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Studies on nutraceutical properties of *Flacourtia jangomas* fruits in Assam, India

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Abstract
Searching of nutraceutical properties of fruits has in recent years received much attention in different parts of the world owing to its relevance in discovery of health beneficial foods. *Flacourtia jangomas,* a semi-cultivated fruit plant, having some medicinal as well as economic value, is found frequently in the Brahmaputra valley of Assam and adjoining areas in the northeastern parts of India. The present study has been carried out to make a survey for bioactive compounds, like, alkaloids, flavonoids, phenols, terpenoids, tannins and saponins, and to determine the total phenol and flavonoid contents in the ripe fruits of *F. jangomas.* It has found that the methanol extract of the fruits contains most of the searching bioactive compounds and the total phenol and flavonoid contents are 20 mg/g and 2mg/g respectively. Findings of the study have provided the evidences that the fruits of *F. jangomas* have the nutraceutical potency.

Keywords: *Flacourtia* jangomas, nutraceuticals, secondary metabolites

1. Introduction
Nutraceutical, comparatively a newer term in the food science research, is used to describe any product derived from food sources that provides extra health benefits, *i.e.*, able to decrease the risk of disease, in addition to the basic nutritional value found in food. The term nutraceutical is a hybrid of nutrition and pharmaceutical, coined in 1989 by Stephen Defelice [1]. Searching of the nutraceutical properties of foods has in recent years received much attention in different part of the world owing to its relevance in discovery of health beneficial foods. Fruits have been recognized as one of the most valuable sources of nutraceuticals due to presence of bioactive compounds like alkaloids, terpenoids, tannins, saponins and polyphenols. The most commonly occurred polyphenol in food are flavonoid and phenolic acid [2]. These bioactive compounds are produced through secondary metabolism in different plants. The nutraceutical value of these substances lies because they have definite physiological action on the human body [3].

Assam, a state in the North-Eastern region of India, is one of the richest biodiversity hotspot of the world due to diverse topography, climate and agro-ecological conditions. A number of plant species including fruits have their origin in this region, many of which are still grown in wild or semi-wild states. Despite the vast genetic diversity of these fruits, only a few have been grown as commercial crops for their economic, social and religious importance. However, a number of fruits remained confined in semi-wild or semi-domesticated conditions and are rarely known in other parts of the country. These underutilized fruits have multipurpose uses and therefore play significant role, especially, for the wellbeing of rural people by providing nutrition, household income and employment. Many of these fruits have been used as traditional medicinal plants and some have found important place in the Indian system of Medicine and Unani since time immemorial. Moreover, these plant species are good tools for scientific investigations as well as important genetic resources of the country. *Flacourtia jangomas* (Lour.) Raeusch (family: Flacourtiaceae), locally known as 'Panial', is a semi-cultivated fruit plant found frequently in the Brahmaputra valley of Assam and adjoining areas in the northeastern parts of India. The plant has some medicinal as well as economic values. It is mainly cultivated for its edible fruit and hard wood. The fruits are dark red or purple when ripe (Photo-plate 1) and eaten raw or used for making jams and preserves [4-5]. Different plant parts are also pharmaceutically used for the treatment of asthma, pre- and post-natal blood purification [6]. The fruits are used in bilious conditions and in diarrhea [7]. In the line of phytochemical analysis, however, works have been found to be scanty [8].

Thus, to explore the nutraceutical properties, a phytochemical study for secondary metabolites and the total phenol and flavonoid contents in the fruits of *F. jangomas* was conducted.

2. Materials and Methods

2.1 Collection of plant material and preparation of Extract

The ripe fruits were collected during the months of June-July from the locality. Fresh fruits were dried under shade, powdered, extracted in methanol by soxhlet apparatus. The extract was evaporated under reduced temperature and pressure and finally lypohilized. The residue was used to perform preliminary qualitative tests for detection of secondary metabolites as well as to conduct quantitative analysis for total phenol and flavonoid contents.

2.2 Phytochemical Screening for secondary metabolites

Chemical tests were carried out qualitatively on the extract following standard procedures to identify the phytochemical constituents [9-10].

A. Test for alkaloids

Dragendorff's test: In a test tube containing 1 ml of extract, few drops of Dragendorff's reagent was added and the colour developed was noticed. Orange colour did not appeared, indicated the absence of alkaloids.

