EXTRACTION AND PHYTOCHEMICAL SCREENING OF SYZYGIUM CUMINI SEEDS IN VIDARBHA REGION OF INDIA

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ABSTRACT

In India, an extraordinary variety of climatic regions, ranging from tropical in the south to temperate and alpine in the Himalayan north, where elevated regions receive sustained winter snowfall. The nation's climate is strongly influenced by the Himalayas and the Thar Desert. Vidarbha has three main seasons: the wet Monsoon and post-Monsoon season from June to October, the cool dry winter from October to March and the hot dry season from April till the onset of rains. The temperature of Vidarbha ranges from a minimum of 12-25°C to a maximum of 30-48°C with relative humidity varying from 10-15% to 60-95%. Annual precipitation is 1700 mm 90% of the precipitation falls in four months, i.e. from June to September. Syzygium Cumini have various names in India and other country likes jamun, jambolan, Java plum or black plum, is an evergreen tropical tree in the flowering plant from family Myrtaceae, and is one of the most popular fruits. It is planted in various regions spontaneous. It is native of India, Myanmar, Bangladesh, Sri Lanka, Nepal, China, Australia, Thailand, Colombia, United States of America, Zimbabwe Zambia and Mexico. The entire or whole part of the plants has been widely used in the treatment of various diseases in the traditional and folk medicine. The edible part of fruits (jamun) contain vitamin C, gallic acid, tannins, anthocyanins, includes cyanidin, petunidin, malvidin-glucoside and other components. The seeds of Syzygium Cumini possess anti-diabetic, antipyretic, anti-inflammatory, hypolipidaemic, psychopharmacological, anti-diarrheal, antioxidant and antibacterial activities. Now today study, as per climatic condition in Vidharba region of India, the phytochemical studies were carried out with using methanol, petroleum ether and
ethanol extracts of the seeds of *Syzygium cumini* from the family Myrtaceae which is collected from local market of Chandrapur district of Maharashtra state in India in July, 2017. Preliminary the phytochemical screening of all extracts revealed the presence of phytoconstituents like alkaloids, tannins, saponins, phenols, flavonoids, terpenoids, steroids and amino acids and absence of anthraquinone glycosides.

**KEYWORDS:** In India, an extraordinary Chandrapur district of anthraquinone glycosides.

**INTRODUCTION**

*Syzygium cumini* (Family Myrtaceae) is also known as *Syzygium jambolanum* and *Eugenia cumini*. Other common names are Jambul, Black Plum, Java Plum, Indian Blackberry, Jamblang, Jamun etc. Today these trees are found growing throughout the Asian subcontinent, Eastern Africa, South America, Madagascar and have also naturalized to Florida and Hawaii in the United States of America (Warrier et al., 1996). The tree fruits once in a year and the berries are sweetish sour to taste. The ripe fruits are used for health drinks, making preserves, squashes, jellies and wine (Warrier et al., 1996). In association to its dietary use, all parts of the tree and, importantly the seeds are used to treat a range of ailments, the most important being diabetes mellitus (Sagrawat et al., 2006). Different parts of the jambolan were also reported for its antioxidant, anti-inflammatory, neuropsycho-pharmacological, anti-microbial, anti-bacterial, anti-HIV, antileishmanial and antifungal, nitric oxide scavenging, free radical scavenging, anti-diarrheal, antifertility, anorexi-genic, gastroprotective and anti-ulcerogenic and radio-protective activities (Sagrawat et al., 2006). Almost 25% of globally prescribed drugs obtained from the various plants sources, used as the natural products for diseases prevention and control as well as in drug development. It has also been very well documented in the world forum WHO’s report that state about 80% of world’s population are dependent on plants to meet their primary health care needs (Ahmadullah and Nayar, 1999). Plant metabolites have been of great interest to human for long time due to their pharmacological relevance. A large proportion of world population, especially in the developing countries depends on traditional system of medicine for various diseases. Plant based drugs constitute a major share of medicine in India, china viz ayurveda, yoga, unani, siddha, homeopathy and naturopathy, except allopathy (Vaidya & Devasagagam, 2007).

