CENTRAL NERVOUS SYSTEM DEPRESSANT ACTIVITY OF ALCOHOL AND AQUEOUS ROOT EXTRACTS OF PERGULARIA DAEMIA (FORSK.) CHIOV.

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Summary

Pergularia daemia (Forsk.) Chiov. (*Asclepiadaceae*) is an important medicinal plant used to cure mental disorder, anaemia, leprosy, asthma and piles. The plant is known as Uttaravaruni and Yugaphala in *Sanskrit.* The present study was undertaken to evaluate the central Nervous System depressant activity of the roots. Central nervous system depressant activity was induced on Swiss albino mice using chlorpromazine and pentobarbitone sodium induced sleeping time. Alcohol and aqueous extracts showed significant central nervous system depressant activity, comparison was done between control and drug treated groups using Tukey kramer multiple comparison test. It is concluded that alcohol and aqueous root extract of the plant possesses central nervous system depressant activity which confirms the uses of drug mentioned in *Ayurveda*.

Keywords: *Pergularia daemia*, Pharmacology, Root, Central nervous system depressant activity.

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Introduction

Pergularia daemia (Forsk.) Chiov. (*Asclepiadaceae*) is a perennial twining herb occurs in warm parts of India, and is known as Uttaravaruni and Yugaphala in *Sanskrit*, used in Ayurvedic medicine[1]. Plant possesses stomachic, laxative and diuretic properties, used to cure cough, biliousness, sore eyes; and has anthelmintic, emetic and expectorant properties[2]. Leaf paste mixed with castor oil is applied to joints to cure inflammation, liver complaints, spleen enlargement and leaves have hypoglycaemic activity[3]. The leaf extract is useful for eye troubles, urinary discharges, leucoderma, parturition and treatment of catarrhal affections[4]. *Pergularia daemia* (Forsk.) Chiov. have been documented for antifertility, anti-inflammatory, antipyretic, analgesic, hepatoprotective, antibacterial properties[5,6,7,8].

The present investigation was carried out to determine central nervous system depressant activity of the alcohol and aqueous root extracts of *Pergularia daemia* using swiss albino mice.

Methods

Plant material: The roots of *Pergularia daemia* (Forsk.) Chiov. were collected from forest of Savanadurga, Bangalore and authenticated by Dr. S.N. Yoganarasimhan (Taxonomist and Research Co-ordinator, Department of Pharmacognosy, M.S.Ramaiah College of Pharmacy, Bangalore). A voucher herbarium specimen (*Lokesh Nikajoo* 006) in flowering and fruiting condition was deposited at the herbarium. The roots were washed, shade dried, powdered, passed through sieve no. 60 for preparation of coarse powder.

Preparation of the extracts: Alcohol extract: The powder was extracted with ethanol 95% v/v in a Soxhlet extractor by continuous heat extraction and concentrated in a rotary flash evaporator at a temperature not exceeding 50ºC. Alcohol extract was prepared in distilled water containing 2% v/v Tween 80 (as a suspending agent).

Aqueous extract: The aqueous extract was prepared by maceration in 2 % v/v chloroform water. The macerate was filtered through Whatman No.1 filter paper and concentrated in a rotary flash evaporator at a temperature not exceeding 50ºC and extract was prepared in distilled water for experimental purpose.

Phytochemical analysis: Phytochemical analysis of different extracts was carried out by using root powder. Different solvents like petroleum ether (60°-80°) benzene, chloroform, acetone and ethanol (95%) were used. After extracting with each solvent, the residue was dried in oven below 50ºc and finally macerated with chloroform water for 24 hours. Each extract was concentrated by evaporating the solvent to dryness. The dry extracts were subjected to preliminary phytochemical screening for detection of various phytoconstituents[4].

Animals: Swiss albino mice weighing 18-25 g of either sex were used for the study. The animals housed in the animal house maintained under standard hygienic conditions, at 20 ± 2^{o} C, humidity (60 ± 10%) with 12 h day and night cycle, providing food and water ad libitum. The study was carried out as per CPCSEA (Committee for the purpose of Control and supervision of Experiments on Animals) norms after obtaining approval from the Institutional Animal Ethical Committee of MSRCP.

