

Bombax ceiba: A Potential Anti-Anxiety Drug

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ABSTRACT

Background: *Bombax ceiba* is a famous plant used extensively in traditional medicine for various diseases. However, data pertaining to its effects at CNS level is limited. **Objective:** To analyze the anti-anxiety potential of ethanolic extracts of *Bombax ceiba* leaves on rats using the elevated plus maze protocol. **Materials and Methods:** Five groups containing six rats each were treated with respective drugs. Control rats were treated with 0.5ml of 10% ethanol i.p. (negative control). Single treatments of diazepam (2mg/kg i.p) were used as a positive control and varied levels of ethanolic extracts of *Bombax ceiba* leaves (150 mg/kg, 250 mg/kg, 400 mg/kg, i.p.) were applied to other groups. The index of anxiety was measured by observing the entry and time duration in the open arm of the EPM. **Results:** Extract at 400 mg/kg significantly increased the time duration and number of entries into the open arm which was statistically significant (p value <0.05) when compared to the control group (ethanol). Diazepam showed the most significant results when compared to other groups (p<0.0005). Extracts at 150 mg/kg and 250 mg/kg demonstrated negligible results that were statistically insignificant. **Conclusion:** The results indicate the anti-anxiety potential of *Bombax ceiba* leaves at higher concentration. Further studies are required to analyze the implicated phytochemicals and the mechanism at cellular level.

Key words: Anti-anxiety, *Bombax ceiba* leaves, Diazepam, Ethanol, Elevated plus maze.

INTRODUCTION

Psychological diseases are very common nowadays, with an increasing incidence.¹ A 2009 WHO survey indicated that mood disorders and anxiety were the most common mental health problems around the world.¹ Epidemiological studies have shown that one third of the population is affected with anxiety disorders at least once in their lifetime. These disorders may create diminishment of life and a financial burden along with health care costs.

Anxiety is usually treated with the sedative hypnotic group of drugs. The benzodiazepine group of drugs are generally prescribed but carry the burden of low safety margin and dependency.^{2,3} Of the newer groups, partial agonist at serotonin receptors have gained wide acceptance. These drugs, however, take up to 4 weeks to show effect and are quite costly as well.⁴ Some patients are generally resistant to these drugs and have limited response to treatment.

On the other hand, traditional healing with plants and their extracts has been used for treating mental disorders since many years. Some famous plants are *Hypericum perforatum* and *Nigella sativa* for depression.^{5,6} *Passiflora* and *Chamomile* for anxiety disorders.^{7,8} The effects are due to certain phytochemical constituents in these plants.

Bombax ceiba is a tree from the Malvaceae family which grows in Indonesia, Malaysia, China, Hong Kong and Taiwan. The tree has red blossoms with five petals. Tibetans refer to it as “salmari”, while in Europe and America, it is ordinarily referred as the cotton tree. It is utilized as a tea in parts of China.

This tree is rich in various phytochemicals. Extracts have confirmed the presence of alkaloids, flavonoids, glycosides, coumarins, proteins and amino acids.⁹ *Bombax ceiba* has numerous documented effects, some of which are anti-inflammatory, aphrodisiac, antimicrobial, hepatoprotective, anti-diabetic, anti-aging and hypotensive.¹⁰⁻¹⁶ Evaluation at the CNS level, however, is limited. Therefore our study is aimed at analyzing the anti-anxiety potential of this plant analyzing the anti-anxiety potential of this plant.

MATERIALS AND METHODS

Plant material

Bombax ceiba leaves were washed with water and shade dried. After crushing into a powder, 100 gm was placed in 500 ml of 70% ethanol in a dark place for three days with frequent agitation. The obtained extract was then filtered and dried with a rotary evaporator.

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Animals and grouping

The experiments were performed according to university guidelines as per the research committee and ethical committee approval. The subjects consisted of 30 albino rats, 125-150 gm in weight and of both genders. These were obtained from the animal house at the College of Pharmacy, King Khalid University, Saudi Arabia. The animals were allowed free access to water and food *ad libitum*. They were kept in normal temperatures with a 12-h light dark cycle. The animal experiments were carried out as per the National Institutes of Health guide for the care and use of Laboratory animals.

