

EFFECT OF *RANDIA DUMETORUM* LAM ON CLONIDINE AND HALOPERIDOL-INDUCED CATALEPSY IN MICE

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Summary

In the present study we evaluated the anticataleptic efficacy of ethanolic extract of *Randia dumetorum* fruits in clonidine and haloperidol-induced catalepsy in mice. Five groups (n=5) of albino mice were used in the study. Catalepsy was induced by clonidine (1 mg/kg, s.c.) and haloperidol (1 mg/kg, i.p) in clonidine and haloperidol-induced catalepsy in mice respectively. The degree of catalepsy (cataleptic score) was measured as the time the animal maintained an imposed posture.

In the clonidine-induced catalepsy model significant ($p < 0.01$) reduction in the cataleptic scores was observed in all the groups treated with *Randia dumetorum* Lam (180, 360, 720 mg/kg, p.o.) however there was no significant reduction in the cataleptic scores in the haloperidol-induced catalepsy model in mice. The results obtained in the present study indicate that the anticataleptic activity exhibited by ethanolic extract of *Randia dumetorum* Lam fruits can be attributed to the antihistaminic activity; however *Randia dumetorum* does not possess antidopaminergic and antiserotonergic activity.

Keywords: *Randia dumetorum*, Clonidine, Haloperidol, Catalepsy.

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Introduction

Randia dumetorum Lam. (Sans. – Madana, Eng - Emetic nut, Hindi – Mainphal, Mah - Gelaphal) is a small thorny tree found all over India upto an altitude of 1350 meters in the hills. *Randia dumetorum* has been recommended in Ayurvedic system of medicine for variety of diseases. The fruit is said to be anti-asthmatic, emetic, expectorant, diaphoretic, nauseant, anthelmintic, abortifacient and antispasmodic. Bark is a sedative and nervine calmative. It is administered internally and applied externally in the form of a paste in rheumatism and to relieve pain of bruises and bone-aches during fevers and to disperse abscesses. It also acts as an astringent and is useful in diarrhea and dysentery¹.

Catalepsy is a condition in which the animal maintains imposed posture for long time before regaining normal posture. Catalepsy is a sign of extrapyramidal side effects of drugs that inhibit dopaminergic transmission or increase histamine release in brain. Clonidine, a α_2 -adrenoceptor agonist, induces dose-dependent catalepsy in mice, which is inhibited by histamine H₁ receptor antagonists but not by H₂ receptor antagonist². Intracerebroventricular injection of histamine in conscious mice induced catalepsy, which was inhibited by H₁ receptor antagonist but not by H₂ receptor antagonists³. It is known that clonidine releases histamine from mast cells⁴. There are histamine containing mast cells in brain⁵. Brain histamine does play a definite role in the production of the extra pyramidal motor symptoms of catalepsy. Therefore it has been suggested that the cataleptic effect of Clonidine in the mouse be mediated by histamine (via H₁ receptors), which is released from the brain mast cells in response to stimulation of α_2 adrenoreceptors by Clonidine².

Neuroleptic agents also induce catalepsy, but by different mechanism. Neuroleptics inhibit dopamine D₂ receptors in the substantia nigra^{6, 7}. Dopamine receptors in the striatum are involved in this neuroleptic-induced catalepsy⁸. Dopamine receptors have been shown to be equally distributed post-synaptically on striatal neurones and pre-synaptically on cortico-striatal terminals⁹. There is considerable evidence that blockade of dopamine transmission produces catalepsy^{6, 10} in rats and extrapyramidal side effects in humans¹¹.

The objective of the present study was to evaluate the anticataleptic efficacy of ethanolic extract of *Randia dumetorum* fruits in clonidine and haloperidol-induced catalepsy in mice.

Material and methods

Plant material

Fruits of *Randia dumetorum* Lam. were collected from local market of Pune and authenticated by Botanical Survey of India, Pune, where a sample specimen (Voucher number: PBN 01) has been deposited.

Extraction

Dried and coarsely powdered fruits of *Randia dumetorum* Lam. were subjected to solvent extraction in soxhlet extractor using ethanol as solvent (Hot method). Ethanol extract was dried and mixed with equal parts of gum acacia. The yield obtained was 6 % w/w.

Animals

Male albino mice (Swiss strain) weighing 25-28 g were housed under standard laboratory conditions of light and dark cycle (12:12), in groups of five each. The animal had free access to food and water. The ethical committee of the institute approved the protocol of the study.

