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# Article

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# Antiasthmatic and antiallergic potential of methanolic extract of leaves of *Ailanthus* excelsa

Dinesh Kumar,\*,1 Z. A. Bhat,1 P. Singh,1 V. Khatanglakar,2 S.S. Bhujbal2

<sup>1</sup>Department of Pharmaceutical Sciences, University of Kashmir, India, <sup>2</sup>Padam Shree Dr. D. Y. Patil Institute of Pharmaceutical Science and Research, India

Abstract: The aim of study was antiasthmatic potential of methanolic extract of leaves of Ailanthus excelsa Roxb., Simaroubaceae. Traditionally or in Indian system of medicine, A. excelsa is used in the treatment of asthma, cough, colic pain, cancer, diabetes and also used as antispasmodic, antifertility, bronchodilator. Stem bark of A. excelsa already reported for its potential against asthma. The pollens of Ailanthus excelsa reported allergic in nature and the time of collection of leaves were important in this study, generally the flowering stage of plant was avoided for the collection due to maximum chance of pollens at that time. Methanolic extract of leaves of A. excelsa was evaluated using in vitro goat tracheal chain preparation model and in vivo- Milk induced leucocytosis, eosinophilia, Clonidine induced catalepsy in mice model while Passive paw anaphylaxis and Clonidine induced mast cell degranulation in rat model. The extract showed the presence of flavonoids, terpenoids, saponins, quassonoids and test was also positive for alkaloids and steroids. The extract also showed the presence of guercetin which is flavonoid and detected on the preparative TLC plate with the help of standard quercetin. Dose response studies of methanolic extract of leaves of A. excelsa Roxb. were conducted at 100 μg mL<sup>-1</sup> in vitro and 100, 200, 400 mg kg<sup>-1</sup> p.o. in vivo models. The treatment with methanolic extract of A. excelsa at different dose level showed the significant (\*p<0.05, \*\*p<0.01, \*\*\*p<0.001) antiasthmatic activity. Inhibition or decrease the release of inflammatory mediators potentiates the antiasthmatic as well as antiallergic activity of methanolic extract of leaves of A. excelsa.

## Introduction

The use of medicinal plants for the treatment of human diseases has increased considerably worldwide. Evaluation of the effects of these plants on organs and systems has contributed to the development of the scientific basis for their therapeutic application, and also has enriched considerably the therapeutic arsenal for the treatment of a number of diseases (Elizabetsky, 1986).

Ailanthus excelsa Roxb. is a tree belonging to family Simaroubaceae, indigenous to central and southern India. Commonly it is known as a plant of Heaven. The traditional claims, phytochemical investigations, and pharmacological evaluation and some Ayurvedic formulations provide the backbone to make this tree as a plant of Heaven. This is not wrong to say that it is largactil because it has number of activities. In Indian system of medicine it is used in panic diarrhoea, bronchatitis and dysenteries (Nadkarni, 2000; Anonymous, 1996). In addition, Ailanthus excelsa was shown to have antipyretic activity (Suresh et al., 1995). Even though

A. excelsa was reported to be useful in a many ailments like stem bark of A. excelsa was potent antiasthmatic. bronchodilator (Kumar D et al., 2010a; Kumar D et al., 2010b; Kumar D et al., 2010c). From a pharmaceutical perspective flavonoids possess a remarkable spectrum of biochemical and pharmacological activities. The leaves were reported to contain different flavonoids like kaempferol (5,4',5,7-tetrahydroxy flavone), luteolin (3',4',5,7-tetrahydroxy flavone), apigenin (4',5,7trihydroxy flavone) (Lavhale & Mishra, 2007). Thus, drug development has been encouraging researchers to find strategies to treat allergic diseases and the medicinal plants have been the target of these studies and an important tool to treat immediate-type allergic response (Dai et al., 2004). Scientific evaluation of the leaves of plant was not reported by these models for its antiasthmatic and antiallergic activity. Hence, in the present study, we have evaluated the methanolic extract of leaves of Ailanthus excelsa Roxb. for its antiasthmatic as well as antiallergic activity using animal models.

