Evaluation of Herbal Extract of *Gentiana diffusa* for Antianxiety Activity

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Abstract

The present study was designed to authenticate the anti-anxiety activity (by using elevated plus maze model) of methanolic extract of the leaves of *Gentiana diffusa.* by authors Swiss Albino mice were treated with different doses of the leaf extracts (200 mg / kg p.o.) and Diazepam (2mg/ kg, p.o) was used as a positive control. Results of study show that methanolic extract in higher doses (200 mg/kg) possesses marked anti-anxiety activity and was comparable to the effect produced by diazepam. The plant can be developed as a commercial source of anxiolytic agent. Further studies are in process to isolate the active constituent responsible for this activity and mechanism of action.
Keywords: Leaves, Methanol, Anxiety, Diazepam

Introduction

Herbal medicines are prevalent as remedies for diseases and play a key role in the human health-care of a massive common of world's population. World's populations rely on the use of traditional medicine, which is predominantly based on plant material. [1] The traditional medicine refers to a broad range of ancient natural health care practices and these medicinal practices have originated from time immemorial and developed gradually, to a large extent, by relying or based on practical experiences, without significant references to modern scientific principles. Although, herbal medicines are effective in the treatment of various ailments, very often these drugs are unscientifically exploited or improperly used. Therefore, these plant drugs justify detailed studies in the light of modern science. [2] Plant and plant products have been used for treating various illnesses. Today, several medicinal plants and their products are still being employed as house remedies, over the counter drugs as well as raw materials for the pharmaceutical industry and they represent a substantial proportion of the global drug market.

Anxiety is a merged progressive behavioral and physiological adjustment of the organism, which ultimately leads to wide variety of central nervous system (CNS) disorders, if remain untreated. In addition to individual genetic factors external influences, such as nutrition, smoking, alcohol, socioeconomic status, environmental conditions etc., can strongly contribute to its predicted arrival. [3] In Indian system of medicinal plants, many plants have been prescribed for the treatment of CNS

disorder. *Gentiana diffusa* is one such plant reported for the treatment of CNS disorders. Literature survey revealed that plants having flavonoids and terpenoids, alkaloids and tannins showed potent activity for the treatment and prevention of stress and anxiety. Thus, it was hypothesized that *Gentiana diffusa* should also showed proposed activity.

Experimental work

Collection & Identification of Plant material

The crude drug of *Gentiana diffusa* was collected from available sources. The plant part (leaves) will be shade-dried, powdered to 40 mesh particle size and stored in air tight container. Plant specimens were identified and authenticated. The powder drug of whole plant was dried in shade & used for extraction.

Extraction of plant drug

The powdered plant material (about 90 gm) was extracted with methanol (95%) in a soxhlet apparatus. The solvent was removed under reduced pressure, which obtained a dark brown sticky residue with respect to dried plant material. The dried extract was stored in a desicator till further study. The percentage yield of methanolic extract was calculated as 5.0% w/w.

In vivo study for the assessment of anti-anxiety activity of plant drug

Animals

Albino rats of approximately 8-10 weeks of age weighing 40 to 60g were taken. The institutional animal ethical committee permitted the study. The animals were housed in standard cages, at room temperature (25 ± 3ºC), with 12hr light cycles, all the animals were fed with standard pellet diet. They were given 2 weeks' time to get acclimatized with laboratory conditions.

Grouping of animals

Group I: Received Vehicle (Control)

Group II: Standard group received Diazepam (2mg/kg)

Group III: Test group received methanolic extract of *Gentiana diffusa* (200 mg/kg)

Elevated plus maze (EPM) model of anxiety

The plus-maze apparatus consisting of two open arms (16 × 5 cm) and two closed arms (16 × 5 × 12 cm) having an open roof, with the plus-maze elevated (25 cm) from the floor was used to observe anxiolytic behaviour in animals. Each mouse was placed at the centre of the elevated plus maze with its head facing the open arms. During this 5 minutes experiment, the behavior of the mouse was recorded as: (a) the number of entries into the open arms, (b) average time spent by the mouse in the open arms (average time = total time spent in open arms/number of entries in arms). Extract of *Gentiana diffusa* was administered orally using a tuberculin syringe fitted with oral canula. The dose administration schedule was so adjusted that each mouse was having its turn on the elevated plus-maze apparatus 45 minutes after the administration of the dose. During the entire experiment, the animals were allowed to socialize. [4,5,6]

Statistical Analysis

Experimental data are expressed as mean ± standard error of mean (SEM). Statistical analysis was performed using Dunnet vs. control test followed by one-way ANOVA to identify the differences between pair of groups. Data were considered significant at $p < 0.01$.

Results and Discussion

In vivo study for the assessment of anti-anxiety activity of plant drug

For the assessment of anti-anxiety activity of *Gentiana diffusa,* Number of entries in open arms and Average time spent in open arms were assessed. The results were recorded and calculated.

Table 1 Evaluation of anti-anxiety activity of plant drug

Treatments	Dose	Number of entries in open Arms Mean ± S.	Average time spent in open arms Mean ± S.
Control	Vehicle	2.15 ± 0.15*	2.30 ± 0.40**
Standard	Diazepam (2mg/kg)	5.10 ± 0.28**	10.70 ± 0.54**
Methanol extract	(400 mg/kg)	4.50 ± 0.20*	7.30 ± 0.18**

The data is expressed as Mean ± S.D.; *$P<0.05$, **$P<0.01$

Conclusion

The conclusion of the present study suggest that it can serve as a valuable source of information and provide appropriate standards to ascertain the quality of this plant material in future study or application, moreover it could be a possible source therapeutic agents in preventing or slowing the development of blood and related compounds. The flavonoids of the plant appear most likely as the active ingredients responsible for the antianxiety effect of plant.

Acknowledgment

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