Drugs and chemicals

The following drugs and chemicals were used for this study:

Drugs: Clonidine – Unichem, India

Haloperidol – Searle, India

Chlorpheniramine maleate - Research Lab Fine Chem. Industries, India.

Chemicals: Ethanol AR grade.

Effect on Clonidine-Induced Catalepsy in mice^{12, 13}

Bar test was used to study the effect of ethanolic extract of *Randia dumetorum* Lam on clonidine-induced catalepsy in mice. All the animals in the different experimental groups (n = 5) were administered clonidine (1 mg/kg, s.c.) 1 hr after administration of vehicle distilled water + equal parts of gum acacia (10 ml/kg, p.o.), standard drug chlorpheniramine maleate (10 mg/kg, i.p.) and ethanolic extract of *Randia dumetorum* Lam (180, 360 and 720 mg/kg, p.o.). The forepaws of mice were placed on horizontal bar (1cm in diameter, 3 cm above the table) and the time required to remove the paws from bar was noted for each animal and the duration of catalepsy was measured at 15, 30, 60, 90, 120, 150 and 180 min.

Effect on haloperidol-induced catalepsy in mice^{12, 13}

Bar test was used to study the effect of ethanolic extract of *Randia dumetorum* Lam on haloperidol-induced catalepsy in mice. All the animals in the different experimental groups (n = 5) were administered haloperidol (1 mg/kg, i.p.) 1 hr after the treatment with vehicle distilled water + equal parts of gum acacia (10 ml/kg, p.o.), standard drug chlorpheniramine maleate (10 mg/kg, i.p.) and ethanolic extract of *Randia dumetorum* Lam (180, 360 and 720 mg/kg, p.o.). The forepaws of mice were placed on horizontal bar (1cm in diameter, 3 cm above the table) and the time required to remove the paws from bar was noted for each animal and the duration of catalepsy was measured at 15, 30, 60, 90, 120, 150 and 180 min.

Statistical analysis

The data is presented as mean \pm SEM. The data was analysed by one-way ANOVA followed by Dunnett's test. $p < 0.05$ and 0.01 was considered significant.

Results

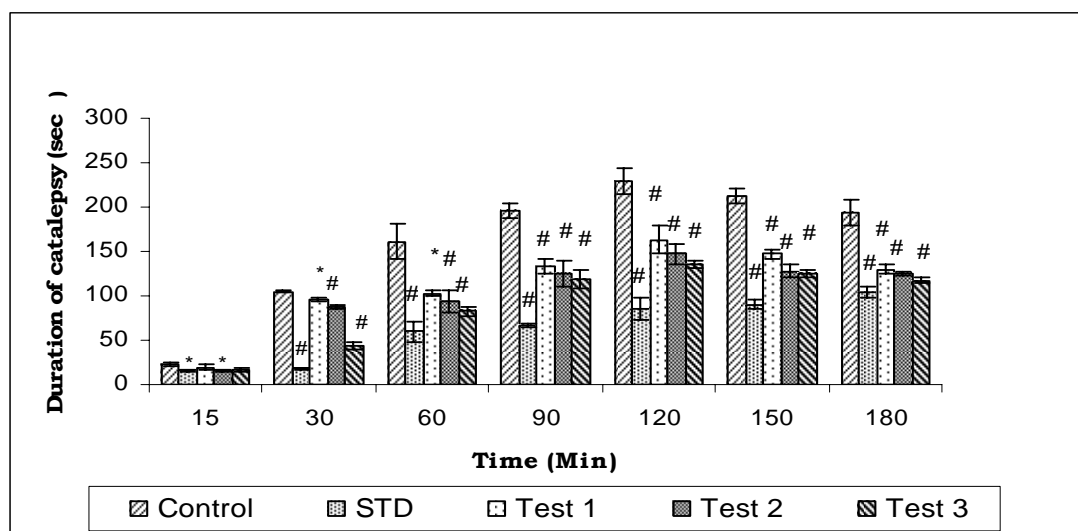
Clonidine Induced-Catalepsy in mice

Clonidine (1 mg/kg, s.c.) produced catalepsy in mice, which remained for 2 hr. The vehicle treated group showed maximum duration of catalepsy (229.2 ± 14.88 sec.) at 120 minute after the administration clonidine. There was significant inhibition ($p < 0.05$) of clonidine-induced catalepsy in the animals pretreated with *Randia dumetorum* Lam extract (180, 360, 720 mg/kg, p.o) and the duration of catalepsy was found to be 163.4 ± 15.27 , 147.4 ± 11.85 and 135.2 ± 4.84 seconds respectively at 120 minute after the administration of clonidine. Chlorpheniramine maleate (10 mg/kg, i.p.) significantly inhibited ($p < 0.01$) the clonidine-induced catalepsy in mice at 120 minute after the administration of clonidine.

