Estimation of Total Iron Content of Aqueous Extract of *Lepidagathis Incurva*

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**ABSTRACT**

*Lepidagathis Incurva* is a common herb which is found in the hot Indo-Burmese and Malaysian regions in forests and thickets at low to medium altitudes. Ethnobotanical journals have reported that the plant has been used by tribals for many different uses like treatment of blindness, ear infection, hematinic, hemostatic etc. The sample specimen for the present study was collected from vaidyamadham, Mezhathur, Trithala, Palakkad, where it has been reported to be used by Ashtavaidyan Vaidyamadham Cheriya Narayanan namboodiri as hematicin. The dosage preferred was eating 10 leaves of *Lepidagathis Incurva* early morning in daily basis; good results have been reported with increase in hemoglobin content within one month. In the present study iron content estimation was done by two methods, UV estimation and atomic absorption spectroscopy. The content of Iron in the microwave digested sample was found to be 1.074mg/L by atomic absorption spectroscopy. The Iron-1,10-phenanthroline complex method was used for UV method and the Fe content was found to be 0.9mg/L. Acute toxicity studies were carried out by following the OECD guidelines. The plant showed good potential for being included in polyherbal formulaions as phytounitent.

**Keywords:** *Lepidagathis Incurva*, Hemostasis, UV estimation, absorption spectroscopy, iron estimation.

1. **INTRODUCTION**

Ethnobotanical studies have reported *Lepidagathis Incurva* being used for blindness, ear ailments, hemostatic. The herb is being used traditionally as hematinic by the tribals at the Kudajadri hill of Kollur, Shimooga district, Karnataka. The practice has been extended and implemented by the Ayurvedic Physicians at vaidyasala palakkad. The present study was aimed at estimating the iron content of the leaves which may prove to be potent phytopharmaceutical, which can be included in Ayurvedic formulations.

Formulations available iron are plenty in the market and are being widely prescribed for the pregnant and anemic patient, many of them are abhoved because of their metallic taste. *Lepidagathis Incurva* seemingly proves to be a potentially active alternative to these.

In the present study an attempt was made to find the iron content of the aqueous extract of *Lepidagathis Incurva* to check out its feasibility as a potent phytopharmaceutical.

2. **MATERIALS AND METHODS**

The *Lepidagathis Incurva* was received as a gift sample from vaidyamadham, Palakkad dist (Kerala) the plant was authenticated by Dr. Jomy Augustine, Head of the Department of Botany, St. Thomas College, Pala, Kottayam.

**ESIMATION OF IRON CONTENT:**

**Spectrophotometric determination of Fe2+ ions using 1,10-phenanthroline:**

1. Weigh accurately about 0.07 g of pure ferrous ammonium sulfate hexahydrate, dissolve it in water, and transfer the solution to a 1-liter volumetric flask. Add 2.5 mL of concentrated sulfuric acid and dilute the solution to the mark. Calculate the concentration of the solution in mg of iron per mL. (Remember, your solution was prepared using Fe (NH4)2(SO4)2•6H2O).

2. Prepare the unknown sample as follows. Add about 20g fresh leaves and approximately 100mL 10% sulfuric acid. Crush these leaves using mortar and pestil and filter. Now

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transfer a 1mL this solution to another 100 mL volumetric flask. This will be referred to as the "prepared unknown". 3. Into another five 100 mL volumetric flasks, pipette (volumetrically) 1, 5, 10, 15, 20, and 25 mL portions of the standard iron solution. Put 50 mL of distilled water into another flask to serve as the blank. To each flask, including the "prepared unknown", add 1 mL of the hydroxylamine solution, 10 mL of the 1,10 phenanthroline solution and 8 mL of the sodium acetate solution. Then dilute all the solutions to the 100 mL marks and allow them to stand for 10 minutes with occasional shaking of the flasks.

4. Using the blank as a reference and any one of the iron solutions prepared above, measure the absorbance at different wavelengths in the interval from 400 to 600 nm. (Note that it is necessary to re-adjust the 0% T and 100%T settings whenever the wavelength is changed). Take reading about 20 nm apart except in the region of maximum absorbance where intervals of 5 nm should be used. Plot the absorbance vs. wavelength and connect the points to from a smooth curve. Select the proper wavelength to use for the determination of iron with 1,10-phenanthroline.

5. Measure the absorbance of each of the standard solutions and the unknown at the selected wavelength (table1). Plot the absorbance vs. the concentration of the standards. Note whether Beer’s law is obeyed. Using the absorbance of the unknown solution calculate the % (w/w) iron in sample solution.

<table>
<thead>
<tr>
<th>Standard solution</th>
<th>Concentration of standard solution (mg/L)</th>
<th>Absorbance (nm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1 mL</td>
<td>0.013</td>
<td>0.047</td>
</tr>
<tr>
<td>5 mL</td>
<td>0.051</td>
<td>0.075</td>
</tr>
<tr>
<td>10 mL</td>
<td>0.103</td>
<td>0.188</td>
</tr>
<tr>
<td>15 mL</td>
<td>0.154</td>
<td>0.293</td>
</tr>
<tr>
<td>20 mL</td>
<td>0.206</td>
<td>0.397</td>
</tr>
<tr>
<td>25 mL</td>
<td>0.258</td>
<td>0.497</td>
</tr>
</tbody>
</table>

Table 1: Spectrophotometric determination of Fe²⁺ ions using 1,10-phenanthroline

**Total iron determination by atomic absorption spectroscopy**

**Microwave digestion**: To prepare the plant materials for total iron determination, accurately weighed samples (0.5 g) were digested with the mixture of 30% H₂O₂ and concentrated 65% HNO₃ (3:5, v/v). Next the samples were transferred to 50 mL volumetric flasks and diluted with the twice distilled water.

3. RESULT AND DISCUSSION

The given sample complies the limit test for iron as per the Indian Pharmacopoeia. spectrophotometric data shows that absorbance of sample solution is found to be 0.175 at 510 nm. So the concentration of sample from the standard graph is calculated as 0.9 mg/L (Fig 1). Atomic absorption spectroscopic method for total iron estimation of microwave digested sample is reported that one liter of extract contains 1.075mg of iron.

Fig 1: concentration vs absorbance

4. CONCLUSION

Trace elements have important role in various human metabolic processes. In the present study Atomic absorption spectroscopy has been used to determine the iron content and to carryout quantitative estimation. The experimental data of the present work will be of immense importance in formulation of new Ayurvedic proprietary medicines and managing dose of a particular formulation.

5. REFERENCES


Conflict of Interest: None Declared

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