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Research Article

EVALUATION OF ANTI-DIARRHOEAL ACTIVITY OF METHANOLIC RHIZOME EXTRACT OF *PICRORRHIZA KURROA* ROYLE EX. BENTH

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ABSTRACT

Picrorrhiza kurroa (Scrophulariaceae) is a small perennial herb growing in the hilly parts of the north-Western Himalayas region in India and Nepal. The objective of the present study was to evaluate and compare anti-diarrhoeal activity of *Picrorrhiza kurroa* royle ex. Benth on magnesium su;phate induced enteropooling and gastro intestinal motility test using charcoal meal methods. The results suggested that there was a significant reduction in the peristaltic movemets in charcoal meal test and reduction in the intestinal fluid secretions in magnesium sulphate induced enteropooling method indicating its anti-diarrhoeal activity. Two doses of the test extract i.e. 250mg/kg, 500mg/kg were used to evaluate the anti-diarrhoeal activity. Atropine sulphate was used as the standard in the gastro intestinal motility test to compare the test results. Loperamide was used as the standard drug in magnesium sulfate induced enteropooling method to compare the test results. The study concluded that the methanolic rhizome extract of *Picrorrhiza kurroa* showed significant Anti-diarrhoeal action.

Keywords: *Picrorrhiza kurroa*, Anti-diarrhoeal action, Loperamide, magnesium sulfate induced enteropooling, gastro intestinal motility test, Atropine sulfate

INTRODUCTION

Picrorrhiza kurroa royle ex. Benth belonging to the family Scrophulariaceae is a small perennial herb that is widely distributed in the north – West India on the slopes of Himalayas between 3000 and 5000mts[1,2]. *Picrorrhiza kurroa* is an important herb in the traditional Ayurvedic system of medicine and has been used to treat liver and brochial problems. Other traditional uses include treatments of dyspepsia, bilious fever, chronic dysentery and scorpion sting. The most important active constituents of *Picrorrhiza kurroa* are the cucurbitacin glycosides, apocyanin, drosin, iridoid glycosides, picrosides and kutkin[3,4]. Diarrhoeal diseases are one of the leading causes of morbidity and mortality in developing countries and are responsible for the death of millions of people each year[5]. There are a large number of epidemiological and experimental evidence pertaining to worldwide acute diarrhoeal disease, which is one of the principle causes of death in infants[6.] As there is no literature available on anti-diarrhoeal action of *Picrorrhiza kurroa*, the present study was taken up to evaluate for Anti-Diarrhoeal action using magnesium sulfate induced enteropooling and gastro intestinal motility test methods of methanolic rhizome extract of *Picrorrhiza kurroa* royle ex. Benth.

MATERIALS AND METHODS

Plant collection, identification and authentication

The plant specimen was collected from S.V University, Tirupati, India and identified as *Picrorrhiza kurroa* Royle ex. Benth. Belonging to the family Scrophulariaceae, Voucher No: SDIP, Ref No: 002 dated 26/10/2012 and authenticated by Dr.Madhavachetty, Botanist, Tirupati. The rhizomes of the plant were dried in vacuum oven at 40° C.

Preparation of plant extract

Rhizomes of *Picrorrhiza kurroa* plant are coarsely powdered and are successively extracted by continuous hot percolation method using Soxhlet apparatus employing methanol followed by distillation to recover the excess solvent. Methanolic extraction yielded sufficiently good quantity of the product. The extract was later subjected to drying and stored in a desiccator for further use[7]. The extract is soluble in water. Therefore, from the dried methanolic extract, accurately 250mg/ml and 500mg/ml solutions were prepared using distilled water.

Standard used for the activity

Loperamide and Atropine sulfate were used as the standard drugs to compare the test results. Loperamide was prepared in the concentration of 3mg/kg in distilled water and Atropine sulfate was prepared in the concentration of 5mg/kg body weight.

Animals used for the activity

Male wistar rats (150-180 gms) were used for the study and kept at the laboratory animal house of Sree Dattha Institute of Pharmacy for acclimatization to laboratory environment. They were kept in well cross ventilated room at 27±2°C for 1 week before the commencement of experiment. Animals were provided with commercial rodent pellet diet and water ad libitum.

