
Phytochemical Analysis and Antioxidant Activities of *Garcinia Morella* Desr.

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ABSTRACT

*Phytochemicals are the secondary metabolites produced by plants in defense that are responsible for many useful medicinal aspects for mankind. *Garcinia morella* Desr. is well known for their edible fruits with high medicinal values, commonly known as 'kujee thekera' in Assam, India. The present paper is an attempt to perform the qualitative screening of phytochemicals of different leaf extract in ethanol, methanol, water and dichloromethane. The preliminary screening of different leaf extracts showed the presence of secondary metabolites like flavanoids, phenols, tannin coumarin, diterpenoids, etc. The antioxidant activity analysis performed by "DPPH assay" of the methanolic leaf extract with Ascorbic acid as the positive control revealed 64% DPPH scavenging at 100µg/ml concentration and IC₅₀ value of 16µg/ml. Whereas, the concentration 100 µg/ml of *G. morella* leaf extract exhibited 30.81% of DPPH scavenging with IC₅₀ of 225.70µg/ml.*

Key words: *Phytochemicals, antioxidant, *Garcinia morella*.*

INTRODUCTION

Plant derived substances are of great interest owing to their versatile applications. Medicinal plants are the richest bio-resource of drugs used in traditional system of medicines, nutraceuticals, food supplements, pharmaceutical intermediates and chemical entities for synthetic drugs (Ncube *et al.*, 2008). In recent times, medicines based on *Traditional Knowledge* have gained worldwide attention. The scientific community is focusing on proper pharmacological validation and identification of lead components of the plants used as sources of traditional medicines for the treatment of various ailments (Gurib *et al.*, 2006). The North East India is the home for such valuable traditional herbal remedies.

Phytochemicals or the chemicals produced by plants in defense have proved to be of immense usefulness to mankind. Many researches and studies have been done on plant phytochemicals for drug discoveries and still this trend of research is running successfully all over the world (Faraz *et al.*, 2003; Parekh and Chanda, 2008). Phytochemicals like flavanoids, steroids, phenols, tannins, etc have been extracted from plants for drug designing (Raina *et al.*, 2014). These phytochemicals or secondary metabolites are also known to have antioxidant activities. Antioxidant activity on the other hand is the capability of plant secondary metabolites to inhibit the free radicals produced in animal or human body. Many plants contain antioxidants compounds which protects the cell against the damaging effects of *Reactive Oxygen Species* (ROS) (Helen *et al.*, 2012). Thus, this role of phytochemicals throws light on how much of importance they play in the benefit of mankind. Dietary intake of phytochemicals may promote health benefits, protecting against chronic degenerative

disorders, such as cancer, cardiovascular and neurodegenerative diseases (Milbury *et al.*, 2002) and majority of foods, such as whole grains, beans, fruits, vegetables and herbs contain several phytochemicals.

Garcinia morella Desr. belonging to the family Cluciaceae, is one such medicinal plant used by traditional healers for the treatment of inflammatory disorders. It is an evergreen tree commonly known as Indian Gamboge. It is known as “*Kujee thekera*” in Assamese and “*Tamal*” in Bengali and Hindi. It is mainly distributed in India, Sri Lanka and Philippines and is present either in the form of wild or cultivated. In India, it is widely distributed in the regions of Sothern India, North-eastern India (Assam, Khasi Hills), Western-Ghats and also in the regions of West Bengal up to an altitude of 3000ft (Andersion, 1874). *Garcinia morella* is mainly a source of *Gamboge*- a gum resin with a wide range of uses (as in food, paints, medicines, etc.) that is commonly harvested and yellow latex (milky juice) of the plant is dried and sold in the market as *Kokum* or *Gamboge*; it is traditionally used by the local people of Assam in many ways; the fruit, leaves, gums are exploited by the locals for medicinal and cooking purposes; Oils and juice of fruits are cooling for fever, diabetes and jaundice (Barua *et al.*, 2012). Fruits are sliced and dried under sunlight to preserve it for long time which is used in treating dysentery by the *Bodo* tribes of Udalguri district of Assam (Patiri and Borah, 2007).

