EVALUATION OF ANTIANXIETY AND ANTIDEPRESSANT ACTIVITY OF CASSIA OCCIDENTALIS LEAVES

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ABSTRACT

Anxiety and Depression are widespread psychiatric disorders affecting around 5% of the population. Furthermore, it is difficult to predict which patient will respond to any given treatment. In the traditional systems of medicine, many plants and formulations have been used to treat anxiety and depression for thousands of years. The present study was designed to evaluate the antianxiety and antidepressant activity of the ethanolic and aqueous extracts of Cassia occidentalis leaves in rodents. Antianxiety activity was tested by exposing rats to unfamiliar aversion in different methods like elevated plus maze model and actophotometer. The results infer that reduced aversion fear elicits antianxiety activity. The antidepressant activity was tested by using despair swim test and tail suspension test. The results infer that reduced immobility time elicits antidepressant activity. It was concluded that ethanolic and aqueous extracts of Cassia occidentalis leaves having antianxiety and antidepressant activity. Ethanolic extract of Cassia occidentalis leaves showing more significance activity over the aqueous extract.

Keywords: Cassia occidentalis, Antianxiety activity, Antidepressant activity, Elevated plus maze, Actophotometer, Despair swim test.

INTRODUCTION

According to the World Health report 1, approximately 450 million people suffer from mental disorders or heart diseases, yet only a small minority of them receives even the most basic treatment. This amounts to 12.3% of the global burden of disease and will rise to 15% by 2020 2. In the search for new therapeutic products for the treatment of neurological disorders, medicinal plant research, worldwide has progressed constantly demonstrating the pharmacological effectiveness of different plant species in a variety of animal models 3. Anxiety and depression are extremely dramatic and debilitating multifacetic disorders and it is now becoming clear that without knowledge of clinical and biological aspects of anxiety and depression, it is impossible to offer effective treatment strategies for the patients. Over the past decades, there has been intensive study of a variety of neurobiological aspects of depression and anxiety. Currently the most widely prescribed medications for anxiety disorders are benzodiazepines. But the clinical applications of benzodiazepines as antianxiety agents are limited by their unwanted side effects. Therefore the development of new pharmacological agents from plant sources are well justified. Cassia occidentalis a native plant of southern India, called as Kasmard in Sanskrit, Kasondi in Hindi and Coffee senna in English belongs to family Caesalpiniaceae. Its common name is ponnavarai. The parts used are leaves, seeds and roots. It is used for fever, menstrual problems, tuberculosis and as a tonic for general weakness and illness 4. Cassia occidentalis leaf extracts have anti bacterial 5,6,7, antiplasmodial 8, antimutagenic 9, hepatoprotective10 and anti diabetic activity11. The purpose of the present study was to evaluate the antianxiety and antidepressant activity of ethanolic and aqueous extracts of Cassia occidentalis leaves.

MATERIALS AND METHODS

Plant material

Collection and authentication of plant materials

The leaves of Cassia occidentalis belonging to the family Caesalpiniaceae were collected in the month of april 2011 from the local areas of Anantapur district, Andhra Pradesh, India. The plant material was identified and authenticated by Dr. J. Raveendra Reddy, M.Pharm., PhD, Department of Pharmacognosy, Raghavendra Institute of Pharmaceutical Education and Research, Anantapur and voucher specimen (13/11) was preserved in Department of Pharmacology, Raghavendra Institute of Pharmaceutical Education and Research, Anantapur, India.

Processing of sample leaves were dried in shade for 25 days and then powdered to get a coarse powder. This powder was stored in an air tight container and used for successive extraction.
Drugs and chemicals
Fluoxetine (Crescent Therapeutics limited, Himachal Pradesh), Diazepam (Ranbaxy Laboratory limited.), Ethanol (Loba Chemicals Mumbai).

Antianxiety and antidepressant activity
The ethanolic and aqueous extracts of Cassia occidentalis leaves were tested for antianxiety activity using elevated plus maze and actophotometer and antidepressant activity using despair swim test and tail suspension test.

Treatment
Animals were divided into four (I-IV) groups.

Group I - Control group received distilled water (1ml, p.o).
Group II - Standard group received Diazepam (5mg/kg i.p).
Group III - Test group received ethanolic extract of Cassia occidentalis (500mg/kg p.o).
Group IV - Test group received aqueous extract of Cassia occidentalis (500mg/kg p.o).