Table 1: Effect of *Randia dumetorum* Lam on Clonidine-induced catalepsy in mice.

Groups (n=5)	Duration of catalepsy (sec) at Mean \pm SEM						
	15 min	30 min	60 min	90 min	120 min	150 min	180 min
I	23.20 \pm 1.65	104.40 \pm 1.20	161.20 \pm 20.21	196.20 \pm 8.57	229.20 \pm 14.88	212.00 \pm 8.22	193.40 \pm 14.73
II	16.00 \pm 1.58*	18.20 \pm 1.11 [#]	59.40 \pm 10.84 [#]	67.00 \pm 2.77 [#]	84.60 \pm 12.48 [#]	90.60 \pm 6.16 [#]	103.60 \pm 6.63 [#]
III	19.40 \pm 2.71	95.80 \pm 2.47*	102.60 \pm 3.07*	132.60 \pm 8.36 [#]	163.40 \pm 15.27 [#]	147.20 \pm 5.35 [#]	129.80 \pm 5.07 [#]
IV	15.80 \pm 1.11*	87.20 \pm 1.39 [#]	93.80 \pm 13.21 [#]	125.00 \pm 14.92 [#]	147.40 \pm 11.85 [#]	128.00 \pm 6.93 [#]	124.20 \pm 1.88 [#]
V	17.00 \pm 1.44	44.00 \pm 3.75 [#]	82.80 \pm 5.25 [#]	119.20 \pm 10.59 [#]	135.20 \pm 4.84 [#]	125.60 \pm 3.75 [#]	117.60 \pm 2.73 [#]

Values are mean \pm SEM; n = 5 in each group, * $p < 0.05$, # $p < 0.01$, compared to control group (One way ANOVA followed by Dunnett's test).

Figure 1: Effect of *Randia dumetorum* Lam on Clonidine induced catalepsy in mice.

*p<0.05, # p<0.01, compared to control group (One way ANOVA followed by Dunnett's test).

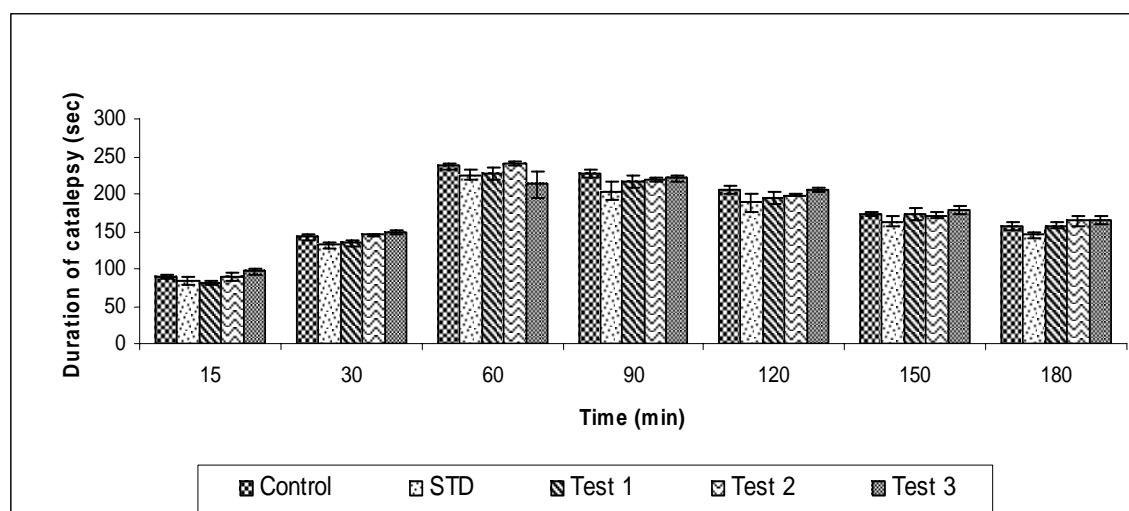
Haloperidol-Induced catalepsy in mice

Randia dumetorum Lam extract did not inhibit haloperidol induced catalepsy in mice.

