



Short Communication

Preliminary study on the central nervous system depressant effect of *Picrorhiza kurrooa* Royle. (Scrophulariaceae) in mice models

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SUMMARY

Picrorhiza kurrooa Royle. is a well known medicinal plant among the indigenous medical practitioners of India. Present study is the first time to report the activity on the central nervous system. Preliminary study of the hot water extract showed significant depressant activity on the hole board test as evidenced from the ambulation and head dipping scores. The extract showed better activity compared to diazepam on the duration of pentobarbital induced sleeping time.

Keywords: *Picrorhiza kurrooa*, CNS, depressant, Hellebore

INTRODUCTION

The prolonged use of modern tranquilizers and psychotropic drugs lead to a variety of autonomic, endocrine, allergic, hematopoietic and neurological side effects. Moreover, such agents primarily relieve the symptoms and offer a palliative relief of a temporary nature (Koslow *et al.*, 1995). During the last two decades, pharmacotherapy with psychoactive drugs has been increasingly recognized as most effective in the management of anxiety, stress and psychosomatic disorders. In many countries herbal medicines (e.g., St John's

Wort, Valerian) are as commonly prescribed as conventional medications for the treatment of psychiatric problems (Miller and Murray, 1998). The potential of plants as sources for new centrally acting drugs is still largely unexplored, only a small percentage have been investigated phytochemically, and the fraction submitted to biological or pharmacological screening is even smaller (Hamburger and Hostettmann, 1991).

Indian system of medicine has used plants towards the treatment of several ailments in humans for the last 5000 years (Baliga *et al.*, 2003). *Picrorhiza kurrooa* Royle. (English name: Hellebore) is a bitter plant have a remarkable reputation among the indigenous medical practitioners. The plant is well-known in the Ayurveda and has traditionally been used to treat disorders of the

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liver and upper respiratory tract (Jadhav and Pal, 2005). There are numerous reports about the hepatoprotective activity of *Picrorhiza kurrooa* and its constituents kutkin and picroliv (Ansari *et al.*, 1988; Ansari *et al.*, 1991; Visen *et al.*, 1991; Visen *et al.*, 1996; Santra *et al.*, 1998). In addition, the plant and its constituents showed anticholestatic (Saraswat *et al.*, 1993), antioxidant (Govindarajan *et al.*, 2003; Vijayakumar *et al.*, 2005), anti-inflammatory (Singh *et al.*, 1993), cyclooxygenase-2 enzyme inhibitory (Zhang *et al.*, 2005), hypolipidemic (Khanna *et al.*, 1994), and immunomodulatory activities (Wang and Pang, 2002; Vijayakumar *et al.*, 2005). Although numerous pharmacological activities have been reported for *Picrorhiza kurrooa*, however no neuropharmacological activity was found on the literature. The present study thus was undertaken to preliminary evaluate the central nervous system depressant activity of *Picrorhiza kurrooa* using mice models.

MATERIALS AND METHODS

Plant materials and extract

The rhizome of *Picrorhiza kurrooa* Royle. (Family: Scrophulariaceae) was collected from authenticated herbal shop in Dhaka and identified by Mr. M.K. Miah of Bangladesh National Herbarium, Dhaka, Bangladesh where voucher specimens were preserved. The rhizome was then dried and finely powdered by a grinding machine (Mesh size #80). The hot water extract was prepared by boiling 100 g of the powdered plant materials in 1600 ml water and was filtered and evaporated to give 400 ml of hot water extract.

Animals

Non-fasted mice (male, Swiss-webstar strain, 20 - 25 g body weight) bred in the animal house of the Department of Pharmacy, Jahangirnagar University, were used for the experiments. The animals were provided with standard laboratory food and tap water *ad libitum* and maintained at natural day

night cycle. The animals were grouped (n = 6) according to body weight. The extract was administered orally at a dose of 10 ml/kg. The research was carried out according to the rules governing the use of laboratory animals as acceptable internationally.

Hole board test

Experimental method each animal was placed carefully in the center of the field and the number of holes passed, head dipping, and the number of fecal boluses excreted recorded for a period of two min at 0, 30, 60, 120 and 240 min after the oral administration of the extract (Alamgir *et al.*, 2002).

Pentobarbital-induced sleeping time test

The extract was administered per oral 30 min before the administration of pentobarbital (i.p.; 40 mg/kg body weight). Diazepam (1 mg/kg i.p.) was used as a positive control. The animals were observed for the onset and the duration of sleep, as evidenced by the observation of the loss of righting reflex (Williamson *et al.*, 1996).

Statistical analysis

Unpaired t-tests were performed by SPSS 9.05 to test the level of significance. Probability value of 0.05 or less ($p < 0.05$) was considered as significant. The data were expressed as a ratio of sleeping time mean value of experimental animals vs. control animals.