#### **Materials and Methods**

Plant material

Leaves of Ailanthus excelsa Roxb.. Simaroubaceae, were collected in August 2008 from local area of Pimpri, pune-18 (India) and identified by the Regional Research Institute of Ayurveda Kothrude, Pune (India). A voucher specimen - 899 was authenticated. Leaves were washed with tap water superficially and dried under shade, powdered pass through 40 mesh sieve. The powdered material was extracted with methanol (95%) using Soxhlet apparatus. The extract obtained was dried in rotary vacuum evaporator, yielding a dark greenish brown colored powdery mass (10%). Phytochemical evaluation was done according to the method of Khandalwal (2003). TLC of quercetin was done according to the method of Wagner & Bladt (1996).

Animal

Isolated adult goat tracheal tissue, Albino mice and Albino rates (Wistar Strain) of either sex weighing 20-25 g and 150-200 g respectively were used for studies. Isolated adult goat trachea tissue was obtained immediately after slaughter of the animal. Pieces of the trachea were collected in the ice cold oxygenated Krebs solution. The albino mice and albino rats were obtained from animal house of National Toxicological Laboratory, Pune. They were housed in polypropylene cages with standard pellet chow and water *ad libitum*. In all experimental sets, five rats and five mice were used for each treatment.

Acute toxicity studies

Mice were selected for this study. They were divided into eight groups each containing six animals. Methanolic extract of leaves of *A. excelsa* was administered orally in varying doses (0.50, 0.75, 1.00, 1.25, 1.50, 1.75, 2.00, 2.50 and 5.00 g/kg) to these animals. They were continuously observed for 2 h to detect changes in the autonomic or behavioural responses like alertness, spontaneous activity, irritability, urination, etc. Any mortality during experimentation and the following seven days was also recorded. A group of animals treated with vehicle (distilled water) was served as control. Based on the results of preliminary toxicity testing the doses of 100, 200 and 400 mg/kg *p.o.* were chosen for further experiments (Kumar D et al., 2010c).

Antiasthmatic and antiallergic activity

In vitro method: isolated goat trachea chain preparation

Isolated adult goat tracheal tissue was obtained

immediately after slaughter house of the animals. Trachea was cut into individual rings and tied together in series to form a chain. Trachea was suspended in bath of Krebs solution and was continuously aerator at 37+0.5 °C. DRC of histamine in plane Krebs solution and in 100  $\mu$ g/mL *Ailanthus excelsa* extract in Krebs solution was taken. Graph of percentage of maximum contractile response on ordinate and concentration of histamine on abscissa was plotted to record dose response curve of histamine, in absence and in presence of drug extract (Kulshrestha et al., 1983; NagChaudhari et al., 1974).

*In vivo* method: Effect of test drug on leukocytosis, eosinophilia: (Brekhman, 1969; Vadnere et al., 2007).

Mice were divided into five groups, five animals in each group. Animals belonging to group-I received distilled water (DW) 10 mL/kg, (p.o.). Animals belonging to group II, III, IV, V received boiled and cooled milk injection in dose of 4 mL/kg, (s.c.). Animals belonging to groups III, IV and V received test extract of *Ailanthus excelsa* Roxb. in dose 100, 200 and 400 mg/kg, p.o. respectively, 1 h before milk injection. Blood samples were collected from each mouse from the retro orbital plexus, under light ether anaesthesia. Total leukocyte count and total eosinophilia count and was done in each group before drug administration and 24 h after milk injection. Difference in total leukocyte count and total eosinophilia count before and 24 h after drug administration was calculated.

Passive paw anaphylaxis in rats

Rats (Wistar) were given (s.c.) three doses of 100 µg of egg albumin adsorbed on 12 mg of aluminium hydroxide gel prepared in 0.5 mL of saline on 1st, 3rd, 5th day. On 10th day of sensitization blood was collected from the retro orbital plexus and collected blood was allowed to clot and the serum was separated by centrifugation at 1500 rpm. Animals were divided into five groups (n=5). Animals belonging to group I served as control and were administered only the vehicle (10 mL/kg p.o.). Animals belonging to groups III, IV & V, received three doses (100, 200, 400 mg/kg p.o.) respectively of Ailanthus excelsa extract. Animals of group II, as positive control/ standard group received dexamethasone (0.27 mg/kg p.o.). The animals were passively sensitized with 0.1 mL of the undiluted serum into the left hind paw of animals. The contra lateral paw received an equal volume of saline. Drug treatment was given 24 h after sensitization. Animals were challenged in the left hind paw with 10 µg of egg albumin in 0.1 mL of saline, and the paw inflammation was measured using a Plethasmometer. The difference in the reading prior to, and after antigen challenge represented the edema volume and the percent inhibition of volume was calculated by using the following formula.

