Original Research Article

Comparative Study of Phytochemical Constituents in Flower of Wedelia trilobata, Achyranthes aspera and Chrysanthemum from Durg District of Chhattisgarh, India

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ABSTRACT

The variety of medicinal plants have been known since ages for their therapeutic and pharmacological potentials. Such plants are known to have the ability to synthesize a wide range of chemical compounds and secondary metabolites. The active biological functions and medicinal values of such plants is attributed to the presence of such bioactive compounds. In the present study, the significant phytocompounds were screened from the methanolic and chloroformic flower extracts of three important medicinal plants commonly found in Durg district of Chhattisgarh, viz., Wedelia trilobata, Achyranthes aspera and Chrysanthemum, and then comparatively analyzed. The significant secondary metabolites viz., Cardiac glycosides, steroids, Alkaloids, Flavonoids, Terpenoids, Tannins and Saponins were qualitatively analyzed from different flower extracts and for which the standard protocol was followed. The methanolic flower extracts of three of the plants were found to have the rich sources of phytoactive compounds as compared to the chloroformic flower extract. Cardiac glycosides were richly present in the flower extracts of Wedelia trilobata, and Chrysanthemum. Flavonoids, Tannins, Terpenoids and Alkaloids were present in bulk in Chrysanthemum flower extract.

Keywords
Phytochemical compounds, Cardiac glycosides, Steroids, Flavonoids, Terpenoids, Alkaloids

Introduction

Chhattisgarh is the ‘Herbal state’ of India, rich in the natural sources of plants and herbs with medicinal values. The medicinal plants have been the important part of the the Indian traditional systems of medicines and herbal drugs since age. The tremendous beneficial properties of medicinal plants to fight against free radicals and to prevent epidemics of infectious diseases, has led to the rapid and intense exploration of more and more of such plants with therapeutic and pharmacological potentials (Anil Kumar Dhiman, 2006). The properties like anti-cancerous, anti-fungal, anti-bacterial, anti-inflammatory, anti-microbial, analgesic and antioxidant ones are often attributed to the presence of a wide range of phytochemical compounds in the different extracts of the
medicinal plants. The beneficial and medicinal properties of such plants are known to result from the combinations of the secondary products present in the plants. Hence, the medicinal plants are used as the rich source of many potent and powerful drugs (Uniyal et al., 2006). The most prominent phytochemical compounds are Cardiac glycosides, Steroids, Flavonoids, Terpenoids, Alkaloids, Saponins and Tannins.

The three commonly growing medicinal plants of Chhattisgarh, selected for the present study were Wedelia trilobata, Achyranthes aspera and Chrysanthemum. Wedelia trilobata, now with a new nomenclature as Sphagneticola trilobata is very attractive, glossy, ground cover herb with small, bright yellow blooms. It is well known for its anti-leishmanial, anti-inflammatory, antioxidant and anticarcinogenic properties (Brito et al., 2006; Lin et al., 2007; Roseni et al., 2009; Govindappa et al., 2011; Haldar et al., 2011; Kataki et al., 2012). Achyranthes aspera is an indigenous medicinal plant in the form of the perennial herb with woody base and greenish white inflorescence. It is known for spermicidal activity, and also for anti-parasitic, anti-microbial, analgesic, anti-inflammatory, wound healing and antioxidant activities (Gokhale et al., 2002; Edwin et al., 2008; Sutar et al., 2008; Zafar et al., 2009; Gayathri et al., 2009; Malavili et al., 2009; Vijaya Kumar et al., 2009; Zahir et al., 2009; Shravastava et al., 2011). Chrysanthemum is another ornamental perennial flowering herb known for its white and colourful blooms. It has anti-inflammatory and cardioprotective effects, cytotoxic, antioxidant and antimutagenic activities (Miyazawa et al., 2003; Ji et al., 2004; Xie et al., 2009; Hanen et al., 2009; Ding et al., 2010).

The medicinal value of Wedelia trilobata comprises of its use in the treatment of hepatitis, in clearing off the placenta after birth, and in cure of fever, sores, cold and inflammation (Xuesong et al., 2006). Achyranthes aspera is traditionally used in treatment of asthama, cough, oedema, piles, skin eruptions, pneumonia, rheumatism, and also in treatment against bites of poisonous snakes and reptiles (Khare et al., 2007; Nadkarni et al., 2009). Chrysanthemum and its herbal infusions are used in the treatment of viral and bacterial infections, sinusitis, blood pressure, digestive and skin problems and influenza (Cheng et al., 2005; Lin et al., 2010; Kim et al., 2011; Sharma et al., 2011).

The wide range of bioactivities, and medicinal and pharmaceutical properties of the extracts of Wedelia trilobata, Achyranthes aspera and Chrysanthemum is accounted for the presence of phytochemical constituents enormously. The present study reveals the comparative analysis of the various important phytoactive compounds in the flower extracts of three of the selected medicinal plants, prepared in methanol and chloroform solvents separately.