ANTIANXIETY ACTIVITY
Elevated plus maze (EPM) model
The apparatus comprises of two open arms (35x5cm) and two closed arms (30x5x15cm) that extend from a common central platform (5x5cm). The floor and walls of the closed arms are made of wood and painted black. The entire maze is elevated to a height of 50 cm above the ground level. Rats weighing (150 – 200gms) were housed in a pair of 10 days prior to the test in the apparatus. During this time the rats were handled by the investigator on alternate days to reduce stress. 30 min and 60min after oral administration of the drug treatment, each rat was placed in the center of the maze facing one of the enclosed arms. During five minutes session, number of entries into open arm and time spent in the open arm were noted. The procedure was conducted preferably in a sound attenuated environment.

Locomotor activity
The locomotor activity can be easily studied with the help of actophotometer, the rats were grouped and treated with drugs. Each animal was placed individually in actophotometer and the basal activity score of all the animals were recorded for 10 mins after 30 and 60 min of drug treatment.

ANTIDEPRESSANT ACTIVITY
Despair Swim Test Apparatus
For the determination of antidepressant activity, forced swim test (FST) protocol was employed. During the test, animals were individually placed in a glass cylinder (20 cm in height, 14 cm in diameter) filled with water up to a height of 10cm, at 25 ± 2°C. All animals were forced to swim for 5 min and the duration of immobility was observed and measured during the 5 min interval of the test. Immobility period was regarded as the time spent by the rats to float in water with no struggle and making only those movements necessary to keep its head above the water. In order to check the fitness level of each test animal, a pre-test was carried out 24 h before the FST by subjecting each test animal to a session of 15 min swimming.

Tail suspension test
Tail suspension test was performed based on the method prescribed. The mice were suspended 58cm above the floor by means of an adhesive tape, placed approximately 1cm from the tip of the tail. The total duration of immobility was quantified during a test period of 5min. Mice were considered immobile when they were completely remain motionless.

RESULTS
Physical properties of the extracts
The colour, texture and the percentage yield of the ethanolic and aqueous extracts of leaves of cassia occidentalis were tabulated in table 1.

Acute toxicity
The acute toxicity study revealed the non toxic nature of all the extracts even at a higher dose of 4g/kg body weight of mice for oral route of administration. For the present study the dose is being selected as 500mg/kg p.o.

Phytochemical analysis
After subjecting to screening, both the ethanolic and aqueous extracts of leaves of cassia occidentalis revealed the presence of flavonoids, glycosides, tannins and saponins. The details of phytochemical constituents are given in table 2.

Table 1: Physical properties of Cassia occidentalis leaves extracts.

<table>
<thead>
<tr>
<th>Plant part</th>
<th>Type of extract</th>
<th>% Yield</th>
<th>Texture</th>
<th>Colour</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cassia occidentalis leaves</td>
<td>Ethanolic extract</td>
<td>13.8</td>
<td>Gummy</td>
<td>Reddish brown</td>
</tr>
<tr>
<td>Cassia occidentalis leaves</td>
<td>Aqueous extract</td>
<td>11.3</td>
<td>Gummy</td>
<td>Greenish</td>
</tr>
</tbody>
</table>

Table 2: Phytochemical analysis of Cassia occidentalis leaves extracts

<table>
<thead>
<tr>
<th>Phytochemicals</th>
<th>Ethanolic extract</th>
<th>Aqueous extract</th>
</tr>
</thead>
<tbody>
<tr>
<td>Flavonoids</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Glycosides</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Saponins</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Tannins</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Alkaloids</td>
<td>-</td>
<td>-</td>
</tr>
</tbody>
</table>

+ indicates presence; – indicates absence of the phytochemical constituents which were screened using various identification tests.

Table 3: Effect of Ethanolic and Aqueous Extracts of Cassia Occidentalis Leaves on Locomotor Activity (Actophotometer) In Rats at Different Time Intervals.