Percent inhibition =  $1 - (V_{t} / V_{c}) \times 100$ 

 $V_t$  = Mean relative change in paw volume in test group;  $V_c$  =Mean relative change in paw volume in control group.

Prior drug treatment animals were sensitizes with serum. Next 24 h after drug treatment animals again challenged for 10 µg egg albumin and edema inhibition was calculated (Gokhale & Saraf, 2002; Pungle et al., 2003: Oliver et al., 2003).

# Clonidine induced catalepsy in mice:

Albino mice were divided into five groups (n=5). Control group received saline (10 mL/kg) and other groups received single dose of extract (100, 200, 400 mg/kg *p.o.* body weight) respectively. Chlorpheniramine maleate (10 mg/kg, *i.p.*) was used as standard. All the groups were received clonidine (1 mg/kg, *s.c.*) one h after the drug administration and the duration of catalepsy was measured at 15, 30, 60, 90, 120, 150 and 180 min (Ferre et al., 1990; Taur et al., 2007).

# Clonidine-induced mast cell degranulation:

Rats were divided into five groups, five animals in each group. Animals belonging to group-I received vehicles 5 mL/kg, (p.o.) Animals belonging to group-II received sodium cromoglycate 50 mg/kg, (i.p.). Animals belonging to group-III, IV and V received methanolic extract of Ailanthus excelsa Roxb. in dose (100, 200 and 400 mg/kg, p.o.) respectively. The treatment was continued for seven days. On day 7th, 2 h after the assigned treatment mast cells were collected from the peritoneal cavity. 10 mL of normal saline solution was injected into peritoneal cavity and abdomen was gently massaged for 90 s. The peritoneal cavity was carefully opened and the fluid containing mast cells was aspirated and collected in siliconised test tube containing 7 to 10 mL of RPMI-1640 Medium (pH 7.2-7.4). The mast cells were then washed thrice by centrifugation at low speed (400-500 rpm) and the pallet of mast cells were taken in the medium. The mast cells suspension approximately (1 x 10 6 cells/mL) was challenged with 0.5 µg/mL of clonidine solution and stained with 1% toluidine blue and observed under high power microscope field (400x). Total 100 cells were counted from different visual areas and the number of intact and degranulated cells was counted. The percent protection was calculated (Lakadwala et al., 1980).

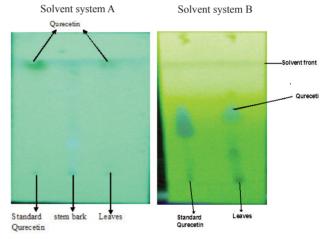
Statistical analysis

The statistical analysis was performed by using one-way analysis-of-variance (ANOVA) Followed by Dennett's test for individual comparison of groups with control.

#### Results

# Phytochemical investigation

The crude extract showed positive test for coumarin, flavonoids, quassinoids, terpenoids, alkaloids, steroids and saponins. The flavonoid as quercetin was identified with the help of standard quercetin on preparative TLC plates (Merck) in solvent system A & B (Solvent system A: ethyl acetate:glacial acetic acid:formic acid:water; 100:11:11:26; Solvent system B: ethyl acetate:glacial acetic acid:formic acid:water; 100:11:11:10). The spot detected under short wavelength in UV chamber. (Figure 1). Quercetin is one of the important constituent which helps in the treatment of asthma, allergy and many more allergic diseases.



**Figure 1.** Identification of quercetin on *Ailanthus excelsa* Roxb., Simaroubaceae, by TLC (solvent system A: ethyl acetate:glacial acetic acid:formic acid:water; 100:11:11:26; Solvent system B: ethyl acetate:glacial acetic acid:formic acid:water; 100:11:11:10.

In vitro method: effect of methanolic extract of leaves of Ailanthus excelsa Roxb. (AELM) on histamine induced contraction in goat tracheal chain preparation

In the present study, it was observed that *Ailanthus excelsa* inhibits the contraction produced by histamine in these tissue preparations. Histamine (10  $\mu$ g/mL) was taken in different dose level and DRC was plotted in absence and in presence of *Ailanthus excelsa* extract. Study showed that *Ailanthus excelsa* extract inhibits significantly (\*p<0.05, \*\*p<0.01, \*\*\*p<0.001) percentage contraction at concentration 100  $\mu$ g/mL in

goat tracheal chain preparation. Dose dependent response relationship was seen (Table 1).