Mayer's test: To 1 ml of the extract, 2 ml of Mayer's reagent was added, a dull white precipitate did not found, indicated the absence of alkaloids.

B. Test for flavonoids

Alkaline reagent test: To the test solution, a few drops of sodium hydroxide solution were added. Formation of intense yellow colour which turns to colourless by addition of few drops of dilute acetic acid indicated the presence of flavonoids.

Shinoda test: To the test solution, a few drops of concentrated HCl and a few pieces of magnesium turning were added. Development of pink or magneta red colour indicated the presence of flavonoids.

C. Test for phenolic compounds

Ferric chloride test: To the test solution, a few drops of ferric chloride solution were added. A dark green colour indicated the presence of phenolic compounds.

D. Test for tannins

Lead acetate test: To the test solution, a few drops of 10% lead acetate solution were added. Precipitate formation indicated the presence of tannin.

E. Test for terpenoids

Salkowski's test: Extracts were treated with chloroform and filtered. The filtrates were treated with few drops of concentrated sulphuric acid, shaken well and allowed to stand. Appearance of red colour in the lower layer indicates the presence of steroids. Formation of reddish brown colour of interface after addition of concentrated sulphuric acid to the side carefully (without shaking) indicated the presence of terpenoids.

F. Test for saponins

Foam test: Crude extract was mixed with 5 ml of distilled water in a test tube and it was shaken vigorously then some drops of olive oil were added. The formation of stable foam was taken as an indication for the presence of saponins.

2.3 Determination of total phenol and flavonoid content

Folin-Ciocalteu method, as described by Nabavi *et al* [11], was used for phenol content determination. Briefly, 100 mg plant sample was dissolved in 10 ml methanol of 50% (v/v with distilled water). The solution was filtered. 0.5 ml of the filtrate was mixed with 2 ml of Folin-Ciocalteu reagent (1:1 diluted with distilled water) and mixed thoroughly. After five minutes 2 ml of 10% Na_2CO_3 solution was added. The solution was warmed for one minute, and then cooled. After one hour at room temperature absorbance was measured at 760 nm with UV-Visible spectrophotometer. Sample blank was concomitantly prepared containing 0.5 ml distilled water, 2 ml of Folin-Ciocalteu reagent and 2 ml of 10% Na_2CO_3 dissolved in water. Total phenol content was calculated as gallic acid equivalent from a calibration curve. The calibration curve was prepared by preparing gallic acid solutions at concentration 10, 25, 50, 100, 200 and 250 µg/ml in methanol (50%). Total phenol content is expressed in terms of gallic acid equivalent as mg/g of dry mass.

Colorimetric aluminum chloride method, as described by Nabavi *et al.* [11], was used for flavonoid content determination. Briefly, 100mg plant sample was dissolved in 10 ml of methanol. The solution was filtered. 2 ml of the filtrate was mixed with 0.1 ml of 10% aluminum chloride, 0.1 ml of 1M potassium acetate and 2.8 ml of distilled water. The mixture was shaken and kept at room temperature for 30 minutes. The absorbance of the reaction mixture was measured at 415 nm with UV-Visible spectrophotometer. Total flavonoid content was calculated as quercetin from a calibration curve. The calibration curve was prepared by preparing quercetin solutions at concentrations 5, 10, 25, 50, 80 and 100 µg/ml in methanol. The blank sample was prepared in similar way by replacing aluminum chloride with distilled water. Total flavonoid content is expressed in terms of quercetin equivalent as mg/g of dry mass.

3. Result and Discussion

The present study carried out on the methyl alcoholic extract of fruits revealed the presence of most of the studied secondary metabolites except alkaloids i.e., flavonoids, phenols, tannins, terpenoids and saponins were found to be present. The result is summarized in Table 1.

