Phytochemical screening of different extracts of *Iris variegata*

Ghulam Mustafa Rather & Ranjana Singh, Arif Hussain Bhat

**ABSTRACT**

Medicinal plants have been used in different systems of medicine from centuries. Among the most important medicinal plants *Iris* species have also been included in this category which belongs to family Iridiaceae. It has widerange of medicinal and pharmacological uses. Rhizomes of some *Iris* species are rich sources ofbioactive molecules, such as flavonoids, Steroids, tannins, saponins, iridal type triterpenoids, diterpenes and quinons.Isoflavonoids are the major constituents of all *Iris* species and about 50 different isoflavonoids in the form of diglucosides, triglucosides are reported in *Iris*.Consumption of isoflavonoids is reported to reduce the risk of cardiovascular disease and cancer and also have diverse biological activities like antimicrobial, estrogenic and insecticidal. Further research work is needed to explore the wide range of applications of *Iris* species, so that maximum utilization can be done for human welfare.

1. Introduction

The ancient written evidence about medicinal plants and their usage for drug formation came from Sumerian clay slab over 5000 years ago in Nagpur. For drug preparation it comprised 12 procedures of over 250 plants (Kelly 2009). The Indian holy books Vedas mention treatment with plants which are abundant in country. Various spice plants used even today originate from India (Tucakov 1971).Plants such as myrtle and incense were utilized for the treatment during rituals according to Bible and Talmud (Dimitrova 1999). In the 18th century, the work of Linnaeus was remarkable which help us in naming and classifying the plants (Jancic 2002). Early 19th century was a turning point in the knowledge and use of medicinal plants. The isolation of glycosides marked the beginning of scientific pharmacy. The discovery of active substances such as tannins, saponosids, etheric oils, vitamins, hormones etc by the up gradation of chemical methods (Dervendzi 1992). In early 20th century, stabilization methods for fresh medicinal plants were proposed and much effort was invested in the study of the condition of manufacturing and cultivations of medicinal plants (Lukic 1985, kovacev 2000). For the discovery of secondary metabolites the study of plants continues and among them family Iridiaceae plays an important role. In this family *Iris* the largest genus of family Iridiaceae (Mathew 1985, Townsend et al., 1985). Phytochemical studies on 18 Iris species all over the world have been reported to produce 46 isoflavonoid aglycones perhaps the largest number of isoflavonoids in a single genus of non-leguminous plants (Rastogi, et al., 1991). In view of the medicinal properties of various species of *Iris* and pharmacological effects of active constituents present study was carried out to determine the different phytochemical constituents like flavonoids, terpenoids, steroids etc in *Iris variegata*.

2. Material and methods:

The rhizomes of *Iris variegata* were collected from high altitudes of Gulmarg 2657 m (Latitude: 34°02’ 60.00’ N and Longitude: 74° 22’ 48.00’ E) of District Baramulla Jammu and Kashmir India, during July-August in 2017. Taxonomical identification of the plant was carried at centre for biodiversity and taxonomy Department of Botany University of Kashmir. The herbarium of the plant was deposited at department of botany, university of Kashmir under voucher No. 2603 on 04-07-2017.

The rhizome was thoroughly washed 2-3 times with running tap water, then these rhizomes were dried in shade for 3-4 weeks, every part was then cut into pieces, then the plant material (rhizomes) were grinded in grinder. The plant material was kept in a bottle and was labeled. The powered plant material was weighed using an electronic balance. The plant material was now ready for extraction by cold extraction with N-hexane and in soxhlet extractor with chloroform 95% and ethanol 5%, methanol 60% and distilled water. The extraction was done for 48 hours in each solvent. The crude extracts thus obtained were then filtered through filter paper and then concentrated in vacuum evaporator. The amount of crude extracts obtained thus weighed (Table: 1) and the yield was calculated as:

\[
\text{Percentage yield} = \frac{\text{Wt. of crude extract (gms.)}}{\text{Wt. of the powdered sample used (gms.)}} \times 100
\]

<table>
<thead>
<tr>
<th>S.No.</th>
<th>Solvent</th>
<th>Wt. of plant material (gm)</th>
<th>Vol. of solvent (ml)</th>
<th>Wt. of extract (gm)</th>
<th>Percentage %</th>
</tr>
</thead>
<tbody>
<tr>
<td>01</td>
<td>N-Hexane</td>
<td>300</td>
<td>500</td>
<td>42.85</td>
<td>14.2</td>
</tr>
<tr>
<td>02</td>
<td>Chloroform</td>
<td></td>
<td>200</td>
<td>21.05</td>
<td>7.01</td>
</tr>
<tr>
<td>03</td>
<td>Methanol</td>
<td></td>
<td>200</td>
<td>42.4</td>
<td>14.13</td>
</tr>
<tr>
<td>04</td>
<td>Distilled water</td>
<td></td>
<td>200</td>
<td>6.3</td>
<td>2.1</td>
</tr>
</tbody>
</table>

Table: 1
3. Phytochemical screening:

It is a series of tests that determine the presence or absence of chemical substances present in Iris variegata.

