Vol 1|Issue 1|Jan – Jun 2011 |1-4.

e-ISSN 2248 – 9142
print-ISSN 2248 – 9134

International Journal of
Current Pharmaceutical & Clinical Research

www.ijcpcr.com

# ANTI-ASTHMATIC ACTIVITY OF WHOLE PLANT OF *Azadirachta indica L.*

***K.K. Senthil Kumar**

*Department of Pharmaceutics, Cherraan's College of Pharmacy, Siruvani Main Road, Coimbatore, India.

**ABSTRACT**

Over the past decade, herbal and ayurvedic drugs have become a subject of world importance, with both medicinal and economical implications. A regular and widespread use of herbs throughout the world has increased serious concerns over their quality, safety and efficacy. Thus, a proper scientific evidence or assessment has become the criteria for acceptance of herbal health claims. *Azadirachta indica* L. (Meliaceae) widely distributed because of both religious and medicinal applications. We examined the effect of ethanol extract of roots of *Azadirachta indica* 25, 50, 100mg/kg doses orally in the isolated goat tracheal chain preparation, passive paw anaphylaxis in rat. The extract showed significant dose-dependent antiasthmatic activity in all these models.

**Keywords:** Antiasthmatic activity, *Azadirachta indica* , Meliaceae

## INTRODUCTION

*Azadirachta indica* L. (Meliaceae) is a Tree 6 to 25 m tall Cultivated and naturalized in lowland areas, Native to India and Malaysia, and now widely distributed because of both religious and medicinal applications. The Chemical *Constituents* [**1-6]** include Androstadiendione derivatives, azadirachtins and derivatives, azadirinin, azadirol, azadiradione derivatives, limbolide, limbonin, limocins, limocinone, lophenol, margocilin, margolone, margolononе, margosin, isomargosinolide, margosolone, meldenin derivatives, margosinone, melia lactone, melia polysaccharides, melicitrin, 6- methoxymellein, myricetin glycoside, naheedin, nimbadiol, nimbaflavone, nimbanal, nimbandiol, nimbidin, nimbidinin, nimbidiol, nimbidol, nimbilicin, nimbilin, nimbin derivatives, nimbinene, 6-deacetyl nimbinal, nimbinin, deacetylnimbinolide, nimbinone, nimbiol, nimbione, nimbionol, nimbisonol, nimbocidin, nimbocinol, nimbocinone, nimbolide, nimbolins, nimbonolone, nimolinin, nimolinone, nimosone, nimbin polysaccharides, several organo sulphur compounds, cholesterol, cycloartanol derivatives, cycloeucalenol, daucosterol, ergostadienol, beta sitosterol, fraxidin, 5-hydroxymethyl furfural, gedunin derivatives, hyperoside, kaempferol its glycoside, quercetin glycoside, iso rhamnetin, 3-deacetylsalannol, salanin, salannolactams, salannolide, scopoletin, tiglic acid, vepaol, 1,3-diacetyl vilasinin.The Literature survey concluded that the plant possess Insect antifeedant, insecticidal, antiarthritic, anti-inflammatory, antiulcerative, antitumour, antipyretic, antiviral, cytotoxic, nematocidal, molluscicidal, fish poison, antifertility, anti-implantation, insect repellant, larvicidal, abortifacient, antifungal, spasmolytic, wound healing acceleration, hypotensive, antihyperglycemic, analgesic, CNS depressant, antifilarial, dermatitis producing. To treat asthma, diabetes and syphilis. Antipyretic, antidysenteric, for skin diseases and as an insecticide [6-10]. Even though *Azadirachta indica* L. was reported to be useful in a many ailments, scientific evaluation of the plant was not reported for its anti asthmatic activity. Hence, in the present study, the anti asthmatic activity of extract of whole plant of *Azadirachta indica* L. was studied using different in vivo and in vitro animal models.

Corresponding Author:- **K.K.Senthil Kumar** Email:-kksenthilqa@yahoo.co.in

**e-ISSN 2248 – 9142**
**print-ISSN 2248 – 9134**

## MATERIALS AND METHODS

### Plant collection

The Plant material of whole plant of *Azadirachta indica* L. used for investigation was collected from S.V. University at Tirupathi, Chittoor (Dist.), Andhra Pradesh, India. The plant was authenticated by Dr. K. Madhava Chetty, Department of botany, S.V.University, Tirupathi.