RESULTS AND DISCUSSION

Intact animals are considered the best method for investigating the action of drugs on central nervous system. Hole board test evokes a pattern of behavior characterized by exploration (head dipping through the holes), locomotion (ambulation past the holes), and emotional defecation. Decrease in sleeping latency and increase in sleeping time are classically related to central nervous system depressant drugs (Williamson *et al.*, 1996). As like

Table 1. Effect of *Picrorhiza kurrooa* on hole board experiment

Group	0 min	30 min	60 min	120 min	240 min
Ambulation					
Control	23.25 ± 4.41	21.25 ± 3.67	16.75 ± 3.47	17.16 ± 3.20	19.91 ± 3.13
<i>P. kurrooa</i>	17.16 ± 4.36	4.00 ± 1.54 ^b	4.00 ± 4.04 ^a	4.66 ± 1.20 ^a	7.00 ± 3.17 ^a
Head Dipping					
Control	14.91 ± 0.98	11.58 ± 1.65	13.08 ± 3.04	14.75 ± 2.75	18.25 ± 3.10
<i>P. kurrooa</i>	12.00 ± 0.85	1.16 ± 0.74 ^b	2.83 ± 1.32 ^a	0.50 ± 0.50 ^b	1.83 ± 0.54 ^b
Defecation					
Control	2.00 ± 0.40	1.08 ± 0.259	0.58 ± 0.19	0.41 ± 0.19	0.33 ± 0.14
<i>P. kurrooa</i>	1.83 ± 0.87	0.333 ± 0.33	1.00 ± 0.51	0.66 ± 0.49	1.16 ± 0.54

Values are expressed as Mean ± S.E.M. in min

^a*P* < 0.01, ^b*P* < 0.001

many other centrally active drugs, barbiturates work on the cerebral cortex and thus produce their actions. Pentobarbital, a barbiturate class hypnotic drug by an allosteric modification of GABA_A receptor increases the chloride conductance and potentiates GABA_A mediated postsynaptic inhibition (Katzung, 2001). Inhibition of Cytochrome P 450 enzyme system (e.g. CYP1A2, 2C9, 2C19, 2D6, 3A4 and 4A9/11) can lessen pentobarbital metabolism and therefore increase sleeping time (Tsuj et al., 1996).

The hot water extract of *Picrorhiza kurrooa* significantly (*P* < 0.01 - *P* < 0.001) reduced the ambulation of the animals from 30 min to 240 min compared to the control animals (Table 1). The exploratory head dipping behavior was also seen to decreased from 30 min to 240 min (*P* < 0.01 - *P* < 0.001). Significant difference was not observed to the defecation compared to control. The findings in this experiment indicate possible central nervous system depressant activity of the *Picrorhiza kurrooa* extract.

The extract showed a non-significant decrease on the onset of the sleeping time (Table 2) but the duration of the pentobarbital induced sleeping time was significantly increased (*P* < 0.001). Depressant action (*P* < 0.001) of the extract is slightly better than the diazepam in terms of the duration of sleep. The findings suggest the action may be attributed to the cerebral mechanism

Table 2. Effect of *Picrorhiza kurrooa* on the hypnotic action of pentobarbital

Group	Onset of sleep	Duration of sleep
Control	353.8 ± 68.7	2519.1 ± 261.2
<i>P. kurrooa</i>	230.0 ± 24.0	6380.0 ± 607.0 ^a
Diazepam (1 mg/kg)	243.6 ± 16.2	5000.4 ± 139.8 ^a

Values are expressed as Mean ± S.E.M. (n = 6) in sec
^a*P* < 0.001

involved in the regulation of the sleep.

At the end of our discussion it must be kept in mind that the compounds present in the extracts are water soluble substances, therefore triterpenoids and flavonoids commonly detected in the extracts might be present in glycosidic forms. On the other hand, it should be considered that other classes of compounds and any other polar substance can be responsible for the central nervous system activity examined in this work (Jiménez et al., 2001). The results given in this paper are a preliminary evaluation of the plant and we suggest other CNS models to further evaluate their potency and bioactivity guided isolation of the pure compound(s).