MATERIALS AND METHODS

Collection of plant material: The fresh leaves of *Garcinia morella* were collected from the Botanical Garden, Gauhati University, Assam, India.

Preparation of plant extract: The fresh leaves were collected and washed under running tap water to remove soil particles and then shade dried for 45 days and finally the dried leaves sample were ground well into a fine powder with the help of mixer grinder. A 20 gm air dried plant material was soaked into 200 ml organic solvents, *viz.* water, ethanol, methanol, diethyl methane for 24 hrs in a rotary shaker at 150 rpm in 30° C. The extracts were filter through the Whatman No: 1 filter paper and was allowed to evaporate. The condensed extracts were stored in airtight container at 4° C till further investigation.

Preliminary phytochemical screening: Phytochemical screening was performed to identify phytochemicals in the ethanol, methanol, water and dichloromethane extracts of plant leaves. The qualitative screening of powdered crude drugs for their active ingredients was carried out using the standard procedures (Trease and Evans, 1983; Brindha *et al.*, 1981).

DPPH antioxidant scavenging assay: The free radical scavenging activity was measured by the 1, 1-Diphenyl-2-picrylhydrazyl (DPPH) method proposed by Leong and Shui. DPPH solution of 0.1 mM was prepared freshly in methanol and kept away light and then from the initial absorbance was measured at 517 nm using Spectrophotometer (Beckman Coulter, DU730).

Final concentration of Standard ascorbic acid and plant extracts were make (stock sol=1mg/ml) at various concentration (25, 50, 75, 100) and then taken and final volume is adjusted to 10ml with methanol. The final volume were adjusted to 1ml by adding 1.25µl of methanolic extract and 375 µl of methanol in an ependorf tube of 1ml and then placed in dark for 30mins at 27⁰c. Methanol was used as blank and the experiment was expressed as the

inhibition percentage (%) of free radical by the sample and was calculated as the formula followed.

$$\text{Radical Scavenging Activity (\%)} = (\text{Abs control} - \text{Abs sample}) / \text{Abs control} \times 100$$

Where, Abs control is the absorbance of DPPH + methanol; Abs sample is the absorbance of DPPH radical + sample (i.e. extract or standard)

The IC₅₀ value of both the standard Ascorbic acid and the MEPT was calculated by using the software Prism 7.

RESULTS

Phytochemical studies are done on different plant extracts of *G. Morella* on various solvents. Because of their different solvent polarity they showed difference in presence or absence of chemicals. Water extract showed the presence of alkaloids, tannin and diterpenoids. Ethanol extract showed the presence of phenol, tannin coumarin, diterpenoids, and flavonoids. Methanol extract showed the presence of alkaloids phenol, tannin, coumarin, glycosides, flavonoids, and phytosterols. Dichloro methane showed the presence of phenol, coumarin, and diterpenoids.

Table 1. Photochemical screening of *Garcinia morella*

Serial No	Name of the phytochemicals	Water	Ethanol	Methanol	Dichloro methane
1.	Alkaloids	++	-	+	-
2.	Reducing sugar	-	-	-	-
3.	Phenols	-	++	+	+
4.	Tannins	+++	+	++	-
5.	Saponins	-	-	-	-
6.	Coumarins	-	++	++	+
7.	Glycosides	-	-	+	-
8.	Diterpenoids	+	++	-	+
9.	Flavonoids	-	+	+	-
10.	Phytosterols	-	-	+	-

(+++ Strong reactivity; ++ Moderate reactivity; + Weak reactivity; - Not detected)

DPPH radical scavenging activity: In the DPPH radical scavenging assay, antioxidants react with DPPH, and convert it to yellow coloured 1, 1-diphenyl 1, 1- picryl hydrazine. The degree of discoloration indicates the radical scavenging activity. In this test, *G. morella* extract exhibited a considerable antioxidant activity but not more than ascorbic acid. The antioxidant activity of ascorbic acid is highest with 64% DPPH scavenging at 100µg/ml concentration and IC₅₀ value of 16µg/ml. Whereas, the concentration 100 µg/ml of *G. morella* leaf extract exhibited 30.81% of DPPH scavenging with IC₅₀ of 225.70µg/ml.