**Test for flavonoids** (Ammonia test):
5ml of dilute ammonia solution were added to a portion of crude extract followed by addition of concentrated sulphuric acid, formation of yellow coloration in the extract indicates the presence of flavonoids.

**Test for glycosides** (Borntrager’s Test):
To the test tubes containing 2ml of extract, 2ml of dilute sulphuric acid was added; it was boiled for 5 minutes and filtered. To the filtrates, equal volumes of chloroform was added and mixed well, organic layers were separated and ammonia was added to this. Pinkish red color of the ammonia layer indicates the presence of glycosides.

**Test for tannins** (Ferric chloride test):
The crude extract was mixed with 1% ferric chloride solution and it gives blue, green or brownish green color which indicates the presence of tannin.

**Test for saponins** (Foam test):
A small amount of extract was shaken with little quantity of water. The foam produced persists for 10 minutes. It confirms the presence of saponins.

**Test for steroids** (salkowski test):
Chloroform solution of extract was shaken with concentrated sulphuric acid and on standing yields red color, which indicates the presence of steroids.

**Test for triterpenes** (Salkowski test):
5ml of the extract was added to chloroform along with few drops of concentrated sulphuric acid. The mixture was shaken well and kept aside for some time. Appearance of red yellow color in the lower layer indicates the presence of triterpenoids.

**Test for diterpenes** (Copper acetate test):
The extract was mixed with solution of copper extract and gives green color, which indicates the presence of diterpenes.

**Test for quinones**:
With 1ml of extract 1ml of concentrated sulphuric acid was added, red color was formed which indicates the presence of quinones.

4. Results and Discussion:

The present study was carried out on phytochemical screening of rhizomes of Iris variegata revealed the presence of medicinal active constituents and the results are presented in Table 2. In the screening process flavonoids, alkaloids, glycosides, tannins, saponins, steroids, triterpenes and diterpenes shows different types of results in different chemical reagents. The medicinal value of plants lies in some chemical substances that have a definite physiological action on human body. Different phytochemicals have been found to possess a wide range of activities which helps in protection against different diseases. For example the importance of saponins and tannins in various antibiotics using in treating common pathogenic strains has recently reported by (Kubmarawa et al., 2007; Mensah et al., 2008).

According to previous studies on Iris species the phytochemicals such as steroids are also found in l.suaveolens (Hacibekiroglu and Kolak 2011) and I. germanica (Ibrahim et al., 2012). Terpenoids in rhizomes of Iris tectorum (Fang et al., 2007). Triterpenoids in I.germanica L (Bonfils et al., 2001) and in I. marsica (vanditti et al., 2017). Flavonoids in L. tenuifolia (Kojima et al., 1997), in L.songaria (Moein et al., 2008), in L. Pseudopumil (Rigano et al., 2009), in I. tenuifolia/Cui et al., 2011, Jansria et al., 2014), in L. kashmiriana (Alam et al., 2017), in I.politanii (purev et al., 2002). Glycosides are found in L.germanica L (Schutz et al., 2011), in L.marsica (vendittiet al., 2017).

<table>
<thead>
<tr>
<th>Secondary metabolites</th>
<th>Tests</th>
<th>N-Hexane</th>
<th>Chloroform</th>
<th>Methanol</th>
<th>Distilled water</th>
</tr>
</thead>
<tbody>
<tr>
<td>Alkaloids</td>
<td>Dragendrof test</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Flavonoids</td>
<td>Ammonia test</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Glycosides</td>
<td>Borntrager's Test</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Steroids</td>
<td>salkowski test</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Saponins</td>
<td>Foam test</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Tannins</td>
<td>Ferric chloride test</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Diterpenes</td>
<td>copper acetate test</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Triterpenes</td>
<td>Salkowski test</td>
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<td>+</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Quinone</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>+</td>
</tr>
</tbody>
</table>

(-) Indicates absence and (+) indicates presence of secondary metabolites.

Table 2: Phytochemical analysis of extract of Iris variegata in different solvent system.
5. Conclusion

Phytochemical screening of rhizome extract of *Iris variegata* indicates the presence of Alkaloids, Glycosides, Diterpenes, Triterpenes, Tannins, Flavonoids, Saponins and Quiones suggested that it is an important source of bioactive compounds that may supply novel medicines. Phytochemical analysis of this plant may be useful in developing new specialized drugs with more efficiency. Further optimization of these phytoconstituents through structural alteration may allow the development of pharmacologically active agents.
References


