

RESEARCH ARTICLE

Evaluation of skeletal muscle relaxant activity of methanolic extract of *Hibiscus rosa sinensis* leaves in albino rats

Sudha Madhuri A, Roopa C R

Department of Pharmacology, Navodaya Medical College, Raichur, Karnataka, India

Correspondence to: Sudha Madhuri A, E-mail: allellisudhamadhuri@gmail.com

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ABSTRACT

Background: *Hibiscus rosa sinensis* which belongs to family Malvaceae is traditionally used in various diseases due to its medicinal properties. Most of the uses were studied in detail by many researchers, but the studies evaluating the skeletal muscle relaxant activity of *H. rosa sinensis* leaves are limited. **Aims and Objectives:** The present study was undertaken to evaluate the skeletal muscle relaxant activity of the methanolic extract of *H. rosa sinensis* leaves (MEHRL) in albino rats. **Materials and Methods:** MEHRL was prepared and tested for skeletal muscle relaxant activity in albino rats weighing 200–250 g. The animals were divided into four different groups. Group I (control) received normal saline 10 ml/kg, Group II (standard) received diazepam 10 mg/kg, and Groups III and IV received MEHRL 200 mg/kg and 400 mg/kg body weight, respectively. Skeletal muscle relaxant activity (motor coordination) on rotarod and locomotor activity on photoactometer was performed. Statistical analysis was carried out using analysis of variance, followed by Dunnett's multiple comparison tests. **Results:** The results from the actophotometer and rotarod test showed that MEHRL significantly reduced the motor coordination and locomotor activity of tested animal. **Conclusion:** MEHRL showed significant dose-dependent skeletal muscle relaxant activity.


KEY WORDS: Actophotometer; *Hibiscus rosa sinensis*; Rotarod; Skeletal Muscle Relaxant**INTRODUCTION**

Skeletal muscle relaxants are drugs that reduce the muscle tone. They act peripherally at the neuromuscular junction (neuromuscular blockers)/muscle fiber itself or centrally in the cerebrospinal axis to reduce muscle tone or cause paralysis. The neuromuscular blocking agents as adjuncts during general anesthesia to provide muscle relaxation for surgery, while centrally acting agents are used for painful muscle spasms and spastic neurological conditions.^[1] However, these drugs cause several adverse effects. Thus,

looking for an effective alternative has always been a priority in this regard.

Hibiscus rosa sinensis (family - Malvaceae) is a perennial shrub available throughout India. Various parts of this plant such as leaves, flowers, and roots have been known to possess medicinal properties such as aphrodisiac, menorrhagic, and oral contraceptive.^[2]

Pharmacological studies have demonstrated that *H. rosa sinensis* exhibits wide range of properties antinociceptive, anti-inflammatory,^[3] antioxidant, wound healing,^[4] anticonvulsant,^[5] antidiabetic, hypolipidemic,^[6-8] anxiolytic, and central nervous system depressant,^[9] promotes hair growth, and prevents graying of hair,^[10] gastroprotective,^[11] and antibacterial.^[12] The studies evaluating the skeletal muscle relaxant activity of *H. rosa sinensis* leaves are limited. The main purpose of the study was to evaluate skeletal muscle relaxant activity of *H. rosa sinensis* leaves.

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MATERIALS AND METHODS

Collection of Plant Material

Leaves of *H. rosa sinensis* were obtained from local market, and the identification was done.

Preparation of Extract

The leaves were shade dried and pulverized to a fine powder. Methanolic extract of leaves was prepared using Soxhlet apparatus using methanol as solvent. The extract was dried under vacuum. The percentage of yield was 8% w/w stored at room temperature and protected from sunlight.

Animals

Adult albino rats of either sex weighing between 200 and 250 g were used in this study. Animals were obtained from NIN, Hyderabad, India. The animals were stabilized for 1 week at a temperature $25 \pm 1^\circ\text{C}$ and $60 \pm 5\%$ relative humidity and 12 h dark-light cycles. They have been given a standard pellet diet and water *ad libitum*. All the experiments were conducted with prior permission taken from the Institutional Animal Ethics Committee.

Drugs and Chemicals

Diazepam (Lupin Laboratories Ltd., India) and other chemicals were analytical grade.

Experimental Models

The rats were randomly allocated into four groups ($n = 6$) and treated as follows:

- Group I: Control received vehicle only (normal saline 10 ml/kg)
- Group II: Standard received 10 mg/kg diazepam
- Group III: Test rats received methanolic extract of *H. rosa sinensis* leaves (MEHRL) 200 mg/kg
- Group IV: Test rats received MEHRL 400 mg/kg.

All the drugs were administered orally.