Polyphenols form a large group of chemicals and most commonly occurred polyphenols in food include flavonoids and phenolic acids. Dietary polyphenols are of current interest because substantial evidence *in vitro* have suggested that they can affect numerous cellular processes like, gene expression, apoptosis, platelet aggregation, intercellular signaling, that can have anti-carcinogenic and anti-antherogenic implications [2]. Apart from these, various experiments have been demonstrated that flavonoids and phenolic acids are potential antioxidant and antioxidant activity of these compounds is due to their ability to scavenge free radicals. Accumulation of free radicals can cause pathological conditions such as asthma, arthritis, inflammation, neuro-degeneration, heart disease, aging effect, etc. [12]. Phenolic compounds, moreover, act as (i) metal chelators, (ii) anti-mutagens and anti-carcinogens, (iii) antimicrobial agents [13]. Tannins and terpenoids are attributed for analgesic and anti-inflammatory activities. Furthermore, tannins contribute property of astringency i.e., faster the healing of wounds and inflamed mucous membrane [14]. Saponin, likewise, has the potential to lower cholesterol levels in humans due to their

hypocholesterolemic effect. Saponins form complexes with cholesterol to reduce cholesterol levels [15].

Total amount of phenol and flavonoid contents were calculated from gallic acid (y = 0.011 – 0.066, R^2 = 0.999) and quecetin (y = 0.032 – 0.077, R^2 = 0.999) standard curves (Figure 1 & 2). The total phenol and flavonoid contents in their extract were found 20 mg/g and 2 mg/g in terms of gallic acid and quercetin equivalent respectively (Table 2). The phenol and flavonoid contents were well confirmed with qualitative investigations. Although, the total amount of flavonoid content (2 mg/g) found to be moderate, but, amount of the total phenol content (20 mg/g) comparatively high which is impressive.

Table 1: Secondary metabolites constituents in the methyl alcoholic extract of fruits of *F. jangomas*

Secondary metabolites	Chemical tests	Indication: '+' for presence '-' for absence
Alkaloids	Mayer's test	-
	Dragendorff's test	-
Flavonoids	Alkaline test	+
	Shinoda test	+
Phenols	Ferric chloride test	+
Tannins	Lead acetate test	+
Terpenoids	Salkowski's test	+
Saponins	Foam test	+

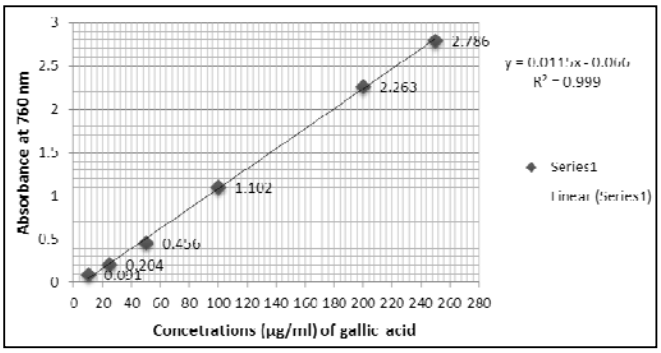

Fig 1: Standard calibration curve of Gallic acid for the determination of total phenol content.

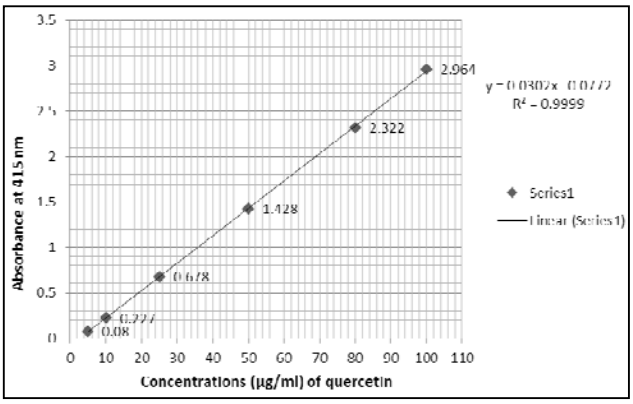

Fig 2: Standard calibration curve of quercetin for the determination of total flavonoid content.

Photo-plate 1: Ripe fruits of *F. jangomas*

Table 2: Total amount of phenol and flavonoid contents of *F.jangomas* fruits.

Plant part/ Extract name	Total phenol content (in mg/g, gallic acid equivalent)	Total flavonoid content (in mg/g, quercetin equivalent)
Ripe fruits/ Methanol extract	20	2

4. Conclusion

In conclusion, the investigation has revealed the nutraceutical potency of the fruits of *F. jangomas* as it contains most of the bioactive compounds which are vital for good health. Further, the findings of the study have provided evidences that crude extract of the fruits contain medicinally important bioactive compounds and justices the uses of the plant in the indigenous medicine for the treatment of different diseases.

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