It is visually noticeable that as a change in color from purple to yellow. Hence, DPPH is usually used as a substrate to evaluate the antioxidant activity of different antioxidants. [9, 10] It has been reported that oxidative stress, which occurs when free radical formation exceeds the body's ability to protect itself, forms the biological basis of several chronic disease conditions such as arteriosclerosis. [11]

Based on the data obtained from the present study, all the test extracts were effective free radical inhibitor or scavenger, which reacts with free radicals, which may limit free radical damage occurring in the human body. The results are summarized in the Table 1. Free radical scavenging activity *in vitro* also increased with increasing concentration in the range of 2-15 µg/ml indicating concentration dependence. The methanol extract was found to demonstrate the most active free radical scavenging potential at the test concentrations, followed by the ethyl acetate and benzene extracts respectively. The presence of polyphenolic compounds only in the methanol extract may be responsible for its maximum antioxidative potential, as polyphenols are well known as natural antioxidants. [12, 13] The present preliminary study therefore confirms remarkable *in vitro* free radical scavenging activity of *Mimusops elengi* leaf against DPPH.

Table 1: DPPH scavenging effect of the different extracts of *Mimusops elengi* leaf.

Extracts	Concentrations (µg/ml)	% of DPPH scavenging activity respectively	Mean ± SEM
Benzene extract of *M. elengi*	2, 4, 6, 8, 10, 15	29.69, 37.27, 54.63, 63.45, 81.63	53.34 ± 4.27
Ethyl acetate extract of *M. elengi*	2, 4, 6, 8, 10, 15	33.72, 54.29, 63.72, 72.81, 87.54	62.42 ± 4.54
Methanol extract of *M. elengi*	2, 4, 6, 8, 10, 15	42.72, 72.27, 81.81, 87.54, 90.45	74.96 ± 5.18
Ascorbic acid	2, 4, 6, 8, 10, 15	29.70, 72.50, 87.54, 92.20, 99.23	76.23 ± 5.27

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