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Research Article

EVALUATION OF ANTIANXIETY EFFECT OF DRIED FRUITS OF *PRUNUS AMERICANA* MARSH

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ABSTRACT

Objective: To evaluate antianxiety effect of dried fruits of *Prunus americana* Marsh. Family Rosaceae.

Methods: Phytochemical screening was performed on various extracts of the fruits. Elevated plus maze and light/dark choice tests were used for screening of antianxiety activity. Diazepam (2 mg/kg) was used as the standard drug.

Results: Phytochemical screening has shown the presence of various constituents such as flavonoids, carbohydrates, tannins, alkaloids, phenols, and saponins. Different concentrations (100 and 200 mg/kg) of the ethanolic extract of *P. americana* have shown promising results. Significant antianxiety activity was observed in ethanol extract at the dose of 200 mg/kg in both models.

Conclusion: The present study suggests that ethanolic extract of *P. americana* contains certain chemical components that are responsible for the antianxiety effect of the fruits of the plant. The plant may be considered for the management of various disorders related to anxiety.

Keywords: *Prunus americana*, Alkaloids, Phenols, Chlorogenic acid, Antianxiety, Rosaceae.

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INTRODUCTION

Plants are an essential piece of the Indian arrangement of prescription (Ayurveda) which is an antiquated and standard framework. India has one of the wealthiest plants medicinal conventions. There are evaluated to be around 25,000 compelling plant-based details, utilized as a part of pharmaceuticals. In the western world, people are aware about herbs and there is an expanding enthusiastic approach toward the nature [1]. Human nervousness is characterized as a sentiment worry or strain stemming from the reckoning of envisioned or incredible danger. This uncertainty influences one-eighth population worldwide and has turned into a critical research region in the field of psychopharmacology. Barbiturates, tricyclic antidepressants (TCAs), and benzodiazepines (BZDs) have been utilized for long time to treat nervousness. The genuine reactions related with these medications, to be specific sleep deprivation, sedation, muscle unwinding, withdrawal and resistance [BZD's, barbiturates and liquor], sexual brokenness, anticholinergic, and antihistaminic impacts (TCA's) have restricted their utilization in patients [2]. The World Health Organization in 2001 revealed that 450 million people were suffering from mental or behavioral issue and just little minority get essential treatment and 12.3% worldwide weight of disease will ascend to 15% by 2020 [3].

Prunus americana Marsh. (common name: American plum, apricot) has a critical place in human wellbeing. Vernacular names of the plant are American wild plum, osage plum, river plum, sand cherry, thorn plum, wild yellow plum, red plum, August plum, goose plum, hog plum, and sloe. Apricot is rich in minerals (potassium) and vitamins (β-carotene). Distinctive parts of the plant are utilized for the treatment of numerous illnesses, for the most part against maladies of bacterial and contagious roots. In Unani system, it is utilized as an antidiarrheal, emetic, and anthelmintic in lever maladies, ear infection and deafness, and as an expectorant for dry throat, laryngitis, lung ailments, and abscesses. It is utilized as soothing, antispasmodic, and also a solution for serious colds and bronchial asthma [4]. Apricot contains oil which is particularly useful for delicate skin and for aroused or dry skin. It has high Vitamin A content [5].

METHODS

Plant material

Dried fruits of *P. americana* were purchased from the local market of Ludhiana and authenticated by Dr. Adarsh Pal Vig (Reference no. 1240, dated 13-9-2011), Department of Botanical and Environmental Sciences, Guru Nanak Dev University, Amritsar.

Preparation of plant extract

The dried fruits of *P. americana* were kept in oven at 30°C for 15 days and then powdered. The powdered fruits were successively extracted with petroleum ether, chloroform, and ethanol using cold maceration method. The resulting extracts were filtered, concentrated, and dried in an oven at 40-50°C. All the extracts were dissolved in respective solvents and were screened for different classes of phytoconstituents [4,6].

Preliminary phytochemical screening

Various qualitative tests were performed to check the presence of active chemical constituents such as phenols, alkaloids, carbohydrates, tannins, flavonoids, and saponins present in the plant [6].

Animals

Acute oral toxicity study was performed. Swiss Albino mice of either sex (40-45 g) were used for the present investigation. Animals were kept under standard environmental conditions at temperature (25±2ºC) and light and dark (12:12 hrs). Mice were fed with standard pellet diet purchased from M/S Hindustan Lever Pvt., Ltd., Mumbai, India, and water *ad libitum*. The experimental protocol was approved by the (Institutional Animal Ethics Committee approval number: 954/ac/06/CPCSEA/12/05).

Acute toxicity study

For toxicity studies, six mice were given orally the test substance in the range of doses 200-1000 mg/kg and the mortality was observed after 72 hrs. Ethanol extract of *P. americana* has shown no mortality up to 1000 mg/kg. Therefore, 1000 mg/kg dose was considered as cutoff dose. Two doses were selected for the experiment (100 mg/kg and 200 mg/kg) [7].

