EFFECT OF *Gloriosa superba* TUBER ON LIBIDO IN MALE ALBINO RATS

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Abstract
In the present study, we examined the aphrodisiac activity of aqueous, chloroform and alcohol extract of Gloriosa superba tuber in male albino rats.The libido were studied in male albino rat of control and experimental groups and compared with those administered with the standard reference drug Sildenafil citrate. Clinical toxicity symptoms such as respiratory distress, salivation, weight loss and change in appearance of hair as well as maternal mortality were not observed at any period of the experiment. The administration of 500 mg/kg body weight dose of plant extract showed increase in mounting frequency. At a dosage of 500 mg/kg body weight, the aqueous, chloroform, and alcohol extracts had a marked influence on libido.
***Keywords**: Aphrodisiac, Gloriosa superba, Male albino rat, libido, tuber.*

Introduction

Plants represent a significant source of medical treatments and are integral to the health of the global community.In traditional medicine, a variety of plants have been used as sex stimulants [1]. Throughout the centuries, Arabs have turned to herbal drugs to enhance sexual performance and raise libido [2]. In African traditional medicine, particularly in Cameroon, Zingiberofficinale and Pentadiplan-drabrazzeana are utilized as aphrodisiacs and for stimulating male sexual function [3].In Egypt, the pollen grains of dates (*Phoenix dactylifera*) and seeds of harmala (*Peganum harmala*) are used to restore sexual potency [4]. (*Punica granatum* was a symbol of immortality and love in oriental regions [5].

Substances that are often regarded as aphrodisiacs have the ability to cross the blood-brain barrier, thereby mimicking or stimulating particular regions involved in sexual arousal within the central nervous system.Some nutritional foods improve the well-being of the individual and consequently improve sexual performance and libido [6].

The species *Gloriosa superba* is classified under the genus *Gloriosa*, which is part of the Liliaceae family. Originally present in forest region of tropical Africa and Asia and is under cultivation in fairly large areas of India. According to Ayurveda, tuber is pungent, bitter, acrid, anthemirtic, laxative, alexiteric and useful in ulcers, leprosy, piles, inflommations, abdominal pains, itching and thirst.*Gloriosa superba* is also claimed to be an abortifacient [7] *G. superba* tuber also shows a hepatoprotective activity [8].

However the validity of the tribal claimed aphrodisiac activity of *Gloriosa superb* has not been proven scientifically. Hence this study was carried out to provide scientific support for its purported folkloric usage.

Material and Method

Collection of plant material

The plant *Gloriosa superba* was collected from Melghat region, identified and authenticated by experts from Botanical Survey of India, Pune, where a voucher specimen with herbarium accession number (SHPAGS6) was deposited.

Animal Stock

Healthy albino Wistar rats, both male and female, approximately 8 weeks in age and weighing between 100 and 160 grams, were procured from the Sudhakarrao Naik Institute of Pharmacy at Pusad. They were housed in a polypropylene cages, maintained at a temperature of approximately 25 ± 2 °C. and a photoperiod comprising 12 hours of light and 12 hours of darkness. The animals were given a standard pelleted diet (Trimurti Lab Feeds, Nagpur) and were permitted to drink water ad libitum.They were allowed a 15 days acclimatization period before the experimental session.

All the experimental protocols were met with the approval of institutional Animal Ethics Committee with registration number (1060/ac/07/CPCSEA (IAEC/01/2009).

Preparation of plant extract

The stem and leaves of *Gloriosa superba* were obtained, shade-dried, chopped into pieces, pulverized using an electric blender, and subjected to soxhlet extraction for 24 hours with distilled water at 60 °C, chloroform at 20 °C, and alcohol at 20 °C.