It can live up to 80-100 years, and planted in the various regions spontaneous. It is native of India, Bangladesh, Sri Lanka, Myanmar, Nepal, China, Australia, Thailand, Kenya,
Colombia, Mexico, United States of America, Zambia, and Zimbabwe. (Adelia et al., 2011) The entire part of the plants has been widely used in the treatment of various diseases in the traditional and folk medicine. The chief active constituents of Syzygium Cumini are myricetine, β-sitosterol, myricyl alcohol, betulinic acid, friedeanol, epifriedeanol, eugenin, β-sitosterol-D-glucoside, Kamepferol-3-0- glucoside, quercetin, astragalin, and gallic acid. The edible part of fruits (jamun) contain vitamin C, gallic acid, tannins, anthocyanins, includes cyanidin, petunidin, malvidinglucoside and other components. The seeds of Syzygium Cumini possess anti-diabetic, antipyretic, hypolipidaemic, psychopharmacological, anti-diarrheal, anti-inflammatory, antioxidant and antibacterial activities. It is reported that seed extracts of Syzygium Cumini given to the animals with the dose of 5gm/kg of bodyweight was more effective than the oral hypoglycaemic or anti-diabetic drug glibenclamide in the case of type 2 diabetes. The preparation of tea from the leaves of Syzygium Cumini has also a hypoglycaemic effect used in the case of diabetes. The flowers are believed to possess an antipyretic effect to reduce the fever. The seeds are additionally used as an anesthetic in South American cultures. The leaves of S. cumini are also used in the treatment of various skin diseases. The Leaves, seeds, fruits and stem of jamun tree have some antimicrobial effect (Sharma S, et al. 2012). Preliminary phytochemical screening also showed the presences of alkaloids, tannins, saponins, flavonoids, phenols, terpenoids, steroids and amino acids and absence of anthraquinone glycosides in seeds extracts of syzygium cumini. The present study was designed to investigate the phytochemical compounds and antibacterial activity of the methanolic, petroleum ether and ethanolic extracts of Syzygium Cumini seeds. (Adelia et al., 2011).

**Taxonomic Classification**

<table>
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<tr>
<th>Kingdom</th>
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<td>Syzygium</td>
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<td>Specie</td>
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MORPHOLOGY
Black plum or jamun is a tropical evergreen tree that grows up to 25-30 meters (80-100 feet) tall, with white gray coloured stems and coarse and discoloured lower bark. The leaves have a characteristic smell like turpentine, and are simple, dark green, opposite, oblong-oval or elliptical, glossy, smooth, leathery in touch and blunt or tapering at the apex point. The leaves are 5-25 centimetres long and 5-10 centimetres wide in size. (Adelia et al., 2011) The midrib of the leaves is prominent and yellowish in colour when mature. The leaf blades have many lateral veins closely parallel. The colours of flowers are white-pinkish, appear in clusters about 4-10 centimetres long, and each being 1-2 cm long and 1.5 cm wide in size across with the four to five united petals and many stamens. The calyx is look like funnel shaped. The fruits appear in the clusters form of 10-50, are ovoid, one seeded berry, 2-4 centimetres long, dark purple, shiny red, dark brown or nearly black in colour. The fruit is generally astringent, sometime unpalatable and flavour varies from sour to fairly sweet (Ayyanar and Subhashbabu, 2012).

Figure: Syzygium Cumínee seeds and fruits.

