

# Mast cell stabilizing effect of hydroalcoholic extract of *Jasminum sambac* leaves against compound 48/80-induced degranulation

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## Abstract

**Introduction:** Asthma is chronic inflammatory disease characterized by swollen airways, obstruction in respiration, etc. On exposure to the allergens, histamine is released from mast cells, which after activating H<sub>1</sub> receptor causes contraction of the airways. An herbal approach which can inhibit the degranulation of mast cell can serve as safer and effective treatment for the management of asthma. **Objective:** The present experiment is designed with objectives to determine the physicochemical properties of *Jasminum sambac* leaves, perform acute oral toxicity studies as per the OECD 420 guideline, and assess the mast cell stabilizing effect of hydroalcoholic (50:50) extract of the plant. **Results:** Physicochemical parameters revealed the standard quality of plant leaves. Phytochemical evaluation showed the presence of phytoconstituents such as flavonoids, glycosides, steroids, saponins, tannins, terpenoids, and phenols. Hydroalcoholic (50:50) extract of *J. sambac* leaves showed no signs of toxicity at the dose of 2000 mg/kg body weight. The lethal dose of the drug is above 2000 mg/kg body weight. The extract showed significant ( $P < 0.05$ ) inhibition of mast cell degranulation when compared to negative control. The percentage of degranulated mast cells was calculated and found to be  $51.33 \pm 3.01^*$  and  $43.0 \pm 2.09^*$  ( $P < 0.05$ ), at doses of 25 mg/ml and 50 mg/ml, respectively. **Conclusion:** It can be concluded that *Jasminum sambac* may serve as the choice of treatment in the management of asthma. The rich phytochemical profile of the plant leaves might be responsible for its mast cell stabilizing action. Isolation of bioactive molecule can be suggested for the further studies.

**Key words:** Asthma, histamine, *Jasminum sambac*, mast cell degranulation

## INTRODUCTION

Asthma is a multifactorial, chronic inflammatory disorder of airways. It is characterized by airway hyperresponsiveness, obstruction due to the inflamed mucous membrane, hyperproduction of mucous, persistent inflammation and infiltration of the airways with airway wall remodeling, and histological changes. Airway remodeling, characterized by thickening of the airway wall, can have profound consequences for the mechanics of airway narrowing and can contribute to the chronic progression of the disease.<sup>[1-5]</sup>

Allergic asthma is a clinical syndrome characterized by T helper cell (Th)1/Th2 imbalance.<sup>[6]</sup> Histamine is stored in granules within mast cells and basophils, where it is

closely associated with the anionic proteoglycans heparin (in mast cells) and chondroitin-4-sulfate (in basophils). From a study, it was evident that bronchoconstriction was one of the first recognized effects of histamine. Inhaled or intravenously administered histamine causes bronchoconstriction, which is inhibited by H<sub>1</sub> receptor antagonists. Histamine contracts both central and peripheral airways *in vitro*, with a more potent effect on peripheral airways.

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Mast cells when exposed to the allergens undergo degranulation which leads to massive release of histamine, which predominantly activates H<sub>1</sub> receptors on airway smooth muscles and endothelial cells. Agents, which can substantially inhibit the degranulation of mast cells and ultimately the release of histamine and/or can antagonize the effect of histamine at H<sub>1</sub> receptor to reduce its inflammatory response, may serve as a useful means in the management of asthma. Synthetic drugs having such properties are known to have a large number of side effect; therefore, the use of herbal medicines becomes the choice of treatment which has proven to be safe and free from side effects.<sup>[7]</sup>

*Jasminum sambac* Linn. (Oleaceae) is commonly known as Jasmine. Conventionally, *J. sambac* was used in the treatment for various illnesses such as rheumatism, paralysis, gallstones, and diabetes mellitus.

Different parts of the plant have been reported to exhibit various pharmacological properties. Essential oil of *J. sambac* is used as a fragrance for skin care products as it tones the skin as well as reduces skin inflammation.<sup>[8]</sup> From the literature, it was revealed that *J. sambac* plant exhibits antifungal<sup>[9]</sup> and anticancer<sup>[10,11]</sup> properties. Essential oil and methanol extract of various parts of *J. sambac* have *in vitro* antimicrobial and antioxidant activities which support the use of the plant by traditional healers to treat various infective diseases.<sup>[8]</sup> Ethyl acetate and water extract of leaves of *J. sambac* showed a reduction in plasma glucose level, lipid profile, and serum urea in diabetic rats.<sup>[12,13]</sup>

Survey on previous research done manifests the versatile pharmacological and phytochemical profile of *J. sambac* Linn. The present study is designed with an objective to standardize the leaves, to perform acute oral toxicity study, and to evaluate the mast cell stabilization activity against 48/80 mast cell activating compound of the hydroalcoholic extract (50:50) of the leaves of *J. sambac*.