Preparation of test samples

Aqueous, chloroform and alcohol extract was suspended in 5 ml/kg of distilled water or olive oil (Figaro- refined olive oil, Spain)and administered

orally. Ethinyl estradiol (Cyclenorm-E – Ethinyl estradiol tablet I.P. 0.01mg) developed by India Nutri Pharma 10 ug/100 g b.w. and progesterone (Susten 100- progesterone I.P.- 100mg) Sun pharmaceutical Industries Limited, Gujarat, India) 0.5 mg/100 g b. w. were administered 48 h and 4 h respectively through subcutaneous injections. Sildenafil citrate suspension was prepared by crushing a tablet of Sildenafil citrate and administered orally at a dose of 5ml/kg in distilled water. Caverta -Sildenafil citrate IP-50mg Ranbaxy, Sirmour, India).

Treatment

The male rats were randomized into 11 groups comprising of 6 animals each. The aqueous, chloroform, and alcohol extract, after reconstitution, was delivered orally to all animals in different groups using an intragastric (ig) soft rubber catheter for 15 days at the doses specified below. Group I- administered with distilled water (5 ml/kg) served as control.

An aqueous extract was given to Group II-IV at the following doses: 100, 250, and 500 mg/kg body weight (b. w.)

Group VI-VII- administered with daily dose of chloroform extract 100, 250, 500 mg/kg b. w. respectively in olive oil (5 ml/kg).

Group VIII-X- administered with daily dose of alcohol extract 100, 250, 500 mg/kg b. w. respectively in olive oil (5 ml/kg).

Group XI- given 5 mg/Kg b. w. of Sildenafil citrate suspension.

Phytochemical analysis

The aqueous, chloroform and alcohol extract of *Gloriosa superba* were subjected to phytochemical and qualitative analysis of alkaloids, tannins, anthraquinone glycosides, saponins, phenolics, flavanoids and steroids [9].

Acute toxicity study

The healthy 60 male albino rats, starved for 3- 4 h, group I was administered with the distilled water (1 ml/rat), group II-X were administered with 1000, 2500 and 5000 mg/kg dose of aqueous, chloroform and alcohol extract was administered and subjected to acute toxicity studies.The rats were observed continuously for 2 h for behavioural, neurological and autonomic profiles and for 24 and 72 h for any lethality or death. No death was observed at highest dose (5000 mg/kg body weight) so its one tenth (500mg/kg) used for studies as per Organization of Economic Co-operation and Development (OECD) 423 guideline [10].

Test for libido

The test was carried out by the method of [11]) modified by [12]. Male rats with sexual experience were organized into five groups, each containing six rats, and were maintained in separate propylene cages on an individual basis during the experiment. Group 1 represented the control group, which received 5ml/kg of distilled water orally, once daily for 15 days at 18:00 h. Group II–IV received suspension of the *Gloriosa superba* extract orally at the doses of 100, 250 and 500 mg/kg, respectively, daily for 15 days at 18:00 h. Group V served as standard group and was given suspension of the Sildenafil citrate orally at the dose of 5 mg/kg, 1 h prior to the commencement of the experiment. To ensure receptivity of female rats, thus were brought to oestrus by the sequential administration of ethiny lestrodial (10 µg/100 g) and progesterone (0.5 mg/100 g) through subcutaneous injections, 48 h and 4 h respectively prior to pairing .The male rats were observed for Mounting Frequency (MF) on the evening of 15th day at 20:00 h. The penis was exposed by retracting the penis sheath and 5% xylocaine ointment (Astrazeneca- Lidocaine USP 5% w/w, Astrazeneca Pharma India Limited, Banglore, India) was applied 15 min before starting the observations. Each male rat was placed individually in a cage and the receptive female rat was placed in the same cage. The number of mountings was noted. The male rats were also observed for intromission and ejaculation.

Statistical analysis

The data are expressed as mean±SE. Statistical analysis was done by using paired and unpaired Student's t-test and one way analysis of variance (ANOVA) [13].

Result

Phytochemical screening of the aqueous, chlololform and alcohol extract of *Gloriosa superba* tuber showed the presence of alkaloids, steroid and saponins, while anthraquinone glycosides, tannins, and phenolic compound were found to be absent.