In-vitro antioxidant property analysis: It was observed that the methanolic extract of *G. morella* have demonstrated dose dependent increase in the DPPH radical scavenging. Ascorbic acid (standard) has shown IC_{50} at 16.33 $\mu\text{g/ml}$ concentration obtained by equation ($Y= 0.2211X + 46.39$) whereas MEPT has shown IC_{50} at 225.70 $\mu\text{g/ml}$ concentration obtained by equation ($Y= 0.1572X + 14.52$) (Table 4.2.; Fig. 4.5).

Table 2. Analysis of DPPH radical scavenging

Concentration ($\mu\text{g/ml}$)	Scavenging %	
	Ascorbic acid	<i>Garcinia morella</i>
25	57.4 ± 4.446	18.83 ± 1.236
50	59.90 ± 2.052	22.18 ± 6.299
75	62.99 ± 1.674	25.53 ± 0.404
100	64 ± 1.218	30.81 ± 0.498

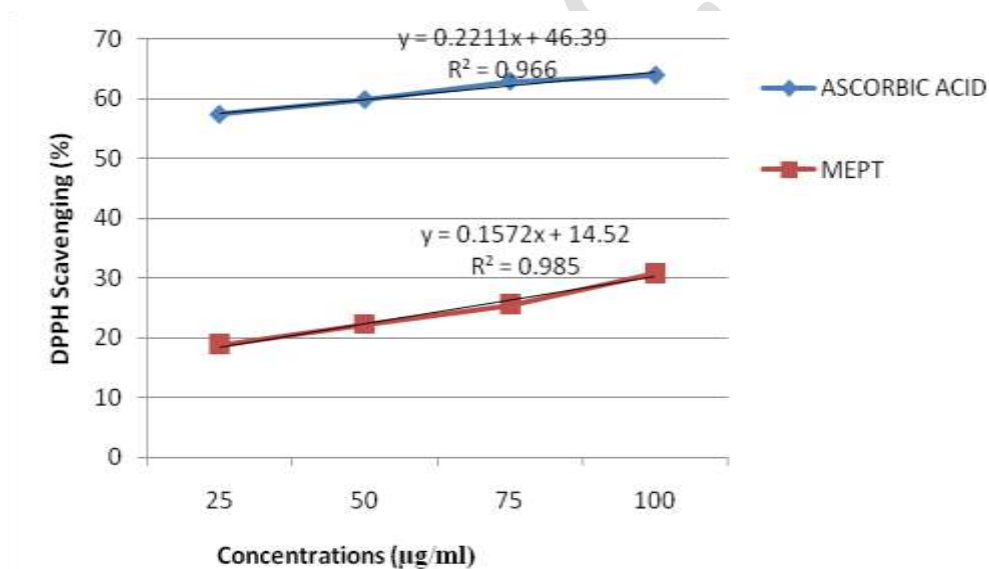


Fig. 1. DPPH radical scavenging activity (%) versus concentration ($\mu\text{g/ml}$).

Table 3. Concentration of extract for 50% free radical scavenging (IC_{50})

Sample extract	DPPH IC_{50} ($\mu\text{g/ml}$)
Ascorbic acid	16.33
<i>G. morella</i> extract	225.70

DISCUSSION

Studies on plant secondary metabolites have been increasing over the last 50 years. These molecules are known to play an important role in adaptation of the plant to their environment, but also represent an important source of active pharmaceuticals (Deka *et al.*, 2017). These chemicals are extremely diverse and many thousands of these have been identified in several major classes. Each family, genus and species produces a characteristic mix of these chemicals, and they can sometimes be used as taxonomic characters in classifying plants. These natural products are potential source of diverse range of medicines or important bioactive compounds for human benefits. There are some reports of phytochemical diversity with antimicrobial activity which includes terpenoids, saponins, phenolics and phenyl propanoids, pterocarpus, stilbens, alkaloids, glucosinolates, hydrogen cyanide, indole and also elemental sulphur, the sole inorganic compounds (Cooper *et al.*, 2001).