## MATERIALS AND METHOD

### Chemicals

The chemicals used in this study were purchased from different suppliers. Ferric chloride, sodium hydroxide, sodium chloride, glucose, sodium bicarbonate, potassium chloride, sodium dihydrogen phosphate, gelatin, magnesium chloride, bovine serum albumin, HEPES, and compound 48/80 were purchased from Sigma-Aldrich. Ketotifen fumarate, ethanol, Mayer's reagent, Dragendorff's reagent, sulfuric acid, ammonia, chloroform, lead ethanoate, acetic acid, hydrochloric acid, Millon's reagent, Molisch's reagent, and magnesium chips were purchased from Merck.

### Plant Material

#### Collection and authentication of plant material

The fresh leaves of *J. sambac* were collected from the local nursery of Bilaspur region in the month of October–November. The plant was authenticated by Dr. V. Rama Rao, Research Officer (Scientist-II), Regional Ayurveda Research Institute for Metabolic Disorders, Bengaluru, Ref. No.: RRCBJ-mus150.

#### Extraction of plant material

The leaves were washed with water to remove any soil or dirt adhering to it. It was then dried in shade, and the completely dried material was reduced to a coarse powder with the help of a mechanical grinder. The powder was labeled and stored in an airtight container for further use. The powdered plant material was extracted using distilled water:ethanol (50:50). Briefly, 100 g of powdered material was macerated with 500 ml of solvent for 48 h with subsequent filtration. The solvent from extract was evaporated to dryness using a rotary evaporator (BUCHI, Switzerland) at 40°C temperature and reduced pressure. The dried extract was weighed, and the percentage yield was calculated. The crude extract was dried in a freeze drier and preserved at +4°C for further studies.<sup>[14]</sup>

### Physicochemical Parameters of *J. sambac* Leaves<sup>[15]</sup>

#### Determination of moisture content (loss of drying)

About 1 g dried powder of the leaves was weighed and placed in Petridish. It was dried in an oven at 105°C for 1 h and cooled in a desiccator; loss in weight was recorded as moisture content.

#### Determination of ash values

The residue of the crude drugs after incineration contains mostly inorganic salts known as ash. Ash values are used to determine the quality and purity of a crude drug. Ash contains inorganic radicals such as phosphates, carbonates, and silicates of sodium, potassium, magnesium, and calcium, and sometimes, inorganic variables such as calcium oxalate, silica, and carbonate content of the crude drug affect “total ash.” Such variable is then removed by treating with acid, and then, acid insoluble ash value is determined.

#### Total ash

Carbon and organic matter present in the drug are converted to ash at high temperature. Accurately 2 g of coarse powder was weighed in a tarred silica crucible. The drug is incinerated by gradually increasing the heat in a muffle furnace up to 450°C for a few hours. After complete incineration, it was kept in a desiccator. The weight of ash with silica crucible was noted. Then, the percentage of total ash was calculated,

at a temperature of 450°C. It mostly contains carbonates, phosphates, silicates, and silica.

#### **Acid-insoluble ash**

Total ash may be treated with HCl, which removes many inorganic salts to yield mainly silica in the residue of acid insoluble ash. Using 25 ml of dilute hydrochloric acid, total ash was washed in a 100 ml beaker. The solution was boiled for 5 min. Filtered through an ashless filter paper, the residue was washed twice with hot water. Filter paper was placed in a silica crucible and incinerated by gradually increasing the heat in a muffle furnace to 450°C for 2 h. After complete incineration, it was kept in a desiccator to cool. The weight of acid insoluble ash with silica crucible was noted. Then, the percentage of acid-insoluble ash was calculated.

#### **Water-soluble ash**

It is produced by separating the water-soluble material from the total ash. In this case, most of the water-insoluble salts which might contribute in total ash were removed to find water-soluble ash content. This was determined in a similar way to acid-insoluble ash, using 25 ml of water in place of HCl.

#### **Determination of extractive values of *J. sambac***

Extractive value is useful for the evaluation of a crude drug as it gives an idea about the nature of the chemical constituents present in a crude drug and is also useful for the estimation of chemical constituents, soluble in that particular solvent used for extraction.