Materials and Methods

Collection and Preparation of Flower Extract

The healthy plants of Wedelia trilobata, Achyranthes aspera and Chrysanthemum were collected from the open fields and gardens of areas of Durg. The fresh flowers were separated from the plants, and washed with clean water. Thereafter, they were shade-dried for a few days. The dried flowers of the three plants were grinded into coarse powder. The flower extract of each plant was then prepared separately, using the solvents methanol (59%) and chloroform by Soxhlet Extraction apparatus.
Methods for Phytochemical screening tests

Test for cardiac glycosides

0.5 ml of flower extract was dissolved in 2 ml Glacial acetic acid containing one drop of 1% Ferric Chloride (FeCl₃). This solution was under layered with 1 ml of Sulphuric acid (H₂SO₄). A brown ring was formed at the interface that indicated the presence of deoxy-sugar.

Test for steroids

0.5 ml of the flower extract was dissolved in 3 ml of Chloroform (CHCl₃) and this was filtered. A few drops of conc. H₂SO₄ was added to the filtrate which formed a lower layer. Appearance of reddish brown color ring showed the positive presence for Steroids.

Test for Flavonoids

5 ml of dil. NH₃ (Ammonia) solution was added to a portion of aqueous filtrate of flower extract. To this conc. H₂SO₄ was added. The appearance of yellow color indicated the presence of Flavonoids. Yellow color usually disappeared on standing.

Test for Saponins

Frothing test, the best test for the detection of Saponins was applied. 0.5 ml of flower extract was added to 5 ml of distilled water in a test-tube. The solution was shaken vigorously, and the stable persistent appearance of ‘froth’ indicated the presence of Saponins.

Test for Tannins

0.5 ml of extract was boiled in 10 ml of distilled water and then filtered. After adding a few drops of 1% FeCl₃ (Ferric Chloride), brownish green or blue black coloration confirmed the presence of Tannins.

Test for Alkaloids

a) Meyer’s test

0.5 ml of flower extract was dissolved in 5ml of 1% HCl (Hydrogen Chloride) and placed in boiling water-bath, followed by filtration. 1ml of filtrate was treated with a few drops of Meyer’s reagent. Appearance of white precipitate or turbidity indicated the presence of Alkaloids.

b) Dry extract precipitation test

4 ml Methanol and 400 ml of Glacial acetic acid, alongwith a few drops of Ammonia was added to the small quantity of dry flower extract. The precipitation indicated the presence of Alkaloids.

Test for Terpenoids

Salkowski test was applied for the detection of Terpenoids in the extract 0.5 gms of flower extract was mixed with 2 ml of CHCl₃ (Chloroform), followed by addition of 3 ml of conc. H₂SO₄ (Sulphuric acid). A reddish brown color at the interface confirmed the presence of Terpenoids.

The phytochemical screening was followed by a comparative study for the presence of different phytochemical compounds in the different flower extracts prepared in two solvents separately, i.e., Methanol and Chloroform. The flower extract of each of the three selected medicinal plants was considered for experimentation, viz., Wedelia trilobata, Achyranthes aspera and Chrysanthemum.
Result and Discussion

The phytochemical analysis of methanolic and chloroformic extracts of flowers of *Wedelia trilobata*, *Achyranthes aspera* and *Chrysanthemum* showed the presence of different phytochemical compounds and absence of some of them as well. Methanolic flower extracts of three of the plants were found to have the rich sources of phytoactive compounds as compared to the chloroformic flower extracts. Cardiac glycosides were richly present in the flower extracts of *Wedelia trilobata* and *Chrysanthemum*. Flavonoids, Tannins, Terpenoids and Alkaloids were detected in *Wedelia trilobata*, and in bulk in *Chrysanthemum* flower extracts. In methanolic flower extract of *Achyranthes aspera*, Saponins, Tannins and Alkaloids showed less or negative presence. The weak presence of Saponins was observed in methanol flower extract of *Wedelia trilobata* and *Chrysanthemum*. Chloroformic flower extract showed the absence of Flavonoids, Saponins and Tannins in the flower extract of three of the selected medicinal plants. However, Steroids and Terpenoids were present in the chloroformic flower extract of all the plants considered for the study. (Table 1)

The plant-derived phytochemical products play an important role in human health and prevention of diseases. Natural or phytochemical constituents are secondary metabolites found in plants, in the form of Phenolic acids and Flavonoids (Apak et al., 2007). These compounds react with nutrients and dietary products, scavenge the free radicals, and thus diminish the risk of dreadful diseases such as arthritis, cancer, osteoporosis, cardiac ailments and early ageing. Among the natural Phenolic compounds found in plants, the Flavonoids are the most important ones. Tannins are polyphenolic compounds, and phenolic units are also present in Alkaloids and Terpenoids.