Various researches are being carried out regarding phytochemical and antioxidant analysis. The qualitative analysis of phytochemical profile of fruit rind of *Garcinia cambogia* or *morella* (Krishnamurthy *et al.*, 2014) showed the presence of flavonoids, terpenoids, saponins, tannins and reducing sugars in ethanol, ethyl acetate and aqueous extracts of the plants. An antioxidant analysis on the fruits of three *Garcinia* sp. (*Garcinia pedunculata* Roxb. ex Buch.-Ham., *G. xanthochymus* Hook. f. and *G. morella* Gaertn. Desr) were reported by some earlier worker where *G. pedunculata* Roxb. ex Buch.-Ham. showed highest antioxidant potential with IC₅₀ value 47.03±13.48 µg/ml whereas *G. morella* showed the lowest antioxidant potential among the three species (Gogoi *et al.*, 2012).

Antioxidant and antifungal activities of polyphenol rich extracts of dried pulp of *Garcinia pedunculata* Roxb. and *Garcinia Morella* Gaertn was evaluated by DPPH assay in CW and HW extract and antioxidant potential was found higher in case of CW extract in comparison IC₅₀ value of HW extract (Sarma *et al.*, 2016). The antioxidant and anticancer activities of *G. morella* (GM) methanol extracts against DLA tumor models and elucidate the probable molecular mechanism of action activity also validated in the fruits of *Garcinia morella* (Choudhury *et al.*, 2016).

The present study was conducted to investigate the phytochemical properties and antioxidant assay of *G. morella* where different extract of leaves were analysed. The Water extract showed the presence of alkaloids, tannin and diterpenoids. Ethanol extract showed the presence of phenol, tannin coumarin, diterpenoids, and flavonoids. Methanol extract showed the presence of alkaloids, phenol, tannin, coumarin, glycosides, flavonoids, and phytosterols. Dichloro methane extract showed the presence of phenol, coumarin, and diterpenoids. Antioxidant potential calculation by DPPH assay showed *G. morella* extract exhibited a considerable antioxidant activity but not more than ascorbic acid. The antioxidant activity of ascorbic acid is highest with 64% DPPH scavenging at 100 µg/ml concentration and IC₅₀ value of 16.33 µg/ml. The concentration 100 µg/ml of *G. morella* methanolic leaf extract exhibited 30.81% of DPPH scavenging with IC₅₀ of 225.70 µg/ml

CONCLUSION

Garcinia morella is an excellent anti inflammatory herb which is used by the locals and tribal of north eastern India to cure stomach ailments, bowel disorders and inflammatory diseases.

This plant is enriched with various types of phytoconstituents present in the form of secondary metabolites that ensures its medicinal value. Keeping this in view the present investigation was carried out on its antioxidant property that may mainstay in exploring other therapeutic potency. Hence phytochemical investigation followed by antioxidant property was evaluated on the leaf of *G. morella* which showed the leaf extract of *G. morella* exhibiting a considerable antioxidant activity but not more than ascorbic acid. The present study showed that this plant can be considered as a good source of natural antioxidant and should be further analyzed for their chemical and biological properties. The curative properties of this plant may depend mainly on the phytochemicals present in it. Presence of some phytochemicals also gives the hint of presence of some antioxidants in plant material, and as it has been reported specially in *G. morella*, the presence of an antioxidant may serve as a raw material for the production of anti-cancerous drugs.

With this study it could be concluded that the phytochemical and antioxidant assay of *G. morella* would give valuable information for further characterization and exploitation of this important groups of medicinal plant of Assam, India

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