#### **Determination of alcohol-soluble extractive values of *J. sambac***

About 5 g of crude powder was weighed and transferred to a conical flask. 100 ml of the 90% ethyl alcohol was added and closed with the cork. It was kept aside for 24 h with frequent shaking. It was filtered, and 25 ml of the filtrate was collected and transferred to a previously weighed, thin porcelain dish. It was evaporated to dryness on a water bath and dried completely in an oven at 90°C kept in a desiccator to cool. The percentage w/w of extractive with reference to the air-dried drug was calculated.

#### **Determination of water-soluble extractive value of *J. sambac***

A procedure similar to that mentioned in the determination of alcohol-soluble extractive values was followed using chloroform:water (1:99) which was used instead of alcohol.

#### **Determination of foaming index**

Weigh accurately 1 g of coarse powder of the plant material and transfer to a 500-ml conical flask containing 100 ml of boiling water. Maintain at moderate boiling for 30 min. Cool and filter into a 100-ml volumetric flask and add

sufficient water through the filter to dilute to volume. Pour the decoction into 10 stoppered test tubes (height 16 cm and diameter 16 mm) in successive portions of 1 ml, 2 ml, 3 ml, and up to 10 ml, and adjust the volume of the liquid in each tube with water to 10 ml. Stopper the tubes and shake them in a lengthwise motion for 15 s, two shakes per second. Allow to stand for 15 min and measure the height of the foam.

#### **Determination of swelling index**

Take accurately weighed 1 g of plant material into a 25-ml glass-stoppered measuring cylinder. 25 ml of water is then added to it and mixture is shaken well after every 10 min for 1 h. The mixture is then allowed to stand for 3 h. The volume in ml occupied by the plant material was measured.

## **PHYTOCHEMICAL SCREENING**

The hydroalcoholic (50:50) extract was subjected to qualitative phytochemical analysis by standard methods to detect the presence of various phytoconstituents in them.<sup>[16-18]</sup> The test solution was prepared by dissolving 3 g of extract in the solvent and filtering the solution through Whatman No. 1 filter paper.

### **Alkaloids**

#### **Mayer's test**

To the extract solution, two drops of Mayer's reagent was added from the sides of the test tube. Formation of a white precipitate indicated the presence of alkaloids.

#### **Dragendorff's test**

1–2 ml Dragendorff's reagent was added to 1 ml of extract solution. Formation of a yellow precipitate showed the presence of alkaloids.

### **Anthraquinones**

#### **Borntrager's test**

Diluted H<sub>2</sub>SO<sub>4</sub> was added to the extract solution in a test tube, and the mixture was boiled. The resulting solution was filtered, and chloroform was added and shaken well in a separating funnel. To the organic layer, ammonia solution was added slowly. The appearance of pink/red/violet color in the lower layer (ammoniacal) indicated the presence of anthraquinones.

### **Carbohydrates**

#### **Molisch's test**

To the extract solution taken in a test tube, few drops of Molisch's reagent were added to it. Further, 1 ml of concentrated H<sub>2</sub>SO<sub>4</sub> was added from the side of the test

tube. After 2–3 min, 3 ml of distilled water was added to it. The appearance of a red or pale violet color at the interface indicated the presence of carbohydrates.

## Flavonoids

### *FeCl<sub>3</sub> test*

1 ml extract solution was taken in a test tube, and few drops of 10% FeCl<sub>3</sub> solution were added. Formation of a greenish-blue or violet color indicated the presence of flavonoids.

### *Lead ethanoate test*

3 ml extract solution was treated with equal volume of lead ethanoate solution. Formation of a buff-colored precipitate indicated the presence of flavonoids.

### *Sodium hydroxide test*

1 ml extract solution was taken in a test tube and 2 ml of 10% NaOH solution was added to it. The yellow color of the solution disappeared on the addition of dilute HCl which indicated the presence of flavonoids.

### *Shinoda's test*

To the extract, 1 ml of ethanol was added. The solution was warmed and filtered. To the filtrate, 2–3 pieces of magnesium chips were added. Then, few drops of concentrated HCl were added to it. Formation of pink/orange/red to violet color indicated the presence of flavonoids.

## Saponins

### *Foam test*

Extract solution was diluted with 2 ml distilled water, shaken, and kept aside for 2–3 min. Persistent frothing on warming the solution indicated the presence of saponins.

