ABSTRACT

The chemicals produced by plants called Phytochemicals. However, the term is generally used to describe chemicals from plants that may enhance health status of organisms, but are not essential nutrients. Rauwolfia serpentina is a medicinally important herb. Its roots and leaves are rich in secondary metabolites and proteins. Its medicinal properties is due to presence of alkaloids. This plant is mainly used as a source of reserpine, but now, more than 50 different alkaloids have been isolated from this plant which are used to treat hypertension, snake bite and breast cancer.\(^1\) Leucas aspera, locally known as ‘Guma’ in Chattishgarh, is a medicinal herb, which has long been used in the Indian subcontinent as a folk medicine for the treatment of a variety of diseases like an analgesic, anti-inflammatory, antioxidant, cytotoxic, snake bite, anticancer etc.\(^2\) Leucas Aspera and Rauwolphia Serpentina were studied to draw connections between their chemical properties and their use as in cancer treatment and treatments for snake bite by local people. Results of study shows that both plants have almost same chemical composition. High quantity of flavonoids, phenolic compounds and alkaloids were found on Rauwolfia serpentina and Leucas Aspera. The values of tannins were very trace on both plants.

KEYWORDS: phytochemicals, metabolite, alkaloids, antioxidant, cytotoxic, anticancer, snake bite.

INTRODUCTION

Medicinal plants are of great importance to the health of individuals and communities. The medicinal value of these plants lies in some chemical substances that produce a definite
physiological action on the human body. Rauwolfia serpentina is a medicinally important herb. Roots of this plant are used for the treatment of many diseases such as anticancer, insomnia, anxiety, excitement, schizophrenia, insanity, epilepsy, hypochondria, diarrhoea, dysentery and snake bite[1] etc in Ayurveda it is said that it stimulate uterine contractions and promote the expulsion of the fetus[2]. Leucas aspera’s water extract is used orally as stimulant, anthelmintic, laxative and diaphoretic. It is also used orally for the treatment of headache, asthma and bronchitis. Hot water extract of entire plant i.e Phanta is also used to treat inflammation, dyspepsia and jaundice. The whole plant extract is used orally to treat scabies, psoriasis and snake bite.[3] The plant is externally used as an insect repellent in the form of Dhupan.[4]

Despite the use of these plants for such purpose there is little information on the nutritional and chemical composition of Rauwolfia serpentina and Leucas Aspera. This work is therefore aimed at to be used in documenting the nutrient and chemical composition of Rauwolfia serpentina and Leucas Aspera.

MATERIALS AND METHOD

Plant Material
The medicinal plants were collected from lands of Kondagaon Amravati forest range Chattishgarh India. The plants were identified by dept. of Botany Dr. C.V. Raman University Kota Bilaspur. The roots of both plants were air-dried milled into powder with the aid of an electrical grinder and finally stored in airtight bottles for further analysis.

CHEMICAL ANALYSIS
All the elements like Ca, Mg, P, S, K, Na, Fe and Zn were determined by the method of shahidi et al.[5] 2 g of each of the plant samples were weighed and subjected to dry ashing in a well-cleaned porcelain crucible at 550°C on a muffle furnace. The resultant ash was dissolved in 5 ml of HNO₃/HCl/H₂O (1:2:3) and heated gently on a hot plate until brown fumes disappeared. To the remaining material into in each crucible, 5 ml of distil water was added and heated until a colorless solution was obtained. The samples in each crucible was transferred into 100 ml volumetric flask by filtration through a whatman No 42 filter paper and the volume was made to the mark with distil water. This solution was used for elemental analysis by atomic absorption spectrophotometer. A10 cm-long cell was used and element in the sample was calculated on percentage of dry matter. Phosphorus content of the digest was determined calorimetrically.[6] In 0.5 ml of the diluted digest, 4 ml of demineralised water, 3
ml of 0.75M H$_2$SO$_4$, 0.4 ml of 10% (NH$_4$)$_6$Mo$_7$O$_{24}$.4H$_2$O and 0.4 ml of 2%(w/v) ascorbic acid were added and mixed. The solution was allowed to stand for 20 min and absorbance was recorded at 660 nm. The content of phosphorus in the extract was determined.

**DETERMINATION OF PHYTOCHEMICALS**

**ALKALOID DETERMINATION:** 5 g of the each sample was weighed into a 250 ml beaker and 200 ml of 20% acetic acid in ethanol was added and covered to stand for 4 h. After filtration the extract was concentrated using a water bath to one-quarter of the original volume. Concentrated ammonium hydroxide was added drop wise to the extract until the precipitation was complete. The whole solution was allowed to settle and the precipitate was collected by filtration and weighed.$^{[7,8]}$

**TANNIN DETERMINATION:** 500 mg of the sample was weighed into 100 ml plastic bottle. 50 ml of distilled water was added and shaken for 1 h in a mechanical shaker. The content was filtered into a 50 ml volumetric flask and made up to the mark. Then 5 ml of the filtrate was pipette out into a tube and mixed with 3 ml of 0.1 M FeCl$_3$ in 0.1 N HCl and 0.008 M potassium ferrocyanide. The absorbance was measured in a spectrophotometer at 120 nm wavelength. A blank sample was prepared using tannin acid, the colour developed at the same wavelength.