The result of the acute toxicity test shows no lethal or any treatment related effects of the extract of *Gloriosa superba* tuber in all treatment groups of animals. No clinical toxicity symptoms, including respiratory distress, excessive salivation, weight loss, or alterations in hair appearance, along with maternal mortality, were noted. Similarly no changes in the behavioural and neurological profiles were observed in treated groups of the rats up to highest dose of 5000 mg/kg body weight. Consequently, one-tenth of this dosage was utilized for additional testing.

Gloriosa supreba aqueous, chloroform and alcohol extract at the dose of 100, 250 and 500 mg/kg b.w. increased the mounting frequency (MF) in a significant manner (F=30.77,F=16.03,F=13.82 at P<0.01) as compared to control. A similar increase in MF was also observed in sildenafil citrate group. Increase in mounting frequency was more evident in aqueous extract (500 mg/kg) treated group of male rats when compared to the control group. However neither intromission nor ejaculation was noted in all the three extract treated group (Table-1).

Table 1: Effect of aqueous, chloroform and alcohol extracts of *Gloriosa superba* on libido in male rats

Treatment groups	Doses (mg/kg body wt.)	Mounting frequency (MF)	Intromission frequency (IF)	Ejaculation (EJ)
Control (Group I)	vehicle	1.16±0.91	Nil	Absent
Aqueous extract (Group II-IV)	100	1.66±0.20**	Nil	Absent
	250	2.83±0.16***	Nil	Absent
	500	4.16±0.16***	Nil	Absent
Chloroform extract (Group V-VII)	100	1.5±0.22 ns	Nil	Absent
	250	2.5±0.22***	Nil	Absent
	500	4±0.22**	Nil	Absent
Alcohol extract (Group VIII-X)	100	1.27±0.16 ns	Nil	Absent
	250	2.33±0.20***	Nil	Absent
	500	3.16±0.16***	Nil	Absent
Sildenafil Citrate (Group XI)	5	2.33±0.26***	Nil	Absent

***P values:* * <0.1, **<.0.01, ***<0.001, when compared with control, ns= non significant. Values are mean±S.E. n=6.**

Discussion

The *Gloriosa superba* tuber has been employed by the tribes of the Melghat area as a sexual enhancer, even in the absence of scientific support. Consequently, this research was undertaken to provide scientific validation for the assertions of the tribal people.

Inthe present study after administration of *Gloriosa superba* tuber (aqueous, chloroform and alcohol) extract (500 mg/kg b.w.) to male rats there was increase in libido (mount frequency (MF) (P<0.001) after genital anesthetization as compared to control. Similar results were obtained by chloroform, methanol, water and butanol fraction of *Eurycoma longifolia*, providing evidence that *Eurycoma longifolia* jack enhanced the libido in sexually experienced male rats.It has been identified as a significant stimulator of sexual arousal in intact, sexually vigorous male rats, occurring without feedback from genital sensations [14].

Similar results were also obtained after administration of *Psoralea carylifolia* seed to male rat at the dose of 100, 200 and 400 mg/kg showed significant increase in mounting frequency after anesthetization ensure the pure libido [15]. *Myristica fragrans* houtt (nutmeg) at the dose of 100, 250, 500 mg/kg significantly increase in mounting frequency (P<0.001) as compared to control group [16].

The effect of aqueous, alcohol and chloroform seed extract of *Moringa oleifera* at the dose of 100, 200 and 500 mg/kg on libido was studied by assessing the MF after genital anaesthetization which does away with the reinforcing effect of genital sensation thus affording study of pure libido[17].

Conclusion

In summary, the research confirms the efficacy of the herb in enhancing libido. The 500 mg/kg dosage of the aqueous extract demonstrated a significant impact on mounting frequency, thereby ensuring the extract's pure libido effects.

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