Composition of Fruit
Analyses of the fruit in the Philippines were reported in 1924 as follows: Waste, 25%; edible portion: water, 80.80%; ash, 0.70; protein, 0.81; sugar, 12.70 (fructose and glucose; no sucrose); acidity (as sulphuric), 0.63%; (as malic) 0.88% (Wester, 1924). The following composition per 100 grams of edible portion was reported for fruits freshly picked at the Lancetilla Experimental Garden, Honduras, in 1948: Moisture, 85.8 gm; ether extract, 0.15 gm; crude fiber, 0.3 gm; nitrogen, 0.129 gm; ash, 0.32 gm; calcium, 8.3 mg; phosphorus, 16.2 mg; iron, 1.62 mg; carotene, 0.004 mg; thiamine, 0.008 mg; riboflavin, 0.009 mg; niacin, 0.290 mg; total ascorbic acid, 5.7 mg (Munsell et al., 1949). Virmani gives the following
analysis: specific gravity, 1.0184; total acidity (as acetic acid), 5.33 per 100 cc; volatile acidity (as acetic acid), 5.072 per 100 cc; fixed acidity, 0.275% as citric; total solids, 4.12 per 100 cc; ash, 0.42; alkalinity of ash, 32.5 (N/10 alkali); nitrogen, 0.66131; total sugars, 0.995; reducing sugars, 0.995; non-volatile reducing sugars, 0.995; alcohol, 0.159% by weight; oxidation value (KMnO4, 186.4); iodine value, 183.7; ester value, 40.42. Other reported constituents of the seeds are: protein (6.3 to 8.5%), fat (1.18%), crude fiber (16.9%), ash (21.72%), calcium (0.41%), phosphorus (0.17%), fatty acids (palmitic, stearic, oleic and linoleic), starch (41%), dextrin (6.1%), a trace of phytosterol, and 6 to 19% tannin (Ranjan et al., 2011). The fruits are avidly eaten by birds and four footed animals (jackals and civets in India). In Australia, they are a favorite food of the large bat called “flying fox”. Analyses of the leaves show: crude protein (9.1%), fat (4.3%), crude fiber (17.0%), ash (6.0%), calcium (1.3%), phosphorus (0.19%) (Giovannucci et al., 2002). It consists mainly of mono- or sesqui-terpene hydrocarbons which are “very common in essential oils.” Constituents of Syzygium cumini seeds are fatty oils (30g/kg), including lauric (2.8%), myristic (31.7%), palmitic (4.7%), stearic (6.5%), oleic (32.2%), linoleic (16.1%), malvalic (1.2%), stercurlic (1.8%) and vernolic acid (3%) and phytoesters such as β-sitosterol. Further constituents are tannins (6%), predominantly corilagin, ellagitannins, ellagic acid, gal-loyl-galactoside and gallic acid (Lock et al., 2009). The leaf oil consists of 16.91% octadecane, 9.98% nonacosane, 9.38% triacontane, 7.38% octacosane, 4.86% Heptacosane, 4.25% hexadecanoic acid and 4.02% eicosane. The seed oil consists of 33.2% 1-chlorooctadecane, 9.24% tetractetracontane, 8.02% decahydro-8a-ethyl-1,1,4a,6-tetramerthynapthalene, 5.29% 4-(2-2-dimethyl-6-6-methylene- cyclohexyl) butanol, 5.15% Octadecane, 3.97% octacosane, 1.72% heptacosane and 1.71% eicosane. (Kumar et al., 2009). Java Plum consist of Energy 251 kJ (60 kcal), Carbohydrates 15.56 g, fat 0.23 g, Protein 0.72 g, water 83.13 g, Vita-min A 3IU, Thiamine (vit B1) 0.006 mg (1%), Ribofla-vin (vit. B2) 0.012 mg (1%), 0.260 mg (2%) Niacin (vit. B3), 0.160 mg (3%) Pantothenic acid (B5), 0.038 mg (3%) Vitamin B60.038 mg (3%), 14.3 mg (17%) Vitamin C, 19 mg (2%) Calcium, 0.19 mg (1%) Iron, 15 mg (4%) Magnesium, 17 mg (2%) Phosphorus, 79 mg (2%) Potas-sium, 14 mg (1%) Sodium.[9] The Fruit Contain 83.70 - 85.80 g moisture, 0.70 - 0.13 g protein, 0.15 - 0.30 g fat, 0.30 - 0.90 g crude fibre, 14.00 g carbohydrate, 0.32 - 0.40 h ash, 8.30 - 15.00 mg calcium, 35.00 mg magnesium, 15.00 - 16.20 mg phosphorus, 1.20 - 1.62 mg iron, 26.20 mg sodium, 55.00 mg potassium, 0.23 mg copper, 13.00 mg sulfur, 8.00 mh chlorine, 8. I.U vitamin A, 0.01 - 0.03 mg thiamine, 0.009 - 0.01 mg riboflavin, 0.20 - 0.29 mg niacin, 5.70 - 18.00 mg ascorbic acid, 7.00 mg chlo-rine and 3.00 mcg folic acid per 100 g of edible portion.
Medicinal Properties
The jambolan has received far more recognition in folk medicine and in the pharmaceutical trade than in any other field. Medicinally, the fruit is stated to be astringent, stomachic, carminative, antiscorbutic and diuretic (Srivastava et al., 1983). Additionally, a fruit extract showed antimicrobial and cytotoxic activities and may potentially be used on topical antimicrobial products. In comparison to other non-traditional fruits jambolao showed considerable high antioxidant activity, which can constitute such as anthocyanins, tannins and flavonols (Gordon et al., 2011). The anthocyanin composition was characterised by the presence of 3,5- diglucosides of five out of six aglycones commonly found in foods (Adelia et al., 2011).

MATERIALS AND METHODS
Collection of Plant Materials
The fresh fruits of Syzygium Cumini were collected from the local region of city Bhadrawati, Maharashtra, India in second week of July, 2017. The collected fruits were washed under running tap water to remove the dust particles, fruit pulp was separated, seeds were cleaned thoroughly, dried at room temperature for 1-2 weeks and finally crushed into the powder by using electrical grinder.