<table>
<thead>
<tr>
<th>Group</th>
<th>Treatment</th>
<th>Photo cell count (Count/600Sec)</th>
<th>% change in activity</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>30min</td>
<td>60min</td>
</tr>
<tr>
<td>I</td>
<td>CONTROL (VEHICLE)</td>
<td>306±5.032</td>
<td>307.7±4.842</td>
</tr>
<tr>
<td>II</td>
<td>Diazepam (5mg/kg i.p)</td>
<td>119±1.528</td>
<td>84.6±2.690</td>
</tr>
<tr>
<td>III</td>
<td>Ethanolic extract (500mg/kg p.o)</td>
<td>145.7±4.910</td>
<td>97.3±1.856</td>
</tr>
<tr>
<td>IV</td>
<td>Aqueous extract (500mg/kg p.o)</td>
<td>167.3±2.404</td>
<td>118.3±1.453</td>
</tr>
</tbody>
</table>

NA: Not Applicable, *P<0.001 when compared to Control

Assessment of antianxiety activity

Elevated plus-maze model
In elevated plus-maze test (EPM), the ethanolic and aqueous extracts of cassia occidentalis leaves at a dose of 500 mg/kg p.o. significantly increased the number of entries and time spent into the open arm. The magnitude of the antianxiety effects 500mg/kg p.o. of ethanolic and aqueous extracts of cassia occidentalis was comparable to that of diazepam 5 mg/kg i.p. (Figure 1 and 2).
A significant decrease in the immobility time directed towards the reduced immobility time by rodents. By performing tail suspension test, the reduced immobility time is thought to reflect either a failure of persistence in escape-directed behavior [i.e., despair behavior] or the development of a passive behavior, meaning the loss of the animal’s ability to cope with stressful stimuli. Markedly showed a significant decrease in the time spent immobile by rodents. By performing tail suspension test, the reduced immobility time directed the antidepressant effect. The motor activity, which is measured by the time spent by the animals is exhibited by decrease in the motor activity and preference to remain at safer places. Antianxiety agents are expected to increase the motor activity, which is measured by the time spent by the animal in the open arms. The spontaneous decrease in basal activity score implicates the reduced anxiety recorded using actophotometer. The widespread use of PST is mainly due to its ability to detect a broad spectrum of antidepressant agents. The test is based on the observation that rodents following initial escape-oriented movements develop an immobile posture when placed inside an inescapable cylinder filled with water. The immobility is thought to reflect either a failure of persistence in escape-directed behavior [i.e., despair behavior] or the development of a passive behavior, meaning the loss of the animal’s ability to cope with stressful stimuli. Markedly showed a significant decrease in the time spent immobile by rodents. By performing tail suspension test, the reduced immobility time directed the antidepressant effect. The antianxiety and antidepressant effects may be due to the flavonoid content present in both the extracts of Cassia occidentalis. However, further studies are required to identify the phytoconstituents responsible for the observed antianxiety and anti-depressant effect.

**DISCUSSION**

The fear due to height induces anxiety in the animals when placed on the EPM. The ultimate manifestation of anxiety and fear in the animals is exhibited by decrease in the motor activity and preference to remain at safer places. Antianxiety agents are expected to increase the motor activity, which is measured by the time spent by the animal in the open arms. The spontaneous decrease in basal activity score implicates the reduced anxiety recorded using actophotometer. The widespread use of PST is mainly due to its ability to detect a broad spectrum of antidepressant agents. The test is based on the observation that rodents following initial escape-oriented movements develop an immobile posture when placed inside an inescapable cylinder filled with water. The immobility is thought to reflect either a failure of persistence in escape-directed behavior [i.e., despair behavior] or the development of a passive behavior, meaning the loss of the animal’s ability to cope with stressful stimuli. Markedly showed a significant decrease in the time spent immobile by rodents. By performing tail suspension test, the reduced immobility time directed the antidepressant effect. The antianxiety and antidepressant effects may be due to the flavonoid content present in both the extracts of Cassia occidentalis. However, further studies are required to identify the phytoconstituents responsible for the observed antianxiety and anti-depressant effect.

**CONCLUSION**

From the results it was concluded that both ethanolic and aqueous extracts of leaves of Cassia occidentalis showed antianxiety and antidepressant activity. These findings suggest that the ethanolic extract of Cassia occidentalis leaves possess more significant antianxiety and anti-depressant activity compared to aqueous extract.

**Statistical analysis**

The results were expressed as mean ± S.E.M. The differences were compared using one way analysis of variance (ANOVA) and subsequently followed by Bonferroni’s test.

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REFERENCES