Effect of AELM on clonidine induced catalepsy in mice

Clonidine (1 mg/kg, s.c.) produced catalepsy in mice, which remained for 2 h. The vehicle treated group showed maximum duration of catalepsy (100.2 $\pm$ 5.96 s) at 150 min after the administration clonidine. There was significant inhibition (\*p<0.05, \*\*p<0.01) of clonidine induced catalepsy in the animals pretreated with methanolic extract of Leaves of *Ailanthus excelsa* Roxb. (100, 200, 400 mg/kg, p.o.) and the duration of catalepsy was found to be 48.6 $\pm$ 4.74 at 150 min and 35.5 $\pm$ 4.59, 34 $\pm$ 1.61 s respectively at 120 min after the

administration clonidine. Chlorpheniramine maleate (10 mg/kg, i.p.) significantly inhibited (\*\*p<0.01) clonidine induced catalepsy in mice at 150 min after the administration clonidine (Table 2).

Effect of AELM on milk-induced leucocytosis in mice

Subcutaneous injection of milk at dose of 4 mL/kg produced a significant (\*\*\*p<0.001) increase in the leucocytes count and eosinophiles count after 24 h of its administration. In the groups of mice pretreated with methanolic extract of leaves of *Ailanthus excelsa* Roxb. at dose (100, 200 and 400 mg/kg, p.o.), there was significant (\*p<0.05, \*\*p<0.01) inhibition of milk-induced Leucocytosis and eosinophilia (Table 3 and 4).

Table 1. Effect of AELM on histamine induced contraction in goat tracheal chain prepatration

Sr. no.	D (10/I.) IV:	-Ve Log molar concentration of histamine	% Maxi	% Maximum Contraction	
	Dose (10μg/mL) Histamine conc.		Control	AELM (100 μg/mL)	
1	0.1	7.08	22.89±1.20	11.44±0.70*	
2	0.2	6.79	$30.72\pm1.26$	16.87±0.62*	
3	0.4	6.48	46.99±1.79	27.11±0.85*	
4	0.8	6.18	65.66±2.09	35.54±0.54**	
5	1.6	5.88	$75.30\pm1.25$	39.16±0.98***	
6	3.2	5.58	81.93±1.52	39.76±0.68***	
7	6.4	5.23	$100.0 \pm 1.25$	50.00±0.70	

n=5; Values are in mean±SEM; AELM: methanolic extract of leaves of *Ailanthus excelsa* Roxb; Control: D.R.C. of histamine in absence of *Ailanthus excelsa* Roxb. extract; AELM: D.R.C. of histamine in presence of methanolic extract of leaves of *Ailanthus excelsa* Roxb. (100 μg/mL); Statistical analysis done by using Student's 't'-test (\*p<0.05, \*\*p<0.01), \*\*\*p<0.001), significantly different from control.

Table 2. Effect of AELM on clonidine-induced catalepsy in mice.

C	Duration of catalepsy (s) at mean±SEM (min)							
Group	15	30	60	90	120	150	180	
Control	41.8±1.66	41.8±1.66	66.6±5.97	71.2±2.13	83.8±2.59	100.2±5.96	68.8±4.55	
Std. (CPM 10 mg/kg)	17.4±1.08**	17.4±1.08**	20.0±2.36**	23.6±3.54**	23.6±3.31**	13.0±2.78**	8.2±2.85**	
AELM 100	24.0±1.98**	39.6±3.39	35.8±2.48**	35.8±2.48**	45.6±6.48**	48.6±4.74**	38.8±3.59*	
AELM 200	26.8±2.76**	39.6±3.06	28.6±2.36**	28.6±2.36**	35.5±4.59**	30.8±3.28**	22.8±2.78**	
AELM 400	26.6±2.79**	32.2±2.67	33.6±1.99**	33.6±1.99**	34±1.61**	26.6±2.9**	24.6±3.11**	

n=5; Values are in mean±SEM, Control: distilled water (10 mL/kg, p.o.); Std: chlorpheniramine maleate (CPM 10 mg/kg, i.p.); AELM 100: methanolic extract of leaves of *Ailanthus excelsa* Roxb. (100 mg/kg, p.o.); AELM 200: methanolic extract of leaves of *Ailanthus excelsa* Roxb. (200 mg/kg, p.o.); AELM 400: ethanolic extract of leaves of *Ailanthus excelsa* Roxb. (400 mg/kg, p.o.): Statistical analysis done by ANOVA followed by Dunnett's test. \*p<0.05, \*\*p<0.01, compared to control group.