## Steroids

### *Libermann–Burchard's test*

3 ml acetic acid was added to the extract solution. The reaction was cooled by keeping the test tube in ice, and then, 1 ml of concentrated H<sub>2</sub>SO<sub>4</sub> was added to the solution. The appearance of a violet to blue or bluish-green color indicated the presence of steroids.

## Terpenoids

### *Salkowski test*

5 ml of extract solution was mixed in 2 ml of chloroform, and 3 ml of concentrated H<sub>2</sub>SO<sub>4</sub> was carefully added to form a layer. Formation of a reddish-brown color at the interface showed the presence of terpenoids.

## Tannins

### *Ferric chloride test*

To 2 ml extract solution, 1 ml 10% FeCl<sub>3</sub> was added. Formation of a bluish-black or bluish-green precipitate indicated the presence of tannins.

### *Glycosides*

To the extract solution, 2 ml of distilled water was added. To this 1 ml, diluted NaOH solution was added. Formation of a yellow color confirmed the presence of glycosides.

### *Phenols*

To 1 ml of extract solution, 2 ml of distilled water was added followed by addition of a few drops of 10% FeCl<sub>3</sub> solution. Formation of a blue or green color indicated the presence of phenols.

### *Proteins*

To the extract solution, 5–6 drops of Millon's reagent was added. Formation of a precipitate which turned red on heating indicated the presence of proteins.

## ACUTE TOXICITY STUDY

### Animals

A total of 10 healthy young adult Swiss Albino mice non-pregnant and nulliparous, weighing between 25 and 30 g (8–12-week-old) were used for the experiment. The experimental protocol was approved by the Institutional Animal Ethical Committee as per the guidance of CPCSEA, Ministry of Social Justice and Empowerment, Government of India (IAEC No. 1275/PO/Re/09/CPCSEA).

### Housing and Feeding Conditions

The animals were housed in standard conditions of temperature (22 ± 2°C), relative humidity (55 ± 5%), and light (12 h light/dark cycles). They were fed with commercial rat feed and water ad libitum.

### Preparation of Animals

The animals were randomly selected, marked appropriately for identification, and kept in clean propylene cages for 7 days before the experiment to allow them for acclimatization to the laboratory conditions. Before dosing, animals were fasted (food but not water was withheld overnight). Following fasting, the animals were weighed again and the dose was calculated according to their body weights. After the dose was administered, food was withheld for 4 h.



**Table 1:** Dose of the extract and frequency of administration

Extract dose	Vehicle	Route of administration	Frequency of administration
Aqueous ethanol (50:50), 2000 mg/kg	Distilled water	Oral route using an intubation cannula	Single dose

**Table 2:** Results of physiochemical evaluation of leaves of *J. sambac*

Parameters	Value (%)
Total ash	10.7
Water-soluble ash	7.5
Acid-insoluble ash	7.8
Alcohol-soluble extractive value	28
Water-soluble extractive value	13.5
Moisture content	6.8
Crude fiber content	12.8
Swelling index	1
Foaming index	<100

*J. sambac*: *Jasminum sambac*

### Preparation of Doses

The extract was dissolved in distilled water and filtered through Whatman No. 1 filter paper. The dose of the extract was prepared shortly before the administration.

### Administration of Dose

Animals were divided into two groups:

- Group I: The control group ( $n = 5$ ) was given distilled water.
- Group II: Test group ( $n = 5$ ) was given a single dose of 2000 mg/kg of 50:50 aqueous ethanolic extract of *J. sambac*, through oral gavage with the help of an intubation cannula [Table 1].<sup>[19-21]</sup>

### Observations

Animals were observed individually at least once during the first 30 min after dosing and periodically during the first 24 h with special attention given during the first 4 h. Animals were observed daily for 2 weeks from the commencement of the experiment. Observations included changes in skin and fur, eyes and mucous membranes, and behavioral pattern, and attention was given to observations of tremors, convulsions, salivation, diarrhea, lethargy, sleep, coma, and mortality. All the observations of individual animals were systematically recorded and compared with that of control animals.

### Body Weight

Individual weights of the animals were determined before the administration of the test extract. All the animals were

reweighed on the 7<sup>th</sup> and 14<sup>th</sup> days of the experiment, and changes in their weights were recorded.