Skeletal Muscle Relaxant Activity (Motor Coordination)^[13]

The muscle relaxant activity was assessed with the help of rotarod. Rats were divided in to four groups consisting of six animals each. The animals retained on rotarod (25 rpm) for 5 min were included in the study. Rats were placed on the rotarod; falloff time from rotarod before and 30 min after drug administration was noted.

The difference in the falloff time from rotarod between control and treated rats was taken as index of muscle relaxation.

Locomotor Activity

The spontaneous locomotor activity was assessed with the help of a photoactometer.^[14] It consists of six built-in light source, photo sensor, and a digital counter to indicate locomotor activity. The device operates on photoelectric cells which are connected to the circuit with the counter when the beam of light falling on photocells is cut off by the animal, and a count is recorded.

To see the locomotor activity, the photoactometer was turned on and each rat was placed individually in the cage for 5 min. The basal activity score for all the animals was noted. All the drugs were administered orally after 1 h, and the activity score for 5 min was observed. The difference in the activity score before and after drug administration was noted. The percentage decrease in motor activity was calculated.

Statistics

Results were expressed as mean \pm standard deviation. Statistical analysis was performed using one-way analysis of variance followed by Dunnett’s test. $P < 0.05$ was considered statistically significant.

RESULTS

Rotarod (Test for Muscle Relaxation)

The test doses MEHRL 200 mg/kg and 400 mg/kg showed significant dose-dependent reduction in motor coordination, that is, 36% and 48%, respectively. The standard drug showed 68.6% reduction in motor coordination [Table 1].

Table 1: Effect of MEHRL on muscle coordination on the rotarod apparatus

Groups	Fall of time (s)		% reduction
	Before	After	
Group I - control NS (10 mg/kg)	311.66 \pm 2.18	313.5 \pm 3.06	-
Group II - standard (10 mg/kg) diazepam	328.5 \pm 2.84	103.16 \pm 2.38*	68.596
Group III - MEHRL (200 mg/kg)	318.33 \pm 1.86	203.5 \pm 2.26*	36.072
Group IV - MEHRL (400 mg/kg)	335.66 \pm 2.32	174.66 \pm 1.84*	47.965

Results expressed as mean \pm SD. $P < 0.001$ *compared to control. SD: Standard deviation, MEHRL: Methanolic extract of *Hibiscus rosa sinensis* leaves

Table 2: Effect of MEHRL on the locomotor activity in the actophotometer

Groups	Actophotometer score		% reduction
	5 min before administration	60 min after administration	
Group I - control NS (10 mg/kg)	177.6±2.96	173.16±1.08	-
Group II - standard (10 mg/kg) diazepam	185.33±2.06	30.66±2.26*	84.45
Group III - MEHRL (200 mg/kg)	198.5±1.02	75.33±2.84*	62
Group IV - MEHRL (400 mg/kg)	173.66±2.18	53.55±1.92*	69.16

Results expressed as mean±SD. $P < 0.001$ *compared to control. SD: Standard deviation, MEHRL: Methanolic extract of *Hibiscus rosa sinensis* leaves

MEHRL (200 mg/kg and 400 mg/kg) showed significant ($P < 0.001$) skeletal muscle relaxant activity.

Actophotometer: (Test for Locomotor Activity)

The percentage of reduction in the locomotor activity after 1 h was 62% and 69% at the doses of 200 mg/kg and 400 mg/kg MEHRL, respectively, and the standard drug showed 84.5% reduction when compared to control.

MEHRL (200 mg/kg and 400 mg/kg) showed statistically significant ($P < 0.001$) reduction in locomotor activity [Table 2].

DISCUSSION

In recent years, the herbal medicines have been extensively used in various diseases due to their safety profile. The objective of the present study was to evaluate the effect of MEHRL on muscle relaxant activity in experimental animals. Actophotometer is used for evaluating the locomotor activity and antianxiety activity in rodents and rotarod for muscle relaxant activity. MEHRL significantly reduced motor coordination and locomotor activity in experimental animals. A decrease in locomotor activity indicates sedative action. Diazepam used as standard here at 10 mg/kg orally showed significant muscle relaxant activity in comparison with control as well as extract. Diazepam has low muscle relaxant sedation ratio, and sedation limits the dose used for muscle relaxation.^[15]

Previous neuropharmacological studies on *H. rosa sinensis* roots showed significant skeletal muscle relaxant and sedative effect.^[16] Our study also collaborates to these findings.

The MEHRL containing flavonoids (hibiscitin) phenolic content as well as terpenoid compounds such as beta-sitosterol and campesterol^[17] is probably responsible for the actions.

The major limitation of the study is that phytochemical analysis was not done to identify the exact constituents. Further, extensive phytochemical analysis and research are necessary to identify the exact constituents and elucidation of its possible mechanism of action underlying the muscle relaxant activity of MEHRL.

CONCLUSION

MEHRL showed significant muscle relaxant activity in a dose-dependent manner.

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