Preparation of Plant Extracts
The powdered sample were percolated by using Soxhlet apparatus successively with the organic solvent such as methanol, petroleum ether and ethanol (70% w/v) respectively. The extracts were taken and kept for further studies.

Screening for Phytochemical Compounds
The seed extracts of Syzygium Cumini were analysed for the presence of Phytoconstituents such as alkaloids, tannins saponins, flavonoids, phenols, terpenoids, steroids, amino acids and anthraquinone glycosides according to the standard methods (Harborne 1998), (Kokate 2001).

Test for alkaloids[Mayer’s Test]
1.36 gm of mercuric chloride dissolved in 60 ml and 5 gm of potassium iodide were dissolved in 10 ml of distilled water respectively. These two solvents were mixed and diluted to 100 ml using distilled water. To 1 ml of acidic aqueous solution of samples few drop of reagent was added. Formation of white or pale precipitates shows the presence of alkaloids.
Test for Tannins

Ferric Chloride Test

Extract (50 mg) + distilled water

Add few drops of neutral 5% ferric chloride solution

Dark green or black precipitates

Indicates presence of tannins

Lead acetate Test

A test tube containing 5 ml of sample

A few drops of 1% solution of lead acetate

Formation of bulky white precipitates

Indicates presence of tannins

Test for Saponins

A drop of sodium bicarbonate was added in the test tube containing 50 ml extract of the sample. The mixture was vigorously shaken and kept for two minutes. A honey comb like froth was formed and it showed the presence of saponins.

Test for Flavonoids

In a test tube containing about 0.5 ml of alcoholic extract of sample, 5-10 drops of diluted Hydrochloric acid and small amount of Mg or Zn were added and the solution was boiled for few minutes. Appearance of reddish pink or dirty brown colour indicates the presence of flavonoids.

Test for Phenols

To 1 ml of the alcoholic solution of the sample, 2 ml of distilled water followed by few drops of the 10% aqueous solution of ferric chloride were added. Formation of blue or deep green colour indicates the presence of phenols.
Test for Terpenoids

In a test tube, 2 ml of chloroform and 5-10 drops of conc. H_{2}SO_{4} were added to 1mg of extract and observed for reddish brown colour that indicated the presence of terpenoids.

Test for Steroids [Salkowski’s Test]

About 100 mg of dried extract of sample was dissolved in 2 ml of CHCl_{3}. H_{2}SO_{4} was added carefully to form a lower layer. A reddish brown colour at the interface was an indicative of steroidal ring.

Test for Amino acids

2 ml of extract sample was treated with the 1-2 drops of ninhydrin reagent. Appearance of violet or purple colour indicates the presence of amino acids.

Test for Anthraquinone Glycosides

Borntrager’s Test: In a test tube containing 5 ml of extract, 2 ml of dilute H_{2}SO_{4} was added, boil for 5 min and filtered. To the filtrates, equal volumes of CHCl_{3} was added and mixed well. Organic layer was separated and 10% ammonia solution was added to this. Appearance of brick pink colour of the ammonia layer confirmed the presence of anthraquinone glycosides.

RESULT AND DISCUSSION

Table I: Phytochemical screening test of Syzygium Cuminiseed extracts.

<table>
<thead>
<tr>
<th>Phytoconstituents</th>
<th>Organic Solvents</th>
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<tbody>
<tr>
<td></td>
<td>Methanolic Extract</td>
</tr>
<tr>
<td>Alkaloids</td>
<td>+</td>
</tr>
<tr>
<td>Tannins</td>
<td>+</td>
</tr>
<tr>
<td>Saponins</td>
<td>+</td>
</tr>
<tr>
<td>Flavonoids</td>
<td>+</td>
</tr>
<tr>
<td>Phenols</td>
<td>+</td>
</tr>
<tr>
<td>Terpenoids</td>
<td>+</td>
</tr>
<tr>
<td>Steroids</td>
<td>-</td>
</tr>
<tr>
<td>Amino acids</td>
<td>+</td>
</tr>
<tr>
<td>Anthraquinone glycosides</td>
<td>-</td>
</tr>
</tbody>
</table>

(+) = Presence of phytoconstituents
(-) = Absence of phytoconstituents
CONCLUSIONS

In this present study, the phytochemical screening activities were performed with methanol, petroleum ether and ethanol extracts of the seeds of *Syzygium Cumini* which are collected from the local region of city Bhadrawati, dist-Chandrapur, Mahararashtra, India. The results of phytochemical screening studies shows the Syzygium Cuminiseeds were rich in alkaloids, tannins, saponins, flavonoids, phenols, terpenoids, steroids and amino acids (as shown in Table I).

REFERENCES