## MAST CELL DEGRANULATION

### Isolation of Mast Cells

The animals were anesthetized using ether, and 20 ml of Tyrod Buffer B (137 mM NaCl, 5.6 mM glucose, 12 mM NaH [CO<sub>3</sub>], 2.7 mM KCl, 0.3 mM Na[H<sub>2</sub>][PO<sub>4</sub>], and 0.1% gelatin), was injected in the peritoneal cavity. The abdomen was gently massaged and the cavity was carefully opened. With the help of a Pasteur pipette, the peritoneal fluid containing the peritoneal cells was aspirated. The fluid was centrifuged at 150 rpm for 10 min at room temperature and resuspended in Tyrod buffer B. It was again centrifuged at room temperature for 15 min at 400 rpm. The cells in the pellet were washed and resuspended in the Tyrod buffer A (10 mM HEPES, 130 mM NaCl, 5 mM KCl, 1.4 mM Ca[Cl<sub>2</sub>], 1 mM Mg[Cl<sub>2</sub>], 5.6 mM glucose, and 0.1% BSA) containing calcium.<sup>[22]</sup>

### Methodology

Mast cells were isolated from peritoneal fluid extracted from six mice and were divided into five respective groups. The experiment was performed in such a manner to make six replicas of each group.

- Group I (normal control): 0.1 ml mast cell suspension.
- Group II (negative control): 0.1 ml mast cell suspension + 0.1 ml of 2µg/ml compound 48/80
- Group III (positive control): Group II + 10 µl/ml ketotifen fumarate
- Group IV (test dose 1): Group II + 0.1 ml of 25 mg/ml extract in normal saline
- Group V (test dose 2): Group II + 0.1 ml of 50 mg/ml extract in normal saline.

All the preparations mentioned above were subjected to incubation, and two to three drops of the suspensions from each test tubes were taken on a glass slide and fixed using a solution of formaldehyde and methanol (1:3). The mast cells were then stained with toluidine blue (0.1%) and slides were observed under a microscope at 40×. The number of degranulated mast cells was counted on each slide and percentage of degranulated mast cells was calculated using the following formula: <sup>[23,24]</sup>

$$\% \text{ of degranulated mast cells} = \frac{(1 - \text{no. of degranulated mast cells})}{\text{Total no. of mast cells}} \times 100$$

**Table 3:** Physical characters and the percentage yield of the extract

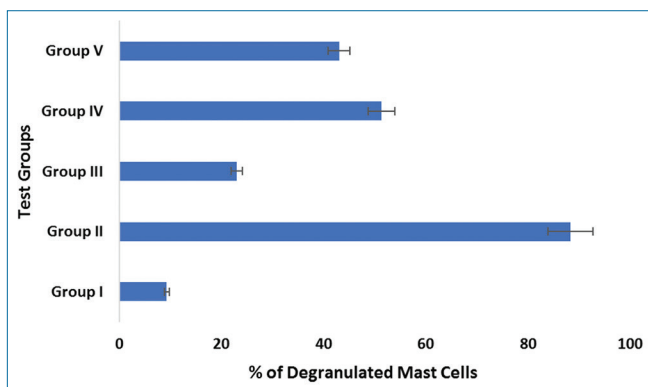
Extract	Consistency	% yield	Color under visible light
Hydroalcoholic Extract of <i>J. sambac</i> leaves (50:50)	Semisolid	21.8%	Dark greenish yellow

*J. sambac*: *Jasminum sambac*

**Table 4:** Phytochemical screening of the extract

Constituents	Test	Inference
Alkaloids	Dragendorff's	-
	Mayer's	-
Anthraquinones	Borntrager's	-
Carbohydrates	Molisch's	+
Flavonoids	Ferric chloride	+
	Lead ethanoate	+
	Sodium hydroxide	+
	Shinoda's	+
Saponins	Foam	+
Steroids	Libermann-Burchard's	+
Tannins	Ferric Chloride	+
Terpenoids	Salkowski	+
Glycosides	Diluted NaOH	+
Proteins	Millon's reagent	+
Phenols	Ferric chloride	+

+: Present, -: Absent



**Figure 1:** Comparison of % of mast cell degranulation between test groups

### Statistical Analysis

The data were analyzed using the software GraphPad Prism (7.03). The results were expressed as mean  $\pm$  standard deviation. One-way ANOVA followed by Tukey's multiple comparison tests was used to analyze the significance between the groups, and  $P < 0.05$  was considered to be statistically significant.