Table 1: Results of phytochemical screening of various extracts of *P. americana*

Phytoconstituents	Chemical test	Petroleum ether extract	Chloroform extract	Ethanol extract
Flavonoids	Shinoda test	-	-	+
	Lead acetate	-	-	+
Alkaloids	Dragendorff's test	-	+	+
	Mayer's test	-	-	-
Tannins	Lead acetate test	+	+	+
	Acetic acid test	-	-	+
Phenols	Ferric chloride test	-	-	+
Carbohydrates	Molisch's test	-	-	+
	Fehling's test	-	-	-
Saponin	Foam test	-	+	-

P. Americana: Prunus americana

Antianxiety activity

Elevated plus maze

The apparatus comprising of two open arms (16×5 cm) and two shut arms (16×5×12 cm) having an open rooftop was utilized to watch anxiolytic conduct in animals [8,9]. Each mouse was put at the focal point of the raised apparatus (25 cm from ground) with its head confronting the open arms. Amid this 5 minutes try, the mice were watched for: (a) The quantity of passages beyond any confining influence arms, (b) normal time spent by the mouse in the open arms (normal time=add up to time spent in open arms/number of sections in arms). Extracts of *P. americana* were administered orally using a tuberculin syringe fitted with oral canula. The animals were allowed to socialize during the time of experiment and care was taken that only height of apparatus invokes anxiety in the animals.

Light/dark choice test

The light/dark box apparatus consisting of two compartments (light/dark) was used. The dark box has six divisions and light box had nine divisions. The light box (white polyvinyl chloride (PVC), 27×27×29 cm) was illuminated by a white light. The dark box (dark PVC, 27×18×29 cm) had a red light. There was a small door to connect two compartments. Every creature was put in the light box with its head confronting the entryway of the dull box. The creatures were watched for number of sections in light compartment and the aggregate time burned through there [10].

For both models, six mice were randomly allocated to the following groups: Vehicle control (physiological saline, 0.9% NaCl), (ethanolic extract of *P. americana*, 100 mg/kg and 200 mg/kg) in carboxymethyl cellulose, diazepam 2 mg/kg [11].

Statistical analysis

The qualities are communicated as mean ± standard error of the mean. ANOVA followed by Dunnett test was used for the statistical analysis.

RESULTS AND DISCUSSION

The antianxiety capacity *P. americana* was assessed utilizing two models of uneasiness, elevated plus maze, and light-dark choice test. These models are generally utilized as trial models for assessing the antianxiety capability of natural products and their compounds. It produces reproducible outcomes. The models were picked as these are successful, straightforward, less tedious, and do not require the mice to be prepared for the test; furthermore, do not make much inconvenience to the creatures while dealing with. The dread because of tallness (acrophobia) creates nervousness in the creatures when these are set on the lifted in addition to labyrinth. The sign of uneasiness and dread in the creatures is displayed by a decrease in movement. This is measured by the time spent by the creature in the open arms. In light-dark choice test, exploration of mice or rats is inhibited by bright illumination, which is highly aversive for rodents. Anxiolytics create a dosage subordinate increment in intersections. Table 1 shows results of phytochemical screening of various extracts of *P. americana*. Phytochemical screening showed the presence of flavonoids, carbohydrates, tannins, alkaloids, and phenols

Table 2: Antianxiety activity of *P. americana* ethanol extract on elevated plus maze model

Treatment	Dose	Mean entries in open arm	Mean time spent in open arms
Control	Vehicle	0.98±0.11	2.93±0.28
Diazepam	2 mg/kg	8.25±0.21**	12.73±0.66**
EEPA	100 mg/kg	2.08±0.25*	4.62±0.30*
EEPA	200 mg/kg	5.96±0.32**	8.97±0.50**

EEPA: Ethanolic extract of *Prunus americana,* n=6; The data is expressed as mean±SEM, *p<0.05 versus control, **p<0.001 versus standard, ANOVA followed by Dunnett test, *P. Americana: Prunus americana*

Table 3: Antianxiety activity of *P. americana* ethanol extract in light-dark box

Treatment	Dose	Mean entries in light compartment	Mean time spent in light compartment
Control	Vehicle	2.66±0.33	17.50±0.56
Diazepam	2 mg/kg	7.83±0.30**	36.33±0.66**
EEPA	100 mg/kg	3.33±0.33*	19.67±1.02*
EEPA	200 mg/kg	6.167±0.30**	30.50±0.61**

EEPA: Ethanolic extract of *Prunus americana*, n=6; The data are expressed as mean±SEM, *p<0.05 versus control, **p<0.001 versus standard, ANOVA followed by Dunnett test, *P. americana: Prunus americana*

in ethanol extract of *P. americana* whereas the petroleum ether extract showed tannins and chloroform extract was found to have alkaloids, tannins, and saponins to some extent. The mean number of entries and time spent by mice in open arms after oral administration of two doses viz. 100 and 200 mg/kg of ethanol extracts of *P. americana* fruits are given in Table 2. Table 3 shows the mean entries in light compartment and the time spent by mice in that compartment after oral administration of ethanol extract of *P. americana* fruits, diazepam (2 mg/kg), and the control (vehicle). Significant antianxiety activity was observed in ethanol extract at the dose of 200 mg/kg in both models. The plant is reported to have polyphenolic phytochemicals (caffeic acid, coumaric acid, rutin, chlorogenic acid, and neochlorogenic acid). Chlorogenic acid, in a dose of 20 mg/kg has shown anxiolytic effect in mice models of anxiety. Identification of compound(s) responsible for biological activity could be used as a prototype(s) to design new substances with antianxiety activity.

CONCLUSION

The present antianxiety study suggests that the ethanol extract of *P. americana* fruits contain certain promising antianxiety substances and may be considered for the purpose of managing the anxiety disorders.

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