Table 3. Effect of AELM on milk-induced leucocytosis in mice.

Sr.no.	Dose	Difference in no. of leucocytes (per cu mm) (mean±SEM)
1	Control (Vehicle, 10 mL/kg, p.o.)	84±8.12
2	Intox. (milk 4 mL/kg)	4850±482.7***
3	AELM100	3600±279.28*
4	AELM200	2060±232.6**
5	AELM400	1630±127.08**

n=5; values are expressed in mean±SEM. Control: vehicle (10 mL/kg, p.o.); Intox: milk 4 mL/kg. AELM100: methanolic extract of leaves of *Ailanthus excelsa* Roxb. (100 mg/kg, p.o.); AELM200: methanolic extract of leaves of *Ailanthus excelsa* Roxb. (200 mg/kg, p.o.); AELM400: methanolic extract of leaves of *Ailanthus excelsa* Roxb. (400 mg/kg, p.o.). \*\*\*p<0.001, Intox. group compared with control group using student't, test and \*p<0.05, \*\*p<0.01, AELM compared to intox. Group using Statistical analysis done by ANOVA followed by Dunnet's test.

Effect of AELM on passive paw anaphylaxis in rats

Antiserum to egg albumin was injected 24 h before administration of the test drugs or standard. Egg albumin was injected after the administration of Ailanthus excelsa Roxb. and dexamethasone. In the vehicle treated group, egg albumin increased the paw volume in the sensitized animals, which was measurable up to the time period of 4 h. Pre-treatment with methanolic extract of leaves of Ailanthus excelsa Roxb. (100 mg/kg, p.o.) does not significantly reduced (\*\*p<0.01) the paw volume at 4<sup>th</sup> h was found to be reduced maximum i.e. 13.98%. Methanolic extract of leaves of Ailanthus excelsa Roxb. (200 and 400 mg/ kg, p.o.) significantly reduced (\*p<0.05, \*\*p<0.01) the paw volume at 4th h and the percentage inhibition was found to be 28.49% which was more than 50% of the standard and 42.47% approximately near to the standard drug. Dexamethasone (0.5 mg/kg, i.p.) significantly reduced (\*\*p<0.01) the paw volume at 4th h maximum and the percentage inhibition was found to be 46.77 % respectively (Table 5 and 6).

Effect of AELM on clonidine-induced mast cell degranulation in rats

Clonidine induced mast cell degranulation was significantly (\*\*p<0.01) inhibited by sodium cromoglycate (50 mg/kg, i.p.) and percent protection was found to be 74.47%. In the groups pre-treated with methanolic extract of leaves of *Ailanthus excelsa* Roxb (100, 200, 400 mg/kg, p.o) there was significant protection (\*\*p<0.01) of mast cells and the percent protection was 19.47, 38.95, and 55.00 % respectively (Table 7).

#### Discussion

The role of plants serving as purifiers of air has been known to us, since times immemorial. To cope with the gradually increasing levels of toxic pollutants, tree

Table 4. Effect of AELM on milk-induced eosinophilia in mice.

		*
Sr.no.	Dose	Difference in no. of eosinophils (per cu mm) (mean±SEM)
1	Control	21.8±3.17
2	Intox. (milk 4 mL/kg)	152.4±9.51***
3	AELM100	128.2±2.42*
4	AELM200	110±4.64**
5	AELM400	94.8±6.823**

n=5; values are expressed in mean±SE;. Control: vehicle (10 mL/kg, p.o.). Intox.: milk 4 mL/kg; AELM100: methanolic extract of leaves of *Ailanthus excelsa* Roxb. (100 mg/kg, p.o.); AELM200: methanolic extract of leaves of *Ailanthus excelsa* Roxb. (200 mg/kg, p.o.); AELM400: methanolic extract of leaves of *Ailanthus excelsa* Roxb. (400 mg/kg, p.o.). \*\*\*p<0.001; Intox. group compared with control group using student't, test and \*p<0.05, \*\*p<0.01, AELM compared to intox. Group using Statistical analysis done by ANOVA followed by Dunnet's test.