Acute toxicity studies: Acute toxicity studies were performed to determine minimum lethal dose of the drug extracts. The alcohol and aqueous extracts were administered orally to overnight fasted animals at doses of 30, 100, 300, 1000 and 3000 mg/kg body weight. After administration of the extracts, animals were observed continuously for the first three hours for any toxic manifestation and thereafter, observations were made at regular intervals for 24 hrs. for one week[10].

Central nervous system depressant activity: CNS depressant activity of alcohol and aqueous extracts was studied by testing locomotor activity in a digital actophotometer and on the potentiation of pentobarbitone induced sleeping time[11].

Locomotor activity: In a digital actophotometer, continuous beam of light falls on photoelectric cells. When the reading is considered as zero, any cut off in the continuity of light by the animal, is recorded on a digital counter in the form of counts. Depending on CNS depressant action of the drug, the animals show reduced locomotor activity.

The animals were divided into six groups of 6 animals each. Group I served as control. After one hour, animals were individually placed in the actophotometer. The digital counts, as the number of line crossings by animal due to beam interruptions, were recorded. The counts correspond to locomotor activity. The cut off time period was 10 minutes. Group II served as standard and treated with Chlorpromazine (3 mg/kg) intraperitonially. Thirty minutes after administration of chlorpromazine, the animals were individually placed in actophotometer. The digital counts, as the number of line crossing by animal due to beam interruptions, corresponding to the locomotor activity, were recorded for 10 minutes. Group III and IV were treated with aqueous root extract and Group V and Group VI were treated with alcohol root extract (500 and 1000 mg/kg body weight respectively) orally and after one hour, animals were individually placed in the actophotometer. The digital counts, as the number of line crossing by animal due to beam interruptions, were recorded for 10 minutes.

Pentobarbitone induced sleeping time: In pentobarbitone induced sleeping time, onset of sleep was noted as the time at which the animal lost its righting reflex. The animals were then placed on their backs leaving sufficient space in between them. The time period between loss of righting reflex and recovery from sleep, as the animal turned to recover its normal posture, was noted as the duration of sleep. Comparison was done between the control and drug treated groups. The animals were divided into six groups of 6 animals each. Group I served as control. Group II served as standard group and was treated pentobarbitone (45 mg /kg) intraperitonially. The onset and duration of sleep were recorded. Group III and IV animals were treated with aqueous root extract and Group V and Group VI animals were treated with alcohol root extract

(500 and 1000 mg/kg body weight respectively) orally. The onset and duration of sleep were recorded.

Statistical analysis: All the values ware statistically analyzed by one-way analysis of variance (ANOVA) followed by Tukey- Kramer multiple comparison test. All values are expressed as mean ± SEM.

Results

The preliminary phytochemical analysis revealed the presence of alkaloids, glycosides, phytosterols, tannins, flavonoids, which helps to undertake further studies on the isolation and identification of specific phytoconstituents. Acute toxicity studies showed no toxic manifestation up to the dose level of 3000 mg/kg body weight.

Reduction in the motor activity by digital actophotometer shows significant ($p<0.001$) Central nervous system depressant effect by both alcohol and aqueous extracts at the dose level of 500 and 1000 mg/kg of body weight as shown in Table 1.

Table 1: Locomotor activity of alcohol and aqueous extracts of *Pergularia daemia* (Forsk.) Chiov. on actophotometer

Treatment	Locomotor activity (Mean ± SEM)
Control	153.33 ± 5.258
Standard (Chlorpromazine 3 mg/kg)	40.5 ± 5.246* * *
Aqueous Extract (500 mg/kg)	83.5 ± 3.695* * *
Aqueous Extract (1000 mg/kg)	53.5 ± 4.890* * *
Alcohol Extract (500 mg/kg)	93.33 ± 7.084* * *
Alcohol Extract (1000 mg/kg)	43.5 ± 3.510* * *

One- way Analysis of Variance ANOVA: p value found to be 0.0001 is considered extremely significant. The data were expressed as mean ± S.E.M.; Tukey Kramer multiple comparison test: ***$p<0.001$ (Extracts vs. control).

The symptoms included decreased respiratory rate and loss of righting reflex, prolongation of pentobarbitone sleeping time and suppression of exploratory effect exhibited by both the extracts which corresponds to barbiturates as shown in Table 2.