Five groups of six rats each were formed and treated with their respective test drugs, as follows:

- Group I: Positive control, Diazepam 2mg/kg, n=6
- Group II: Vehicle control, *Ethanol 10% (0.5 ml), n=6
- Group III: *Bombax ceiba* extract, 150mg/kg, n=6
- Group IV: *Bombax ceiba* extract, 250mg/kg, n=6
- Group V: *Bombax ceiba* extract, 400 mg/kg, n=6

*(Absolute ethanol, HolyLandPharma, Makkah, Saudi Arabia) All extracts were diluted with 0.5 ml of ethanol each for application.

Elevated Plus Maze

The elevated plus maze (EPM) is a plus shaped apparatus which has been used in neurobiological research. It has two open arms and two closed arms placed in a plus shape at a height of 50 cm. Rats prefer to stay in closed areas when in anxiety, a reduction in anxiety is observed by the entry of the animal into the open arm of the maze.¹⁷ Behavioral responses in the elevated plus maze are easily assessed and quantified by an observer. Briefly, rodents are placed in the center intersection of the four arms of the elevated plus maze and their behavior is typically recorded for 5 min.¹⁸

Experimental Protocol

Dried extracts were diluted with 10% ethanol, 0.5 ml each.¹⁹ Each animal was subjected to its respective drug through intra-peritoneal injection. Each animal was placed individually in the center of the maze and observed for 5 min. The number of entries and the time duration spent in the open arm were noted during this period. When the animal placed all four paws in the open arm, it was calculated as one entry. The observations were noted for each animal and the results were analyzed.

Statistical analysis

The mean of each group was calculated and compared through graphs for the two parameters, time spent in the open arm and duration spent in the open arm of the EPM.

The P value was calculated by one-way analysis of variance ANOVA followed by a Dunnett test. The P values <0.05 were considered statistically significant when compared to the vehicle control.

RESULTS

In the present study, we used the elevated plus maze to analyze the anti-anxiety effects of ethanolic extract of *Bombax ceiba* leaves. The EPM model uses the rat's natural fear and aversion of open and elevated places to determine anxiety levels. The number of entries into the open arm of the EPM and the time spent in it is used as an index of the anti-anxiety effect.

The time spent in the open arm of EPM was calculated over 5 min for each animal. Rats tend to stay in closed spaces when feeling fear or apprehension, therefore movement into the open arm of EPM depicts the animals less anxious state. Diazepam treated animals exhibited the least anxiety and preferred to stay and enter in the open arm of the EPM (p value <0.0005 when compared to vehicle control). Animals treated with extracts at 250 mg/kg and 400 mg/kg showed increased time spent in the open arm of the EPM (Figure 1). For the 400 mg extract time spent in open arm when compared to vehicle control (ethanol) yielded p values of < 0.05 which was statistically significant (Figure 1). The values were not significant for animals treated with 250 mg/kg and 150 mg/kg of extract.

The number of entries of each rat into the open arm was also calculated over 5 min. The reading was noted as one when the whole paw of the animal entered the open arm. Entry into the open arm is depicted as an anti-anxiety behavior of the animal. Again, diazepam treated animals exhibited the highest anti-anxiety effect due to increased entry into the open arm of the EPM (p value <0.0005 when compared to vehicle control) (Figure 2). The results were also statistically significant for the 400 mg/kg extract (p value <0.05 when compared to vehicle control), while less anti-anxiety effects were observed for the 150 mg/kg and 250 mg/kg extract with statistically insignificant readings (Figure 2).

DISCUSSION

Our study has confirmed the anti-anxiety potential of ethanolic extract of *Bombax ceiba* leaves. The elevated plus maze was used in this experi-

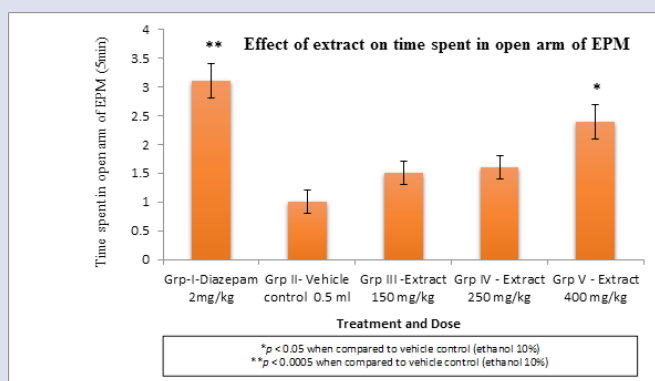


Figure 1: Effects of *Bombax ceiba* ethanolic extract on rats in elevated plus maze. Results are expressed as mean (n=6) with +/- SEM (standard error of mean). The following parameter is shown: time spent in open arm of EPM.