**Table 5.** Effect of AELM on passive paw anaphylaxis in rats.

	Groups		Paw edem	a volume (mL) (mear	n±SEM) (h)	
Sr.no.	Dose	0.5	1	2	3	4
1	Control	0.478±0.024	0.442±0.028	0.408±0.024	0.388±0.021	0.372±0.022
2	Std. (0.5 mg/kg, <i>i.p.</i> )	0.244±0.009**	0.256±0.009**	0.246±0.013**	0.226±0.008**	0.198±0.009**
3	AELM100	$0.442 \pm 0.026$	0.384±0.01661	$0.366 \pm 0.013$	0.352±0.014	$0.320\pm0.013$
4	AELM200	0.366±0.016**	0.336±0.016**	0.366±0.013**	0.288±0.015**	0.266±0.018**
5	AELM400	0.320±0.040**	0.294±0.014**	0.268±0.017**	0.244±0.016**	0.214±0.009**

n=5; values are expressed in mean±SEM; Control: distilled water (5 mL/kg, p.o.); Std.: dexamethasone (0.5 mg/kg, i.p.); AELM100: methanolic extract of leaves of *Ailanthus excelsa* Roxb. (100 mg/kg, p.o.); AELM200: methanolic extract of leaves of *Ailanthus excelsa* Roxb. (200 mg/kg, p.o.); AELM400: methanolic extract of leaves of *Ailanthus excelsa* Roxb. (400 mg/kg, p.o.). Std., AELM100, AELM200, AELM400 compared with Control (ANOVA followed by Dunnett's test), \*\*p< 0.01.

Table 6. Effect of AELM on passive paw anaphylaxis in rats.

Groups		Percentage inhibition of Paw Edema Volume (%) (h)					
Sr. no.	Dose	0.5	1	2	3	4	
1	Std. (0.5 mg/kg, <i>i.p.</i> )	48.95	42.08	39.71	41.75	46.77	
2	AELM100	7.53	13.12	10.29	9.28	13.98	
3	AELM200	23.43	23.98	10.29	25.77	28.49	
4	AELM400	33.05	33.48	34.31	37.11	42.47	

n=5; values are expressed in mean±SEM; Control: distilled water (5 mL/kg, p.o.); Std.: dexamethasone (0.5 mg/kg, i.p.); AELM100: methanolic extract of leaves of *Ailanthus excelsa* Roxb. (100 mg/kg, p.o.): AELM200: methanolic extract of leaves of *Ailanthus excelsa* Roxb. (200 mg/kg, p.o.): AELM400: methanolic extract of leaves of *Ailanthus excelsa* Roxb. (400 mg/kg, p.o.); Std., AELM100, AELM200, AELM400 compared with Control (ANOVA followed by Dunnett's test), \*\*p< 0.01.

Table 7. Effect of AELM on clonidine-induced mast cell degranulation in rats.

Carrie	Mast	Mast cells %		
Groups	Intact	Disrupted	- % protection	
Control	24.0±1.95	76±1.95	-	
Std.	80.6±1.68**	19.4±1.68**	74.47	
AELM100	38.8±2.59**	61.2±2.59**	19.47	
AELM200	53.6±2.01**	46.4±2.01**	38.95	
AELM400	65.8±1.18**	34.2±1.18**	55.00	

n=5; values are expressed in mean±SEM; Control: distilled water (5 mL/kg, p.o.); Std.: sodium cromoglycate (50 mg/kg, i.p.); AELM100: methanolic extract of leaves of *Ailanthus excelsa* Roxb. (100 mg/kg, p.o.); AELM200: methanolic extract of leaves of *Ailanthus excelsa* Roxb. (200 mg/kg, p.o.); AELM400: methanolic extract of leaves of *Ailanthus excelsa* Roxb. (400 mg/kg, p.o.); Std., AELM100 AELM200, AELM400 compared with Control (ANOVA followed by Dunnett's test), \*\*p< 0.01.