Method

Anti-diarrhoeal activity was evaluated using two methods i.e magnesium sulfate induced enteropooling and gastro intestinal motility test using charcoal meal methods.

Magnesium sulfate induced enteropooling method:

Male wistar rats were fasted for 18hrs and divided into four groups of five animals each. Solution of magnesium sulfate was made in the concentration of 10% w/v using distilled water. Group-I animals received normal saline (2ml, p.o.) served as control group. Group-II animals served as standard and received loperamide (3mg/kg, p.o.). Group-III animals received the test extract in the concentration of 250mg/kg and Group-IV animals received the test extract in the concentration of 500mg/kg p.o. immediately after the treatment magnesium sulfate (10% w/v) was administered. After 30minutes following the administration of magnesium sulfate, the rats were sacrificed and the small intestine was removed after tying the ends with threads and weighed. The intestinal content was collected into a graduated cylinder and their volume was measured. The intestine was reweighed and the difference between the full and empty intestine was calculated[8, 9]. The results are tabulated in Table: 1.

Gastro intestinal motility test

Male wistar rats were fasted for 18hrs and divided into four groups of five animals each. Group-I served as the control and was treated orally with distilled water. Group-II animals served as the standard and were treated with atropine (5mg/kg, i.p.). Animals of Group-III received orally 250mg/kg of the methanolic test extract and Group-IV received orally 500mg/kg of the methanolic test extract. After 1 hr, each animal was administered orally with charcoal meal 0.25ml (10 % charcoal in distilled water). Thirty minutes later, the animals were sacrificed. Total small intestine from pylorus to caecum was isolated and the total length and the length travelled by the charcoal meal were measured. This distance was expressed as a percentage of the length of the small intestine[10]. The results are tabulated inTable:2.

Statistical Analysis

Data was expressed as Mean ± Standard error of mean (SEM) and statistical analysis was carried out by one way analysis of variance (ANOVA) followed by Dunnett's t test.

Table: 1 Effect of methanolic rhizome extract of *Picrorrhiza kurroa* on magnesium sulfate induced enteropooling

Treatment	Dose (mg/kg)	Volume of fluid (ml)	Weight of intestinal content (gms)	% inhibition
Control	--	7.85 ± 0.29	11.01 ± 0.29	--
Loperamide	3	4.87 ± 0.23*	06.98 ± 0.23*	34.74
MRPK	250	5.65 ± 0.21*	09.41 ± 0.35*	18.87
MRPK	500	4.93 ± 0.23*	07.56 ± 0.31*	29.98

Results are expressed as Mean ± SEM; n=5 in each group; *p<0.05, MRPK is methanolic extract of *Picrorrhiza kurroa*

Table: 2 Effect of methanolic rhizome extract of *Picrorrhiza kurroa* on Gastro intestinal motility test

Treatment	Dose (mg/kg)	% movement by charcoal
Control	--	87.78 ± 8.02
Atropine sufate	3	38.53 ± 2.62*
MRPK	250	53.65 ± 6.21*
MRPK	500	46.53 ± 5.34*

Results are expressed as Mean ± SEM; n=5 in each group; *p<0.05, MRPK is methanolic extract of *Picrorrhiza kurroa*

RESULTS AND DISCUSSION

The % yield of methanolic extract of rhizomes of *Picrorrhiza kurroa* after 24hrs of hot percolation was found out to be 34%. The preliminary phytochemical screening showed the presence of carbohydrates, glycosides, saponins, steroid like phytochemical constituents. Cucurbitacins, Phenolic, Iridoid glycosides are some of the principle constituents responsible for various pharmacological activities. Iridoid glycosides like kutkin, Picroliv, Picrisides I, II, III & IV, Kutkosides are the chemical moieties that may be responsible for Anti-diarrhoeal activity[11]. Table 1 & 2 reveals that the methanolic extract of *Picrorrhiza kurroa* rhizomes showed significant anti-diarrhoeal activity in a dose dependent manner. Magnesium suphate induced enteropooling method shows that there is a decrease in the volume of fluid accumulation and weight of intestinal content before and after emptying when compared to the control and standard. 500 mg/kg dose showed a significant response. Gastro intestinal motility test revealed that, the test extract showed a marked reduction in the gastro intestinal motility contributing for its constipating effect.

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