REFERENCES

- Alamgir M, Choudhuri MSK, Jabbar S, Rajia S, Khan MTH. (2002) Central nervous system depressant effect of hot water extract of *Ocimum sanctum* L. (Labiatae). *Orient. Pharm. Expt. Med.* **2**, 101-105.
- Ansari RA, Aswal BS, Chander R, Dhawan BN, Garg

- Nk, Kapoor NK, Kulshreshtha DK, Mehdi H, Mehrotra BN. (1988) Hepatoprotective activity of kutkin the iridoid glycoside mixture of *Picrorhiza kurrooa*. *Indian J. Med. Res.* **87**, 401-404.
- Ansari RA, Tripathi SC, Patnaik GK, Dhawan BN. (1991) Antihepatotoxic properties of picroliv: an active fraction from rhizomes of *Picrorhiza kurrooa*. *J. Ethnopharmacol.* **34**, 61-68.
- Baliga MS, Jagetia GC, Rao SK, Babu K. (2003) Evaluation of nitric oxide scavenging activity of certain spices *in vitro*: A preliminary study. *Nahrung Food* **47**, 261-264.
- Govindarajan R, Vijayakumar M, Rawat AKS, Mehrotra S. (2003) Free radical scavenging potential of *Picrorhiza kurrooa* Royle ex Benth. *Indian J. Exp. Biol.* **41**, 875-879.
- Hamburger M, Hostettmann K. (1991) Bioactivity in plants: the link between phytochemistry and medicine. *Phytochemistry* **30**, 3864-3874.
- Jadhav HR, Pal SC. (2005) Chemistry and pharmacology of *Picrorhiza kurrooa* royle ex Benth. *Pharmacog. Mag.* **1**, 140-144.
- Jiménez G, Hasegawa M, Rodríguez M, Estrada O, Méndez J, Castillo A, Gonzalez-Mujica F, Motta N, Vásquez J, Romero-Vecchione E. (2001) Biological screening of plants of the Venezuelan Amazons. *J. Ethnopharmacol.* **77**, 77-83.
- Katzung B. (2001) Basic and Clinical Pharmacology. 8th ed. New York: McGraw-Hill.
- Khanna AK, Chander R, Kapoor NK, Dhawan BN. (1994) Hypolipidemic activity of picroliv in albino rats. *Phytother. Res.* **8**, 403-407.
- Koslow SH, Murthy RS, Coelho GV. (1995) Decade of the Brain: U.S. Department of Health and Human Services.
- Miller LG, Murray WJ. (1998) Herbal Medicinals - A Clinicians Guide. New York: Pharmaceutical Products Press.
- Santra A, Das S, Maity A, Rao SB, Mazumder DN. (1998) Prevention of carbon tetrachloride-induced hepatic injury in mice by *Picrorhiza kurrooa*. *Indian J. Gastroenterol.* **17**, 6-9.
- Saraswat B, Visen PKS, Patnaik GK, Dhawan BN. (1993) Anticholestatic effect of picroliv, active hepatoprotective principle of *Picrorhiza kurrooa*, against carbon tetrachloride induced cholestasis. *Indian J. Exp. Biol.* **31**, 316-318.
- Singh GB, Bani S, Singh S, Khajuria A, Sharma ML, Gupta BD, Banerjee SK. (1993) Antiinflammatory activity of the iridoids kutkin, picroside-1 and kutkoside from *Picrorhiza kurrooa*. *Phytother. Res.* **7**, 402-407.
- Tsuj R, Isobe N, Kurita Y, Hanai K, Yabusaki Y, Kawasaki H. (1996) Species difference in the inhibition of pentobarbital metabolism by empenethrin. *Environ Toxicol. Pharmacol.* **2**, 331-337.
- Vijayakumar M, Govindarajan R, Singh DP, Rawat AKS. (2005) Review of some important plants with antioxidant and other biological activities. *Pharmacog. Mag.* **1**, 129-135.
- Visen PKS, Shukla B, Patnaik GK, Kaul S, Kapoor NK, Dhawan BN. (1991) Hepatoprotective activity of picroliv, the active principle of *Picrorhiza kurrooa*, on rat hepatocytes against paracetamol toxicity. *Drug Dev. Res.* **22**, 209-219.
- Visen PKS, Saraswat B, Patnaik GK, Agarwal DP, Dhawan BN. (1996) Protective activity of picroliv isolated from *Picrorhiza kurrooa* against ethanol toxicity in isolated rat hepatocytes. *Indian J. Pharmacol.* **28**, 98-101.
- Wang X, Pang J. (2002) Protective effects of total glucosides of *Picrorhiza kurrooa* on immune liver injury of rats. *Guangxi Yike Daxue Xuebao* **19**, 524-526.
- Williamson EM, Okpako DT, Evans FJ. (1996) Pharmacological Methods in Phytotherapy Research. Selection Preparation and Pharmacological Evaluation of Plant Material. England: John Willey & Sons Ltd.
- Zhang Y, DeWitt DL, Murugesan S, Nair MG. (2005) Cyclooxygenase-2 enzyme inhibitory triterpenoids from *Picrorhiza kurrooa* seeds. *Life Sci.* **77**, 3222-3230.