

Evaluation of Anti-anxiety Activity of *Grangeamaderaspatana*(L.)Poir. Extracts in Experimental Animals

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Abstract

Grangeamaderaspatana (L.)Poiris a popular Indian medicinal plant belonging to Asteraceae family. This plant is commonly known as Madras Carpet and is grown in wet places. It has long been used in traditional Ayurvedic Indian medicine for various diseases. A wide range of phytochemical constituents have been isolated from this plant. This plant is pharmacologically studied for oestrogenicity, antifertility, analgesic, anti-inflammatory, antiarthritic, cytotoxic, antioxidant, hepatoprotective, diuretic and antimicrobial activities. Despite the widespread uses of the plant, no scientific work is reported in the literature regarding the effect of *G. maderaspatana* against anxiety like states. Chloroform (200 mg/kg, 400 mg/kg) and methanol extracts (200 mg/kg, 400 mg/kg) of *G. maderaspatana* were evaluated for anti-anxiety activity in mice using elevated plus maze apparatus. Among all these extracts, chloroform extract exhibited significant anti-anxiety activity at a dose of 400 mg/kg in mice with respect to control. The chloroform and methanol extract of *Grangeamaderaspatana* possess significant anti-anxiety activity due to presence of terpenoids, steroids and saponin.

Keywords

Anti-anxiety, Asteraceae, Elevated plus maze, Grangeamaderaspatana, Madras carpet



Greentree Group

[Received 21/11/16](#) [Accepted 13/12/16](#) [Published 10/01/17](#)

INTRODUCTION

Herbal medicines have been used by the mankind since time immemorial. Ayurveda, the oldest traditional system of India, revealed that primitive Indians had a wealthy knowledge of medicinal value of diverse plants. India has been endowed with a very rich flora due to the extreme variations in environment and geographical conditions prevailing in the country. With the advent in science, many of the crude drugs used in traditional system have been investigated scientifically¹⁻².

Grangeamaderaspatana (L.)Poiris a weed commonly known as Madras carpet usually growing in sandy lands and waste places. It is reported to contain flavonoids, diterpenes, sesquiterpenoids, steroid, and essential oils. It is a medicinal plant widely used in Indian traditional system of medicine for curing various ailments. The herb is good for pain in the eyes and ears. The root is an appetizer; astringent to the bowels, diuretic, anthelmintic, emmenagogue, galactagogue, stimulant; useful in griping, in troubles of the chest and lungs, headache, paralysis, rheumatism in the knee joint, piles, pain in the muscles, ailment of the spleen and the liver, problems

of the ear, mouth and nose; reduce perspiration (Unani). Plant is stomachic and uterine tonic³⁻⁴.

Anxiety is a cardinal symptom of many psychiatric disorders and an almost inevitable component of many medical and surgical conditions. Indeed, it is a worldwide human sentiment, closely related with appropriate fear and often helping psychologically adaptive use. The most vital clinical generalization is that anxiety is rather rarely a “disease” in itself. In addition indications of anxiety are commonly linked with despair, especially with dysthymic disorder, panic disorder, agoraphobia, neurotic disorder⁵⁻⁶.

Currently different therapeutic regimens are employed to treat anxiety and depressive disorders; but their clinical uses are restricted by their side effects such as psychomotor destruction, potentiation of other central depressant drugs and dependence liability. In the search for novel therapeutics for the treatment of neurological disorders, medicinal plant research has also contributed by signifying pharmacological efficacy of different herbs in a variety of animal models⁷.

Herbal medicines are gaining budding attention because of their cost-effective, eco-friendly feature and true relief from disease condition. Since antiquetime, the herbal medications are effective in the management of various disorders. Many plants have tradition claim in the treatment of several awful diseases but they are not scientifically exploited. Hence, these plant drugs be worthy of detailed studies in the light of contemporary medicine⁸⁻⁹.

MATERIALS AND METHODS

Collection and authentication of plant material:

The plant of *Grangeamaderaspatanawas* collected in the month of December from Saputara, Gujarat, India. The plant was identified and authenticated by Botanical Survey of India, Jodhpur and a herbarium was submitted at Botanical Survey of India

(voucher specimen sample no. BSI/AZRC/I.12012/Tech./2015-16/419).

Preparation of extracts and their phytochemical screening

The coarsely powdered material of the plant of *G. maderaspatanawas* subjected to successive solvent extraction in a Soxhlet apparatus using various solvents like petroleum ether, chloroform, ethyl acetate and methanol, in their increasing order of polarity. Water extract was prepared by maceration. After completion of extraction, the solvent was distilled off and the residue was concentrated and finally dried. The marc left after extraction with each solvent was dried completely in air before subjecting it to next solvent. The vacuum dried extracts were subjected to chemical test to detect the presence of various phytoconstituents. The Phytoconstituents present in various extracts of *G. maderaspatana* is shown in Table 1.

Table 1 "Phytochemical screening of extracts of *G. maderaspatana*"

Sr. No	Chemical constituents	Petroleum ether	Chloroform	Ethylacetate	Methanol	Water
1	Carbohydrate	-	-	-	+	++
2	Protein	-	-	-	-	-
3	Phenolics & Tannins	-	-	-	+	+
4	Saponins	-	-	-	+	++
5	Steroids	++	++	+	-	-
6	Flavanoids	-	-	-	+	+
7	Alkaloids	-	-	-	-	-
8	Terpenes	++	++	+	+	-

+ and - indicate presence and absence respectively.