Table 2: Effect of *Randia dumetorum* Lam on Haloperidol induced catalepsy in mice.

Groups (n=5)	Duration of catalepsy (sec) at						
	Mean \pm SEM						
	15 min	30 min	60 min	90 min	120 min	150 min	180 min
I	89.60 \pm 3.47	142.40 \pm 4.78	237.80 \pm 4.06	226.60 \pm 4.88	206.00 \pm 5.18	173.20 \pm 2.47	156.80 \pm 5.50
II	83.00 \pm 5.87	131.40 \pm 4.34	225.40 \pm 5.88	203.60 \pm 11.45	188.00 \pm 10.99	162.80 \pm 6.58	145.20 \pm 4.53
III	81.40 \pm 3.23	134.20 \pm 3.89	227.20 \pm 7.25	215.20 \pm 7.80	194.40 \pm 7.71	173.40 \pm 7.39	157.80 \pm 4.63
IV	88.80 \pm 5.02	145.40 \pm 1.88	239.80 \pm 3.30	218.40 \pm 2.22	198.60 \pm 2.06	170.80 \pm 3.77	164.00 \pm 6.41
V	96.20 \pm 4.59	148.80 \pm 3.42	213.20 \pm 17.67	221.00 \pm 4.51	205.60 \pm 3.72	178.40 \pm 4.62	164.20 \pm 5.38

Values are mean \pm SEM; n = 5 in each group, One way ANOVA followed by Dunnett's test.

Figure 3: Effect of *Randia dumetorum* Lam on Haloperidol induced catalepsy in mice.

One way ANOVA followed by Dunnett's test

Randia dumetorum Lam extract did not inhibit Haloperidol induced catalepsy in mice.

Discussion

Several drugs are known to induce catalepsy in animals. The neuroleptic agents induce catalepsy by inhibiting dopamine D₂ receptors in the substantia nigra⁶. Chopra and Dandiya (1975) have studied the relative role of acetylcholine and histamine in perphenazine-induced catalepsy and suggested that anticholinergic activity of antidepressant might be due to an increase in dopamine content in brain or their ability to inhibit release of acetylcholine¹⁴. They also showed that different stages of catalepsy appear to be directly correlated with brain histamine content. Uvnas (1969) studied the mast cell degranulation and in correlation with the release of histamine after administration of mast cell degranulating agent (compound 48/80)¹⁵. Lakadwala et al., (1980) have shown that clonidine releases histamine from mast cells in a similar manner to a selective liberator like compound 48/80⁴. Pretreatment with l-histidine, a precursor of histamine potentiated clonidine-induced catalepsy in dose-dependent manner².

Typical neuroleptic-induced catalepsy has been linked to a blockade of post-synaptic striatal dopamine D₁ and D₂ receptors^{6,11}. Despite this evidence, dysfunction of several other neurotransmitters such as acetylcholine¹⁶, GABA¹⁷ and serotonin¹⁸, have also been implicated. In addition to dysfunction of various neurotransmitters in catalepsy, many clinical and preclinical studies have suggested the involvement of reactive oxygen species in haloperidol induced toxicity^{19,20}. Blockade of α_2 -adrenoceptors may alleviate the extrapyramidal effects of neuroleptic agents^{21,22}. Antagonists at α_2 -adrenoceptors facilitate dopamine (DA) transmission as shown by the fact that they enhance the effect of D-amphetamine on locomotor activity²³ and the ipsilateral rotation induced by D-amphetamine in unilateral substantia nigra-lesioned rats²⁴.

Serotonin (5-HT) appears to have complex role in neuroleptic-induced catalepsy. This catalepsy is reduced after stimulation of 5-HT_{1A} and 5-HT_{2A} receptors^{25,26,27,28} and blockade of 5-HT_{2C} receptors²⁹.

In our study, the *Randia dumetorum* Lam fruit extract in all the doses tested showed a significant reduction in the cataleptic scores. The effects were evident till 120 min of observation.

The observation of this study indicated that the ethanolic extract of *Randia dumetorum* Lam inhibited clonidine-induced catalepsy and not inhibited haloperidol-induced catalepsy. From the present study we can conclude that the cataleptic effect of clonidine in the mouse is mediated by histamine release from mast cells and the ethanolic extract of *Randia dumetorum* Lam fruits has antihistaminic activity. The effect of this extract on clonidine-induced catalepsy is probably due to mast cell stabilizing property. However it does not possess antidopaminergic and antiserotonergic activity.

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