Table 2: Effects of alcohol and aqueous extracts of *Pergularia daemia* (Forsk.) Chiov. on pentobarbitone induced sleeping time in mice

Treatment	Onset of sleep (Mean ± SEM)	Duration of sleep (Mean ± SEM)
Control	5.83 ± 0.6009	74.5 ± 7.325
Standard (Pentobarbitone 45mg/kg)	2.83 ± 0.3073***	192.16 ±10.368***
Aqueous Extract (500mg/kg)	3.83 ± 0.3073**	94.5 ± 3.181
Aqueous Extract (1000mg/kg)	3 ± 0.3651***	172.83 ± 9.617***
Alcohol Extract (500mg/kg)	3.83 ± .3073**	108.66 ± 4.185
Alcohol Extract (1000mg/kg)	2.83 ± 0.3073***	183 ± 10.149***

One- way Analysis of Variance ANOVA: p value found to be 0.0001 is considered extremely significant. The data were expressed as mean ± S.E.M.; Tukey Kramer multiple comparison test: ***p<0.001, **p < 0.01 (Extracts vs. control).

Conclusions

The roots of *Pergularia daemia* (Forsk.) Chiov. are used in the treatment of mental disorders in *Ayurveda*[12]. The plant and its different parts as well as extracts possessed musculotropic, antispasmogenic; CNS depressant muscle relaxant and spasmogenic effect which is due to the presence of glycosides[13,14]. Hence the present study is undertaken to prove the CNS depressant effect of alcohol and aqueous root extracts of *Pergularia daemia* (Forsk.) Chiov.

Both extracts showed the CNS depressant activity hence it was concluded that glycosides are one of the phytoconstituents of the plant which are responsible for the CNS depressant activity of root extracts of *Pergularia daemia* (Forsk.) Chiov. The pharmacological studies carried out with reference to uses of the drug mentioned in *Ayurveda*; hence present study was performed to justify the claim. The results obtained from this study confirm the Central nervous system depressant activity of *Pergularia daemia* (Forsk.) Chiov.

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References

1. Yoganarasimhan S. N. Medicinal Plants of India. Vol. 1-Tamil Nadu. Cyber media, Bangalore, 2000: 405.
2. Chatterjee A. and Pakrashi S. C. The Treatise on Indian Medicinal Plants. Vol. 4. Publications and Information Directorate, New Delhi, 1995: 135-136.
3. Pandey G. Dravyaguna Vijnana. Krishnadas Academy, Varanasi, 2001: 568-571
4. Anonymous. The Wealth of India. Vol. 7. Publication and Information Directorate CSIR, New Delhi, 1997: 309-310.
5. Golam S., Gafur M.A., Shah M.A.B., Khurshid A.H.M., Biswas M.H.U., Hassan P. et al. 2001. Antifertility activity of *Pergularia daemia*. J. Medical Sci. 2001, 1: 22-24.
6. Jain S.C., Jain R., Mascolo N., Capasso F. et al. Ethnopharmacological evaluation of *Pergularia daemia* (Forsk.) Chiov. Phytotherapy Res. 1998, 12 (5), 378 - 380.
7. Sureshkumar S.V. and Mishra S.H. Hepatoprotective activity of extracts from *Pergularia daemia* (Forsk.) against carbon tetrachloride-induced toxicity in rats. Pharmacognosy Magazine. 2007, 3: 187 - 191.
8. Senthilkumar M., Gurumoorthi P. and Janardhanan K. Antibacterial Potential of some plants used by tribals in Maruthamalai hills, Tamil Nadu. Natural Product Radiance. 2005, 4: 27 - 34.
9. Harborne J. B. Phytochemical methods. 3rd ed. Chapman and Hall, London, 1998: 32-40.
10. Ghosh M. N. Fundamentals of Experimental Pharmacology. 2nd rev. ed. Scientific Book Agency, Calcutta, 1984: 144-158.
11. Kulkarni S. K. Handbook of Experimental Pharmacology. 3rd rev. ed. Vallabh Prakashan, New Delhi, 1999: 123-125.
12. Yoganarasimhan S. N. Medicinal Plants of India. Vol. 1-Karnataka. Interline, Bangalore, 1996: 356.
13. Rakhit S. Chemical investigations on *Daemia extensa* R. Br. J. Sci. Industr. Res. 1959, 18 (10): 425 - 26.
14. Anonymous. Pharmacological investigations of certain medicinal plants & compound formulations used in *Ayurveda* and *Siddha*. 1st ed. CCRAS, New Delhi, 1996: 350-352.