plantation programs have been undertaken in different countries of the world to help in environmental clean up. Ailanthus excelsa Roxb., Simaroubaceae, is one such exotic avenue tree, the plantation of which has been encouraged under social forestry programs for large-scale tree plantation in different densely populated cities and towns of India. Large areas of several districts are now covered with monoculture plantations of this exotic tree, as a result of which the pollen of this tree has emerged as a major contributor to pollen loads in these areas. Unfortunately, this has posed a real problem and a serious threat as an important aeroallergen to patients suffering from naso-bronchial disorders and sensitive to the pollen of A. excelsa. The allergenic potential of the pollen of A. excelsa in causing several respiratory disorders in sensitive patients has been reported earlier by several workers (Mondal et al., 2007). The leaves were washed with water superficially to remove the pollen grains which are a major factor of allergy and asthma or respiratory disorders. The time of collection was important, flowering time generally avoided for the collection of leaves because the pollens of Ailanthus excelsa were allergic in nature as mentioned above. Quercetin is a ubiquitous flavonoid found in a variety of foods, from raspberries and apples to onions and capers. Quercetin reduces LPS-mediated cytokine production through NF-kB and in particular in the IKB degradation pathway. This is similar in nature to the cascade suppression previously shown in curcumin and magnolol. Of interest, quercetin has also been shown to have antiangiogenic effects in vitro, as well as effects in preventing IL-1-mediated mast cell release of IL-6 without degranulation (Mainardi et al., 2009). Quercetin is the molecule which helps to control and prevent the inflammatory as well as allergic mediator and support to antiasthmatic activity. Allergy and anaphylaxis are the most responsible factor for diseases like asthma, rhinitis, bronchatitis, cold, cough, pain, inflammation etc. Allergic asthma is a chronic inflammatory process occurring due to exposure of allergen resulting in the activation of T-lymphocytes with subsequent release of inflammatory mediators. Immunomodulating agents are

useful in the treatment of allergy by virtue of inhibiting the antigen-antibody (AG: AB) reaction thereby inhibiting release of inflammatory mediators (Leeman et al., 1987). Administration of egg albumin (s.c.) to rat raises the antiserum to egg albumin in the plasma and sub plantar injection of plasma containing these antibodies, then challenged with egg albumin leads to passive paw anaphylaxis in rats (Pungle et al., 2003). The animals pre-treated with Ailanthus excelsa Roxb. extract showed significant reduction (\*\*p<0.01) in the paw volume at all the time intervals. The beneficial effect of Ailanthus excelsa Roxb. could be due to either inhibition AG: AB (hypersensitivity reaction- I) i.e. Producing antiallergic and anti-inflammatory properties. Mast cells are widely distributed in the connective tissue, with a preferential localization adjacent to small blood vessels. The mast cells contain basophile granules literally loaded with active substances which, if allowed to escape themselves or via enzymatically formed products, cause vascular and other tissue reactions similar to those characteristic of inflammatory processes (Uvnas, 1969). In the rat mast cell granules the histamine concentration has been calculated to be around 0.3 M. Both clonidine and compound 48/80 acts through the dynamic expulsion of granules without causing any damage to the cell wall. Clonidine releases histamine from mast cells in a similar manner to a selective liberator like compound 48/80 (Lakadwala et al., 1980). It is known that Sodium cromoglycate, a standard mast cell stabilizer prevents degranulation of mast cells by raising the cyclic adenosine monophosphate (Geetha et al., 1981). It has been known that all pharmacological agents that increase intracellular levels of cAMP relax airway smooth muscle and inhibit the release of autocoids from the tissue and basophiles. The groups of animals pretreated with methanolic extract of Ailanthus excelsa Roxb. resulted in significant reduction in degranulation of mast cells and offered significant protection when challenged with clonidine, indicating mast cell stabilizing activity and which showed the inhibition of allergic and inflammatory mediators. This plant was reported for many respiratory disorders and we have evaluated this plant leaves with

lot of considerations in mind about allergens and found with above mentioned model significant, it may be due to immunomodulatory action or combinatorial effect of phytoconstituents. Further study is required to carry out the isolation and their evaluation for the treatment of asthma in the form of combination of phytoconstituents as well as single molecule.