Experimental Animals

Animals:

Swiss albino male mice weighing 25-30 gms, were used for all sets of experiments in

groups of six animals. They were maintained at controlled room temperature and allowed free access to food and water. The experiments were performed after the experimental protocols approved by the Institutional Animal Ethics Committee of Babaria Institute of Pharmacy and care of animals were taken as per CPCSEA guidelines.

Animals were divided into control group, standard group and extracts treated group. Each group consisted of six animals.

Ethical Committee Approval Number-
BIP/IAEC/2015/04

Acute oral toxicity studies:

Extract containing 2000 mg/kg of different phytoconstituents was administered as per OECD guidelines, orally to six mice. Effects were observed on behavior for 72 hours. Mice were examined for behavioral effects 45 minutes post administration of the extracts. No change in behavior or any abnormality in behavior was observed and no mortality was seen. Thus it was concluded that chloroform and methanol extract of *Grangeamaderaspatan* was nontoxic up to 2000 mg/kg doses. Then 1/5th and 1/10th of the administered dose was chosen for future studies as per OECD guidelines.

Elevated Plus Maze

Animals were divided into six (I-VI) groups. Group I was a negative control and animals were given carboxy methyl cellulose (1% w/v) in a dose of 10ml/kg. Group II was positive control and was given standard drug, Diazepam (1mg/kg). Groups III to IV received chloroform extract at doses of 200 and 400 mg/kg p.o., respectively, Group V to VI received methanolic extract at doses of 200 and 400 mg/kg, p.o., respectively. Test solutions and control were administered orally and standard was administered i.p., 45 minutes prior to observation.

Elevated plus maze model

The elevated plus-maze (EPM) test has been widely validated for measuring anxiolytic and anxiogenic-like activities in rodents¹⁰. This apparatus consists of two open arms (50×10 cm) crossed with two closed arms (50×10×40cm). The arms were linked together with a central square (10×10 cm). The apparatus was elevated to a height of 70 cm in a hazily illuminated room. The animals were divided into six groups containing six animals each. The chloroform extract in doses of 200, 400 mg/kg p.o. and methanolic extract (200, 400 mg/kg p.o.) were administered to groups II-VI. Control group received vehicle only (1% w/v cmc

solution). Diazepam (1mg/kg i.p.) was used as a reference standard for comparison. Each mouse was placed individually at the center of the elevated maze, 45 minutes post administration of the control, extracts and the standard. The number of entries and duration of stay in the open and closed arm in elevated plus maze during a period of 5 minutes were noted¹¹⁻¹². After each test the maze was cautiously cleaned up with a wet tissue paper (10% ethanol solution).

Statistical Analysis

Results are represented as Mean \pm SEM. The test extract, standard and control were evaluated with the help of one-way analysis of variance (ANOVA) followed by Dunnett's Test. P values < 0.05 were considered as statistically significant.

RESULTS AND DISCUSSION

In the present study, the chloroform and methanolic extract of *Grangeamaderaspatana* (200 and 400 mg/kg p.o.) were studied for their effects on the central nervous system in animal model of anxiety using elevated plus maze model.

The elevated plus maze test is based on the principle that exposure of maze leads to move towards variation which is considerably stronger than that induced by

exposure to the enclosed part of the maze¹³. All these behaviours are increased by anxiogenic agents and attenuated by anxiolytics under identical experimental conditions¹⁴.

The anxiety induced by the open field conditions is attenuated by anxiolytic drugs. As shown in figure 1, the findings of the present study indicate that chloroform (200 mg/kg p.o) and methanolic extracts (400 mg/kg p.o) of *Grangeamaderaspatana* showed significant anxiolytic activity in the elevated plus maze test may be due to presence of triterpenoids, steroids and saponins.

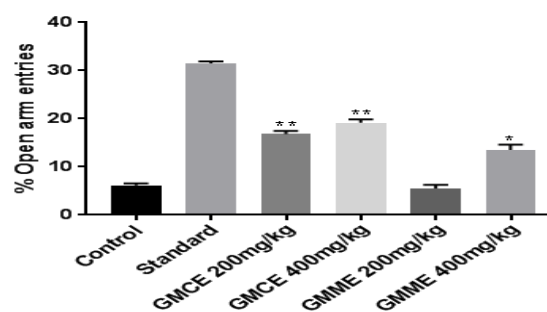


Figure 1a: % open arm entries in Elevated Plus Maze “Anxiolytic activity of chloroform (GMCE) and Methanol (GMME) extract of *Grangeamaderaspatana* in mice. Each bar represents Mean \pm SEM (n=6). One-way ANOVA followed by Dunnett's test, *P values < 0.05 when compared with

control group.”

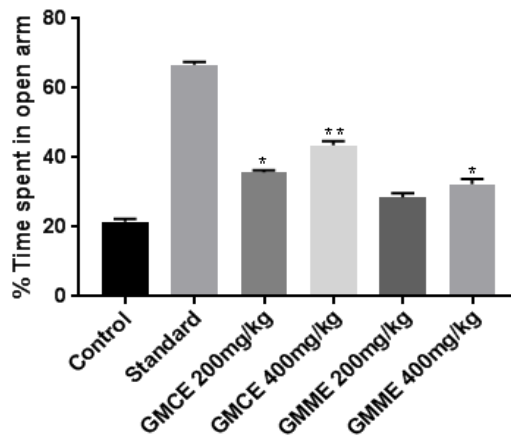


Figure 1b: % time spent in open arm in Elevated Plus Maze

CONCLUSION

The chloroform and methanol extract of *Grangeamaderaspatanapossess* considerable anti-anxiety activity and hence may prove to be valuable and an alternative in the management of anxiety like disorders. The results are sufficient to pursue further studies to suggest the fundamental pharmacological mechanism and also to separate and illustrate responsible bioactive compound.

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