Phytochemical analysis of leaf, bark and fruit extracts of *Baccaurea courtallensis* Muell. Arg

Aiswarya KP, Sruthy Unnikrishnan N, Mahesh S and Laija S Nair

**Abstract**

*Baccaurea courtallensis* Muell. Arg is an evergreen tree of Euphorbiaceae family endemic to the Western Ghats. The bark of the plant is used as a tonic in disorders of mucous membrane and to heal wounds and its root is used in controlling diabetes. The present study was aimed to investigate the phytochemical profile of *Baccaurea courtallensis* Muell. Arg. The leaf, bark and fruit powder was extracted with methanol and hexane. Phytochemical analysis showed the presence of tannins, saponins, terpenoids and phenolic compounds. The present study provides evidence that solvent extract of this plant contains medicinally important bioactive compounds. The phytochemical analysis of the plants is very important commercially and for the production of the new drugs for curing of various diseases.

**Keywords:** Phytochemical screening, qualitative and quantitative analysis, bioactive compounds

**Introduction**

Phytochemicals are natural bioactive compounds found in different parts of the plant that interplay with nutrients and dietary fiber to protect them. Phytochemical studies afford revelation and understanding of phytoconstituents, as much as possible conserving their bioactivities compared with the crude herbal methods that are not easily standardized. Medicinal value of plant lie in some chemical substances that produce a definite physiological action on human body. The most important of these bioactive constituents of plant are alkaloids, tannins, flavanoids and phenolic compounds. Many of the indigenous medicinal plants are used in food for medicinal purpose (Okwu, 2004; Hill, 1952) [10, 5] *Baccaurea courtallensis* Muell. Arg belonging to Euphorbiaceae are used as food and in treatment of infectious diseases such as diarrhoea, dysentery, skin infection (Uduak A E & Kola K A, 2010) [13]. Flowering occur during the month of February and March, fruits are borne during the month of May and June. Fruits are edible. Fruits are harvested by the local tribal population of the region for medicinal purpose (Srinivas et al 2009) [14]. The present study was conducted to screen different phytochemicals present in methanol and hexane extract of different parts of *Baccaurea courtallensis* Muell. Arg.

**Materials and Methods**

a. **Collection and preparation of plant material**

The plant material was collected from Thiruvananthapuram, Kerala, India. The plant parts were shade dried for 1-3 weeks and powdered in mechanical grinder and stored in closed vessel. Then 10 gm dried powder was mixed in 100 ml Methanol and n- Hexane solvents and kept under shaker for overnight. The mixture was filtered through Whatsmann no 1 filter paper to Petriplates and allowed to evaporate the solvents. The extract were kept in sterilized microcentrifuge tube and stored in refrigerator for further use.

b. **Qualitative Phytochemical analysis**

Presence or absence of certain important compounds in an extract is determined by colour reactions of the compounds with specific chemicals which act as dyes. This procedure is a simple preliminary prerequisite before going for detailed phytochemical investigation. Various tests have been conducted qualitatively to find out the presence or absence of bioactive compounds.

**Tannins**

0.5g of powdered sample of each plant is boiled in 20 ml of distilled water in a test tube and Filtered, 0.1% FeCl3 is added to the filtered samples and observed for brownish green or a blue coloration which shows the presence of tannins.
Saponins
10 ml of the plant extract was mixed with 5 ml of distilled water in a test tube and shaken vigorously to obtain a stable persistent froth which indicates the presence of saponins.

Flavanoids
Few drops of 1% NH3 solution is added to the plant extract. A yellow coloration is observed if flavonoid compound is present.

Terpenoids
5 ml of plant extract was mixed with 2 ml of chloroform and 3 ml of concentrated sulphuric acid was carefully added to the mixture to form a layer. An interface with a reddish brown coloration is formed if terpenoid constituent is present.

Glycosides
5ml of plant extract treated with 2ml of glacial acetic acid and a drop of Ferric chloride and a drop of concentrated sulphuric acid was added. Green blue colour to upper layer and reddish brown colour at the junction of two layers indicates the presence of cardiac glycosides.

Phenolic compounds
The extract (500mg) was dissolved in 5 ml of distilled water. To this, few drops of neutral 5% ferric chloride solution were added. A dark green colour indicated the presence of phenolic compounds.

c. Quantitative Phytochemical analysis
Saponin determination
20g of plant sample was dispersed in 200 ml of 20% ethanol, the suspension was heated over a hot water bath for 4hr with continuous stirring at about 55°C. The mixture was filtered and the residue re-extracted with another 200 ml of 20% ethanol. The combined extracts were reduced to 40 ml over water bath at about 90°C. The concentrate was transferred in to 250 ml separating funnel and 20 ml of diethyl ether was added and shaken vigorously. The aqueous was recovered while the ether layer was discarded. The purification process was repeated. 60 ml of normal butanol extract were washed twice with 10 ml of 5% aqueous sodium chloride. The remaining solution was heated in a water bath. After evaporation the sample were dried in the oven in to a constant weight.

Determination of total phenols by spectrophotometric method
The sample was boiled with 50ml of ether for the extraction of the phenolic component for 15 min. 5ml of the extract was pipetted in to a 50 ml flask, then 10 ml of distilled water was added. 2 ml of ammonium hydroxide solution and 5 ml of concentrated amyl alcohol were also added. The samples were made up to mark and left to react for 30 min for colour development. This was measured at 505nm.

