Phytochemical investigation & evaluation for antidiabetic activity of leafy extracts of various Ocimum (Tulsi) species by alloxan induced diabetic model

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ABSTRACT

The various species of Ocimum (Lamiaceae) i.e. Ocimum gratissimum linn., Ocimum americanum linn., Ocimum sanctum linn. and Ocimum basilicum linn. are widely distributed in tribal areas of south eastern Odisha and extensively used traditionally by the tribal people for common colds, headaches, stomach disorders, inflammation, heart diseases, antidiabetic, treatment of various forms of poisoning, malaria and as anthelmintic. The present study is an attempt to preliminary investigation of phytochemical constituents and to explore the antidiabetic activity of methanolic extracts of leaves of various Ocimum species including the comparison of their activity. The Ocimum extracts were screened for phytochemical constituents and evaluated for their antidiabetic activities by alloxan induced diabetic model in Wistar rats. All the tests for phytochemical constituents of methanolic extract of Ocimum species were found to be positive except anthraquinone glycosides and thiol group. All extracts were able to show antidiabetic activity at 0.5 mg/Kg concentration. The activities are well comparable with the standard drug, glibenclamide. The methanolic extract of Ocimum sanctum linn showed better antidiabetic activity in comparison with other species of Ocimum and standard drug. The data were verified as statistically significant by using one way ANOVA at 5 % level of significance (p < 0.05).

Key words: Ocimum; Lamiaceae; Antidiabetic; Alloxan induced diabetic model; glibenclamide.

INTRODUCTION

Anti-diabetic drugs treat diabetes mellitus by lowering glucose levels in the blood. With the exceptions of insulin, exenatide, and pramlintide, all are administered orally and are thus also called oral hypoglycemic agents or oral antihyperglycaemic agents.

Ocimum gratissimum Linn. (Camphor basil in English) is found in hot and the temperate regions of India. The plant grows to a height of one to two feet. The stem and the branches are green or light yellow. The leaves are two to one inches long, oval, pointed and sharp. The seeds are small in size, black in colour, slightly elongated, round at one end and flattened at the other, with thick edges. It stimulates nerve-endings causing a tingling sensation. The leaves taste like cloves; hence they are widely used for flavoring of vegetables etc.

Ocimum americanum Linn. (Hoary basil in English) commonly is found in fields and waste lands, throughout India. A pubescent erect, much branched herb, 15-60 cm high with sub-quadrangular striate branches. Flowers whitish pink found in elongate racemes. Fruits are small; nutlets are pitted and mucilaginous when wetted.

Ocimum sanctum Linn. (Holy basil in English) is a 30-75 cm high erect herb which is grown practically in every part of India. Leaves are 2.5-5 cm. long and 1.6-3.2 cm broad, elliptical, oblong obtuse. Inflorescence is verticillate and flowers are in racemes 15-20 cm. long in close whorls. Odour and taste are aromatic and sharp.

Ocimum basilicum Linn. (Sweet basil in English) is an aromatic plant, nearly glabrous branching herb, 60-90 cm in height with hairy stems, branches green; opposite green leaves simple, opposite, ovate, acute, entire, base cuneate, glabrous on both surfaces and are strongly scented; flowers white or pale purple in simple or much branched racemes; fruits ellipsoid nutlets, black and pitted. There were no reports on the analgesic activity of the leaves extracts of Ocimum species. This prompted us to investigate the analgesic activity of leaves extracts Ocimum species.

MATERIALS AND METHODS

Materials

Sonicator, heating mantle, soxhlet extractor, stop watch, orally feeding needle,dispoven syringe, insulin syringe, digital glucometer (Jonan and Jonsib), glucose strip, and standard drug glibenclamide were supplied by the Department of Pharmacognosy, Jeypore College of Pharmacy, Rondapalli, Jeypore. Methanol (Merck Pvt. Ltd. Mumbai) and other chemicals and reagents were procured from authorized suppliers.