## RESULTS

Total ash value of plant material indicated that the number of minerals and earthy material attached to the plant material

and its value was calculated to be 10.7%w/w. The amount of the acid-insoluble siliceous matter present in the plant was 7.8%w/w. The water-soluble extractive value indicated the presence of sugar, acids, and inorganic compounds. The alcohol-soluble extractive values indicated the presence of polar constituents and its value was found to be 28%w/w. The value for loss on drying was found to be 6.8%w/w; the lesser value of moisture content could prevent bacterial, fungal, and yeast growth. The swelling index was found to be  $<1$  and signifies the presence of trace amount or no mucilage. The foaming index was calculated  $<100$  [Table 2].

The leaves of *J. sambac* were extracted with aqueous ethanol (50:50) using cold percolation method, and the percentage yield is given in Table 3.

Phytochemical screening of aqueous ethanolic (50:50) extract of *J. sambac* leaves revealed the presence of various phytochemical constituents such as flavonoids, glycosides, steroids, saponins, tannins, terpenoids, and phenols, whereas alkaloids and anthraquinones remained absent. The result of phytochemical analysis of the extract is presented in Table 4.

### Mortality and Lethal Dose (LD<sub>50</sub>) Value

There was no mortality reported in control as well as in test group at the limit dose of 2000 mg/kg body weight. The LD<sub>50</sub> value of *J. sambac* extract was found to be more than 2000 mg/kg body weight.

### Wellness Parameters and Body Weight

There were no sign of toxicity and no significant changes observed in wellness parameters of control and treated groups during the course of study. The observations on different parameters in the control and test group are noted and presented in Table 5. The changes in the body weight of the animals were also observed before dosing of extract and thereafter on the 7<sup>th</sup> and 14<sup>th</sup> day of study [Table 6], and there was no significant difference found which might be attributed by animals' normal metabolic functions.

The aqueous ethanolic (50:50) extract of *J. sambac* significantly ( $P < 0.05$ ) inhibited the degranulation of peritoneal mast cells induced by mast cell activator compound 48/80. The result of percentage of degranulated mast cells is shown in Table 7 and graphically represented in Figure 1.

**Table 5:** Observations after dosing in control and test groups

Observations	30 min		4 h		12 h		24 h		48 h		7 <sup>th</sup> day		14 <sup>th</sup> day	
	C	T	C	T	C	T	C	T	C	T	C	T	C	T
Skin and fur	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Eyes	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Mucous membranes	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Salivation	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Lethargy	-	-	-	-	-	-	-	-	-	-	-	-	-	-
Sleep	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Coma	-	-	-	-	-	-	-	-	-	-	-	-	-	-
Tremors	-	-	-	-	-	-	-	-	-	-	-	-	-	-
Convulsion	-	-	-	-	-	-	-	-	-	-	-	-	-	-
Diarrhea	-	-	-	-	-	-	-	-	-	-	-	-	-	-
Mortality	-	-	-	-	-	-	-	-	-	-	-	-	-	-

C: Control group, T: Test group, +: Normal, -: NIL

**Table 6:** Effect of treatment with extract on the body weight of animals (dose 2000 mg/kg) on day 1 (before treatment) and day 14 (after treatment)

Group	Treatment	Body weight (g)	
		Day 1	Day 14
Control	Distilled water	26.74±0.84	27.78±0.87 <sup>a</sup>
Treated	Aqueous ethanolic (50:50) extract	25.86±1.05	26.66±0.99 <sup>b</sup>

Values were expressed as mean±SD, (n=5), a, b: No significant difference at  $P < 0.05$ . SD: Standard deviation

**Table 7:** Percentage inhibition of mast cell degranulations

Treatment	Concentration standard/extract	% of degranulated mast cells
Group I	-	9.33±3.82
Group II	-	88.33±1.72
Group III	10µl/ml	23.0±2.60*
Group IV	25 mg/ml	51.33±3.01*
Group V	50 mg/ml	43.0±2.09*

Values represented as mean±SD, n=6, \*( $P < 0.05$ ) was considered statistically significant

## DISCUSSION

Physiochemical evaluation of the leaves of *J. sambac* was done to standardize plant and was found free from adulterant. The aqueous ethanolic (50:50) extract of the plant was safer at the dose regimen of 2000 mg/kg body weight. The LD<sub>50</sub> of the extract was found more than 2000 mg/kg BW. Mast cell degranulation caused by the compound 48/80 is inhibited by the extract significantly ( $*P < 0.05$ ). Stabilization of mast cells inhibits the release of various inflammatory mediators like histamine and thus provides a protection against histamine-mediated airway inflammation and hyperresponsiveness. The extract may find its usage for the management of asthma.<sup>[25,26]</sup>

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