Terpenoid determination
About 2g of the plant leaf powder was weighed and soaked in 50 ml of 95% ethanol for 24hr. The extract was filtered and the filtrate extracted with petroleum ether (60-80 °C) and concentrated to dryness. The dried extract was treated as total terpenoids.

Tannin determination
The tannins were determined by Folin and Ciocalteu method. 0.1 ml of sample extract was added with 7.5 ml distilled water and added 0.5 ml of Folin phenol reagent, 1 ml of 35% sodium carbonate solution and diluted to 10 ml with distilled water. The mixture was shaken well, kept at room temperature for 30 min and absorbance was measured at 725nm. Blank was prepared with water instead of sample. A set of standard solutions of gallic acid is treated in the same manner as described earlier and read against a blank. The result of tannin are expressed in terms of gallic acid mg/g of extract.

Results and Discussion
The present study carried out on Baccaurea courallensis Muell.Arg revealed the presence of phytoconstituents. The phytochemical active compounds were qualitatively analyzed for leaves, stem, and fruits separately and the results are presented in Table 1. In the phytochemical analysis of leaf, bark and fruit extract tannins, saponins, flavonoids, terpenoids, glycosides, and phenolic compounds showed different results with the solvents used. The result of the qualitative analysis of the leaf, fruit rind and bark are presented in the table -1. Qualitative analysis of the fruit indicated the presence of tannins, saponin, terpenoids and phenolic compound. The plant extracts does not revealed the presence glycosides and flavonoids. The leaf also showed the presence of tannin, saponin, terpenoid and phenolic compound. But with the bark extract only saponin and terpenoid were present. The plant extract revealed to contain saponin were known to produce inhibitory effect on inflammation (Just M.J et al, 1998) [6]. Quantitative estimation was carried out for the phytoconstituents which showed positive result during qualitative analysis (Table -2). Methanolic extract of bark contain greater amount of saponin (83.5mg/g) than thehexane extract (46mg/g). The methanol extract of leaf contained 61mg/g saponin but in hexane extract saponin amount was less (37.5mg/g). Methanolic extract of fruit does not exhibited the presence of saponin but in hexane extract it was 54mg/g. The saponin content is an important source of detergents, surface active agents used in industrial applications and also possesses beneficial health effects (Shi J et al, 2004) [11].

Hexane extract of fruit showed greater terpenoid content (620mg/g). Terpenoids are reported to have anti-inflammatory, antiviral, antimalarial, inhibition of cholesterol synthesis and antibacterial activity (MC Garvey D.J et al, 1995; Guangyi wang et al, 2005) [8, 3]. Phenolic compounds are one of the largest and most ubiquitous group of the plant metabolites (Singh R et al, 2007) [12]. They possess biological properties such as antiapoptosis, antiaging, anticarcinogen, antinflammation, cardiovascular protection and improvement of endothelial function (Han.X, 2007) [4]. In this study phenolic content in various part of plant was studied and methanol extract of leaf showed maximum phenolic content followed by fruit extract. But phenol was absent in the hexane extract of fruit. Several studies have described the anti-oxidant properties of different parts of various medicinal plants which are rich in phenolic compounds (Brown and Evans, 1998; Krings and
Natural antioxidants mainly come from plants in the form of phenolic compounds, such as flavonoids, phenolic acids, tocopherolsetc (Ali et al., 2008) [1] and used for the treatment of degenerative diseases.

During the phytochemical study of tannin in *Baccaurea courtallensis*, hexane and methanol extract of leaf exhibited almost similar amount of tannin whereas the methanol extract of fruit contained only 4.6mg/g tannin.

Table 1: Qualitative phytochemical analysis of different parts of *Baccaurea courtallensis* Muell. Arg

<table>
<thead>
<tr>
<th>Phytochemicals</th>
<th>Bark</th>
<th>Leaf</th>
<th>Fruit Rind</th>
</tr>
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<tbody>
<tr>
<td></td>
<td>Methanol</td>
<td>Hexane</td>
<td>Methanol</td>
</tr>
<tr>
<td>Tannins</td>
<td>-</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Saponins</td>
<td>+</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Flavonoids</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Terpenoids</td>
<td>+</td>
<td>-</td>
<td>+</td>
</tr>
<tr>
<td>Glycosides</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Phenolic Compounds</td>
<td>-</td>
<td>-</td>
<td>+</td>
</tr>
</tbody>
</table>

+ positive - negative

Table 2: Quantitative estimation of phytochemicals in methanolic and hexane extract of different parts of *Baccaurea courtallensis* Muell. Arg

<table>
<thead>
<tr>
<th>Parameters mg/g extract</th>
<th>Bark</th>
<th>Leaf</th>
<th>Fruit Rind</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Methanol</td>
<td>Hexane</td>
<td>Methanol</td>
</tr>
<tr>
<td>Saponin</td>
<td>83.5</td>
<td>46</td>
<td>61</td>
</tr>
<tr>
<td>Terpenoid</td>
<td>530.6</td>
<td>-</td>
<td>506.6</td>
</tr>
<tr>
<td>Phenol</td>
<td>7.4</td>
<td>-</td>
<td>16.6</td>
</tr>
<tr>
<td>Tannin</td>
<td>-</td>
<td>-</td>
<td>20</td>
</tr>
</tbody>
</table>

References