Plant material

The leaves of Ocimum species (O. gratissimum linn., O. americanum linn., O. Sanctum linn. and O. basilicum linn.) were collected from the local area of south eastern Odisha (India) in the month of January 2010. The plant material were identified and authenticated by Botanical Survey of India, Shibpur, Howrah vide Letter No. CNH/I/49/2009/Tech.II/168 dated 05.03.2010. The collected leaves were shade dried under normal environmental condition, powdered, stored in a closed container for further use.

Preparation of Extract

The powdered leaf of each Ocimum species (750 g) was extracted with methanol by using Soxhlet extraction apparatus. The extract was filtered and concentrated in a closed container for further use.

Phytochemical screening

Chemical tests were carried out on all Ocimum species extracts for the qualitative determination of phytochemical constituents as per the standard procedure.

Animals care and handling

This was done as per the guidelines set by the Indian National Science Academy New Delhi, India. Twelve-week-old healthy Wistar rats (150-180g) of either sex bred locally in the animal house of Jeypore College of Pharmacy, Jeypore were selected for the study. They were housed under controlled conditions of temperature of 23 + 2°C, humidity of 50 ± 5% and 10–14 h of light and dark cycles respectively. The animals were housed individually in polypropylene cages containing sterile paddy husk (procured locally) as bedding throughout the experiment and had free access to sterile food (animal chow) (M/s Hindustan Lever Ltd.) and water ad libitum. Animals were caged and all operations on animals were done in aseptic condition. Approval for the research work was obtained by the Institutional Ethical Committee of regd. No. HPI/07/60/IAEC/00013 of date. 07-05-2008.

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Table 1. Phytochemicals detected in methanolic extracts of various Ocimum species.

<table>
<thead>
<tr>
<th>Phytochemicals</th>
<th>O. gratissimum</th>
<th>O. americanum</th>
<th>O. sanctum</th>
<th>O. basilicum</th>
</tr>
</thead>
<tbody>
<tr>
<td>Alkaloids</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Cardiac glycoside</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Anthraquinone glycosides</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Gums mucilage</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Phenolic compound</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Sterols</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Steroids</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Saponins</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Tannins</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Flavones</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Flavanoids</td>
<td>+</td>
<td>+</td>
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<td>+</td>
</tr>
<tr>
<td>Thiod group</td>
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</tr>
</tbody>
</table>

+ = Present and - = Absent

Table 2. Analgesic activity of leafy extracts of various Ocimum species.

<table>
<thead>
<tr>
<th>Group</th>
<th>Treatment (Dose (mg/Kg))</th>
<th>Blood glucose (mg/dl)</th>
<th>Mean ± SEM</th>
</tr>
</thead>
<tbody>
<tr>
<td>I</td>
<td>Control (Distilled water)</td>
<td>76.16±1.99</td>
<td>81.16±2.23</td>
</tr>
<tr>
<td>II</td>
<td>Standard (Gibberellic acid) 0.5(mL)</td>
<td>327.50±6.88</td>
<td>247.33±15.5</td>
</tr>
<tr>
<td>III</td>
<td>O. gratissimum 250</td>
<td>327.33±4.91</td>
<td>272.17±9.64</td>
</tr>
<tr>
<td>IV</td>
<td>O. americanum 250</td>
<td>320.00±6.27</td>
<td>291.50±10.7</td>
</tr>
<tr>
<td>V</td>
<td>O. sanctum 250</td>
<td>322.16±4.82</td>
<td>270.83±16.5</td>
</tr>
<tr>
<td>VI</td>
<td>O. basilicum 250</td>
<td>320.00±6.27</td>
<td>290.83±13.6</td>
</tr>
</tbody>
</table>

All values are expressed in mean ± standard deviation (n=6).
All data were found to be significant at 5% level of significance where p<0.05.

CONCLUSION

Antidiabetic effect against hereditary, immunological, age stress may be elicited through insulin receptors in relevant phenomena. But the extend of activity shown by the crude extracts are less than that of the standard drug glibenclamide except the methanolic extract of Ocimum sanctum linn but many fold more than that of the control group, which justifies its activity. It could be concluded that the Ocimum plant is having anti diabetic activity and better result are obtaining from the methanolic extract of Ocimum sanctum linn. This further study needed to identify the chemical constituents present in extract of this herb that may elicit antidiabetic activity.

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