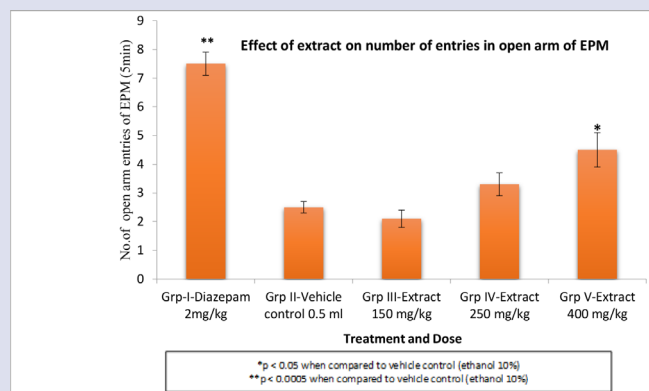


Figure 2: Effects of *Bombax ceiba* ethanolic extract on rats in elevated plus maze. Results are expressed as mean (n=6) with +/- SEM (standard error of mean). The following parameter is shown: number of entries in open arm of EPM.

ment. The elevated plus maze is a widely used behavioural assay for anxiety behaviour of rodents. It is easy to use and yields valid results in a short 5-min test period. The patterns of results obtained using this task are replicable across other species, anxiety/affective behaviour measures, studies and laboratories.

Bombax ceiba is a common traditional medicine plant, which is a rich source of many phytochemicals, including triterpenes/ triterpenoids, flavonoids, anthocyanin, shaminicin, vicenin, scopolin, mangifein and vitexin.²⁰ Lately, two new phytochemicals have been derived, simalin A and B.²¹

The phytochemicals like flavonoids, triterpenoids and lupeol in this plant may be the cause of its anti-anxiety effects. Various plants with similar phytochemicals have shown anti-anxiety effects. Triterpenoids have documented effects at central nervous level, particularly anti-anxiety potential.²² One of the triterpenoids, lupeol, has been proven to provide anxiety-reducing effects.²² Flavonoids have calming effects as well. Apigenin, a flavonoid constituent of the plant *Matricaria chamomile*, acts as a ligand for benzodiazepine receptors in the CNS, causing sedative hypnotic effects.²³ Our plant exhibits phytochemicals such as flavonoids and triterpenoids, which could be reason for CNS calming effects.

There is extensive data on the use of herbs and plants for sedative actions. A famous herb, *Valerian* is extensively used for its anti-anxiety and sleep-enhancing properties. In rat brainstem experiments; the effects of this herb were shown to be mediated by actions at the central GABA receptors.²⁴ Kava is a drink derived from the *Piper methysticum* plant. It is used extensively for its CNS calming effects. Extracts from this plant have been shown to bind to central receptors linked to GABA, dopamine and opiates.^{25,26}

Our study has provided yet another addition to these alternative therapies for anxiety. The implicated phytochemicals and the mechanism at the cellular level have to be determined. Also the toxicological screen and effective dose have to be evaluated. The conventional drugs in market have various adverse effects, mainly addiction and tolerance, this plant can provide an alternative and safer therapy for anxiety.

CONCLUSION

This study confirms the anti-anxiety potential of *Bombax ceiba* leaves. The results have indicated an anxiety lowering effect using a 400 mg/kg extract when administered through the intra-peritoneal route. This provides us with a potential new alternative therapy for treating anxiety disorders.

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ABBREVIATIONS

EPM: Elevated Plus Maze; **CNS:** Central Nervous System; **GABA:** Gaba ammino butyric acid.

CONFLICT OF INTEREST

There are no conflicts of interest.

SUMMARY

Bombax ceiba is a famous plant used in traditional medicine. It is rich in phytochemicals and has various uses. Studies at CNS level are lacking, therefore this research was carried out to evaluate any anti-anxiety potential of its leaves. Ethanolic extract at 400 mg/kg demonstrated anti-anxiety effects in animals by increasing time duration and entry in open arm of EPM. This was statistically significant ($p < 0.05$) when compared

to vehicle control. Our study therefore proposes a potential new addition in treating anxiety disorder.

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