**DETERMINATION OF TOTAL PHENOLS:** For the extraction of phenolic component, the sample was boiled with 50 ml of ether for 15 min. 5 ml of the extract was pipette into 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. The absorbance of the solution was read using a spectrophotometer at 505 nm wavelengths.$^{[7,8]}$

**FLAVONOID DETERMINATION:** 10 g of the plant samples were extracted repeatedly with 100 ml of 80% aqueous methanol at room temperature. The whole solution was filtered through Whatman filter paper no. 42. The filtrate was later transferred into a crucible and evaporated to dryness over a water bath and weighed.$^{[9]}$

**DETERMINATION OF RIBOFLAVIN:** 5 g of the sample was extracted with 100 ml of 50% ethanol solution and shaken for 1h. This was filtered into a 100 ml flask; 10 ml of the extract was pipette into 50 ml volumetric flask. 10 ml of 5% potassium permanganate and 10
ml of 30% H2O2 were added and allowed to stand over a hot water bath for about 30 min. 2 ml of 40% sodium sulphate was added. This was made up to 50 ml mark and the absorbance measured at 510 nm in a spectrophotometer.

**DETERMINATION OF THIAMIN:** 5 g of the sample were homogenized with ethanolic sodium hydroxide (50 ml). It was filtered into a 100 ml flask. 10 ml of the filtrate was pipette and the colour developed by addition of 10 ml of potassium dichromate and read at 360 nm. A blank sample was prepared and the colour also developed and read at the same wavelength.

**DETERMINATION OF Niacin:** 5 g of the sample was treated with 50 ml of 1 N sulphuric acid and shaken for 30 min. 3 drops of ammonia solution were added to the sample and filtered. 10 ml of the filtrate was pipette into a 50 ml volumetric flask and 5 ml potassium cyanide was added. This was acidified with 5 ml of 0.02 N H2SO4 and absorbance measured in the spectrophotometer at 470 nm wavelengths.

**DETERMINATION OF ASCORBIC ACID** (Vitamin C): 5 g of the sample was weighed into an extraction tube and 100 ml of EDTA/TCA (2:1) extracting solution were mixed and the mixture shaken for 30 min. This was transferred into a centrifuge tube and centrifuged at 3000 rpm for about 20 min. It was transferred into a 100 ml volumetric flask and made up to 100 ml mark with the extracting solution. 20 ml of the extract was pipette into a volumetric flask and 1% starch indicator was added. These were added and titrated with 20% CuSO4 solution to get a darkened point.[10]

**Table 1: Phytochemical composition of Rauwolfia serpentina and Leucas Aspera expressed as mg/100 g dry weight**

<table>
<thead>
<tr>
<th>Phytochemicals</th>
<th>Leucas Aspera</th>
<th>Rauwolfia serpentina</th>
</tr>
</thead>
<tbody>
<tr>
<td>Alkaloids</td>
<td>1.25 ±0.11</td>
<td>1.50 ±0.02</td>
</tr>
<tr>
<td>Flavonoids</td>
<td>1.75 ±0.20</td>
<td>1.65 ±0.12</td>
</tr>
<tr>
<td>Phenols</td>
<td>1.64 ±0.02</td>
<td>1.84 ±0.11</td>
</tr>
<tr>
<td>Tannins</td>
<td>0.55 ±0.1</td>
<td>0.85 ±0.20</td>
</tr>
</tbody>
</table>

Results are mean of triplicate determinations on a dry weight basis ±standard deviation.

**Table 2: Mineral composition of Rauwolfia serpentina and Leucas Aspera on mg/100g dry weight**

<table>
<thead>
<tr>
<th>Elements</th>
<th>Leucas Aspera</th>
<th>Rauwolfia serpentina</th>
</tr>
</thead>
<tbody>
<tr>
<td>Calcium</td>
<td>1.08 ±0.30</td>
<td>0.45 ±0.10</td>
</tr>
<tr>
<td>Magnesium</td>
<td>0.12 ±0.11</td>
<td>0.10 ±0.20</td>
</tr>
</tbody>
</table>
Table 3: Vitamin composition of Rauwolfia serpentina and on mg/100g dry weight

<table>
<thead>
<tr>
<th>Vitamin</th>
<th>Leucas Aspera</th>
<th>Rauwolfia serpentina</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ascorbic acid</td>
<td>22.50 ±0.10</td>
<td>41.04 ±0.20</td>
</tr>
<tr>
<td>Riboflavin</td>
<td>0.25±0.01</td>
<td>0.52 ±0.10</td>
</tr>
<tr>
<td>Thiamine</td>
<td>0.15 ±0.20</td>
<td>0.20 ±0.02</td>
</tr>
<tr>
<td>Niacin</td>
<td>0.07 ±0.11</td>
<td>0.05 ±0.10</td>
</tr>
</tbody>
</table>

Results are mean of triplicate determinations on a dry weight basis ± standard deviation.
the plants can play valuable roles in the management of diabetes, which result from insulin malfunction.[12]

These plants are good sources of ascorbic acids, riboflavin, thiamin and niacin (Table 3). Natural ascorbic acid is vital for the body performance.[12]

This study, therefore, has provided some biochemical basis for the ethnomedical use of these plants in the treatment and prevention of infections. As rich source of phytochemicals, minerals and vitamins Rauwolfia serpentina and Leucas Aspera can be a potential source of useful drugs and can be use as nutritional whole food supplements for gaining health benefits against oxidative stress related disorders.

REFERENCES