<table>
<thead>
<tr>
<th>Extract of Plant</th>
<th>Fl. E. of <em>Wedelia trilobata</em></th>
<th>Fl. E. of <em>Achyranthes aspera</em></th>
<th>Fl. E. of <em>Chrysanthemum</em></th>
</tr>
</thead>
<tbody>
<tr>
<td>Solvents</td>
<td>Methanol</td>
<td>Chloroform</td>
<td>Methanol</td>
</tr>
<tr>
<td>Phytochemical Constituents</td>
<td>Cardiac glycosides</td>
<td>++</td>
<td>+</td>
</tr>
<tr>
<td></td>
<td>Steroids</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td></td>
<td>Flavonoids</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>Saponins</td>
<td>$\emptyset$</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>Tannins</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>Terpenoids</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td></td>
<td>Alkaloids</td>
<td>+</td>
<td>-</td>
</tr>
</tbody>
</table>

+ = Present; ++ = Present in bulk; $\emptyset$ = Present in less amount; - = Absent; Fl. E.= Flower Extract
Table 2 Comparative equations about the presence of phytochemicals qualitatively in the flower extracts of Wedelia trilobata, Achyranthes aspera and Chrysanthemum

<table>
<thead>
<tr>
<th>1.</th>
<th>Extract</th>
<th>Methanolic Fl. E. &gt; Chloroformic Fl. E.</th>
</tr>
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<tbody>
<tr>
<td>2.</td>
<td>Phytochemicals</td>
<td></td>
</tr>
<tr>
<td>a.</td>
<td>Cardiac glycosides</td>
<td><em>Wedelia trilobata</em> Fl. E &gt; <em>Chrysanthemum</em> Fl. E. &gt; <em>Achyranthes aspera</em> Fl. E.</td>
</tr>
<tr>
<td>b.</td>
<td>Flavonoids</td>
<td><em>Chrysanthemum</em> Fl. E. &gt; <em>Wedelia trilobata</em> Fl. E. &gt; <em>Achyranthes aspera</em> Fl. E.</td>
</tr>
<tr>
<td>c.</td>
<td>Tannins</td>
<td><em>Chrysanthemum</em> Fl. E. &gt; <em>Wedelia trilobata</em> Fl. E.</td>
</tr>
<tr>
<td>d.</td>
<td>Terpenoids</td>
<td><em>Chrysanthemum</em> Fl. E. &gt; <em>Wedelia trilobata</em> Fl. E. &gt; <em>Achyranthes aspera</em> Fl. E.</td>
</tr>
<tr>
<td>e.</td>
<td>Alkaloids</td>
<td><em>Chrysanthemum</em> Fl. E. &gt; <em>Wedelia trilobata</em> Fl. E. &gt; <em>Achyranthes aspera</em> Fl. E.</td>
</tr>
<tr>
<td>f.</td>
<td>Steroids</td>
<td>Present in sufficed amounts in the Fl. E. of <em>Chrysanthemum, Wedelia trilobata</em> and <em>Achyranthes aspera</em> (Methanolic &amp; Chloroformic)</td>
</tr>
<tr>
<td>g.</td>
<td>Saponins</td>
<td>Present in less amount in the Fl. E. of <em>Wedelia trilobata</em> and <em>Chrysanthemum</em> (Methanolic)</td>
</tr>
</tbody>
</table>

Fl. E. –Flower Extract

Flavonoids are usually referred to as antioxidant and function to provide protection against diseases. Flavonoids get easily absorbed into cell membrane, thereby protect the cells from the damage of free radicals. Also they show inhibitory activity against peroxidation of lecithin (Hanen et al., 2009; Ding et al., 2010). They possess diverse biological activities such as anti-viral, anti-ulcer, cytotoxic and anti-inflammatory ones (Palliwal et al., 2005; Lee et al., 2011; Ghasemzadeh et al., 2011). Terpenoids are significant in plant growth and metabolism.

Alkaloids are often toxic to human beings and many have physiological activities, hence they are widely used in medicines. Many Alkaloids are Terpenoids in nature, and function as growth regulators or as insect repellants and attractants. Steroids, Saponins and Cardiac glycosides are three of the important groups of Triterpenoids. Steroids, at one time, were mainly considered to be animal substances such as sex hormones, bile acids etc. But now their occurrence in higher plants is detected in the form of ‘phytosterols’. Saponins are of economic interest because of their occasional toxicity. The toxic property of cardiac glycosides attribute to their pharmacological activity.

Thus in the present study, the presence of certain significant phytochemical compounds helped in determining that the flowers of *Chrysanthemum* were the richest sources of bioactive compounds owing to their efficient antioxidant and medicinal properties. The flowers of *Wedelia trilobata* were also good sources of phytochemical constituents, attributing to their medicinal and pharmacological activities. Inflorescence of *Achyranthes aspera* was comparatively reported for less sources of phytochemicals. The methanolic flower extracts of *Wedelia trilobata, Achyranthes aspera* and *Chrysanthemum* showed rich presence of phytochemicals as compared to the chloroformic extracts. (Table 2)

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