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## References

- Anonymous, 1996. Indian Pharmacopoeia. Government of India 2, Ministry of Health and Family Welfare, Controller of Publications, New Delhi, A-53-54, A-95-97, A-109.
- Brekhman II, Dardymov IV 1969. New substances of plant origin which increases non specific resistance. *Ann Rev Pharm* 9: 419-430.
- Dai Y, Hou LF, Chan YP, Cheng L, But PP 2004. Inhibition of immediate allergic reactions by ethanol extract from *Plumbago zeylanica* stems. *Biol Pharm Bull* 27: 429-432.
- Elizabetsky E 1986. New directions in ethnopharmacology. *J Ethnobiol Ethnomed 6*: 121-121.
- Ferre S, Guix T, Prat G, Jane F, Casas M 1990. Is experimental catalepsy properly measured? *Pharm Biochem Behav 35*: 753-757.
- Geetha, VS, Viswanathan S, Kameswaran L 1981. Comparison of total alkaloids of *Tylophora indica* and Disodium cromoglycate on mast cell stabilization. *Indian J Pharmacol* 13: 199-201.
- Gokhale AB, Saraf MN 2002. Recent developments in asthma therapy. *Indian Drugs 39*: 121-132.
- Khandelwal KR 2003. Practical Pharmacognosy Techniques and Experiments, Nirali Prakashan, Pune: 10th edition.
- Kulshrestha S, Misra SS, Sharma AL, Sharma P, Singhal D 1983. Response of the goat trachea to some autonomic drugs. *Indian J Pharmacol 15*: 2: 107-110.
- Kumar D, Bhujbal SS, Deoda RS, Mudgade SC 2010b. *In-vitro* and *In-vivo* antiasthmatic studies of *Ailanthus excelsa* Roxb. on guinea pigs. *J Sci Res 2*: 196-202.
- Kumar D, Bhujbal SS, Deoda RS, Mudgade SC 2010c. Bronchodilator activity of aqueous extract of stem bark of *Ailanthus excelsa* Roxb. *Pheog Res 2*: 102-106, doi: 10.4103/0974-8490.62955.
- Kumar D, Bhujbal SS, Patil PS, Buge PV 2010a. In-vitro and In-vivo activities of stem bark of methanolic extract of *Ailanthus excelsa* Roxb. In the management of asthma. *Int J Pharmacol* 6: 284-289, doi: 10.3923/ijp.2010.284.289.

- Lakadwala AD, Dadkar NK, Dohadwala AN 1980. Action of clonidine on mast cells of rats. *Pharm Pharmacol* 32: 790-791.
- Lavhale MS, Mishra SH 2007. Nutritional and therapeutic potential of *Ailanthus excelsa* A Review. *Pharmacognosy Reviews 1*: 105-113.
- Leeman M, Lejeune P, Melot C, Maeije R 1987. Reduction in pulmonary hypertension and in airway resistances by enoximone (MDL 17, 043) in decompensated COPD. *Chest 91*: 662-666.
- Mainardi T, Kapoor S, Bielory L 2009. Complementary and alternative medicine: Herbs, phytochemicals and vitamins and their immunologic effects. *J Allergy Clin Immunol* 123: 283-294.
- Mondal S (Parui), Mondal A, Mandal S 2007. Evaluation of electroelution and immunodiffusion as methods for purification and identification of the allergenic proteins of *Ailanthus excelsa* Roxb, pollen. *Grana* 46: 91-97.
- Nadkarni KM 2000. *Indian Materia Medica*. Popular Prakashan Private Limited Publication, Mumbai: 3<sup>rd</sup> edition, Vol 1: 56-57
- NagChaudhari AK, Lahiri SC 1974. Use of goat trachea isolated tracheal chain preparation. *Indian J Pharmacol 6*: 149-151
- Oliver N, Gerald H, Gert K 2003. Immunological and clinical changes in allergic asthmatics following treatment with Omalizumab. *Int Arch Allergy Immunol* 131: 46-52.
- Pungle P, Banavalikar M, Suthar A, Mengi S 2003. Immunomodulatory activities of boswellic acids of *Boswellic serrata* Roxb. *Indian J Exp Biol 41*: 1460-1462.
- Suresh B, Dhanasekaran S, Elango K 1995. Anti-pyretic activity of some plants in female albino rats: A preliminary report. *Ancient Sci Life 14*: 253-257.
- Taur DJ, Nirmal SA, Patil RY 2007. Effect of various extracts of *Ficus bengalensis* bark on clonidine and haloperidolinduced catalepsy in mice. *Pharmacologyonline 3*: 470-77.
- Uvnas B 1969. Mast cells and histamine release. Ind Pharm 1: 23-32a.
- Vadnere GP, Somani RS, Singhai AK 2007. Studies on antiasthmatic activity of aqueous extract of Clerodendron phlomidis. Pharmacologyonline 1: 487-494
- Wagner H, Bladt S 1996. Plant Drug Analysis; Springerverlag, Publication; Singapur; 2<sup>nd</sup> edition.

# \*Correspondence

Dinesh Kumar

Department of Pharmaceutical Sciences, University of Kashmir, 190006 J& K, India

Tel.+91 9882120308, +91 9419214116, +91 9419854348 Fax: +91 2027420261