### Preparation of extracts

The whole plant of *Azadirachta indica* L. were dried in shade, separated and made to dry powder. It was then passed through the 40 mesh sieve. The powdered material (200 g) was extracted with ethanol using Soxhlet apparatus. The extract was evaporated under reduced pressure using rotary evaporator until all the solvent has been removed to give an extract sample. Percentage yield of Ethanol extract of whole plant of *Azadirachta indica* L. (EEAI) was found to be 12.5% w/w.

### Animals

Isolated adult goat tracheal tissue, and Albino rats (Wistar Strain) of either sex weighing 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 rats were obtained from animal house of Cherraan's College of Pharmacy, Coimbatore. They were housed in polypropylene cages with standard pellet chow and water ad libitum. In all experimental sets, 5 rats were used for each treatment.

## ANTIASTHMATIC ACTIVITY

### 1) Isolated goat trachea chain preparation

Isolated adult goat tracheal tissue was obtained immediately after slaughterhouse 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 80 µg/ml EEAI 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 [12,13].

### 2) Passive paw anaphylaxis in rats

Rats (Wistar) were given (s.c.) three doses of 100 µg of egg albumin adsorbed on 12 mg of aluminum hydroxide gel prepared in o.5 ml of saline on 1st, 3rd, 5th day. On 10th day of sensitization blood was collected from the retro orbital plex and collected blood was allowed to clot and the serum was separated by centrifugation at 1500rpm. Animals were divided into five groups (n = 5). Animals belonging to group I served as control and were administered only the vehicle (10ml/kg p.o.). Animals belonging to groups II, III, IV received three doses (25, 50, 100mg/kg p.o.) respectively of EEAI. Animals of group V, as positive control group received Dexamethasone (0.27mg/kg p.o.). The animals were passively sensitized with 0.1ml 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 hr after sensitization. Animals were challenged in the left hind paw with 10µg of egg albumin in 0.1ml of saline, and the paw inflammation was measured using a Plethysmometer. 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- (Vt / Vc) × 100

Vt = Mean relative change in paw volume in test group

Vc = Mean relative change in paw volume in control group.

Prior drug treatment animals were sensitizes with serum. Next 24 hours, after drug treatment

animals again challenged for10 µg egg albumin and edema inhibition was calculated [13,14].

### Statistical Analysis

The data were expressed as mean ± standard error mean (S.E.M). The Significance of differences among the group was assessed using one way and multiple way analysis of variance (ANOVA). The test followed by Tukey-Kramer multiple comparison tests, the p values less than 0.05 were considered as significance.

## RESULTS

### 1) Isolated goat trachea chain preparation

It was observed that EEAI inhibits contraction produced by histamine in these tissue preparations. Histamine (50µg/ml) was taken in different dose level and DRC was plotted. Study revealed that *Azadirachta indica L.* extract exhibits significant ($p<0.01$) percentage decreased contraction at concentration 80 µg /ml in goat tracheal chain preparation Dose dependent response relationship was seen. (Table-1)

### 2) Passive paw anaphylaxis in rats

There was significant inhibition in rat paw edema at the dose 50mg/kg of EEAI, in all time intervals when percentage inhibition was calculated but more specific effect was seen at 3hour interval time. It was 39.07% and 57.82% for 50mg/kg and dexamethasone respectively. Paw edema volume also significantly ($p<0.01$) decreased in all time intervals at this dose only. Control group showed (0.64 ± 0.03) paw edema volume and that of for 50 mg/kg dose and dexamethasone was (0.39 ± 0.03) and (0.27 ± 0.02) at 3 hour interval. Results are comparable with that of standard dexamethasone. It was seen that further increase in dose showed decrease in activity. (Table-2)

**e-ISSN 2248 – 9142**
**print-ISSN 2248 – 9134**

**Table 1. Effect of *Azadirachta indica* extract on histamine induced contractionon isolated goat tracheal chain preparation**.

| Groups | Dose of histamine ( 50 µg/ml) | Control group % maximum contraction (Mean ± SEM) | Test group % maximum contraction (Mean ± SEM) |
|---|---|---|---|
| 1 | 0.1 | 21.46 ± 1.95 | 9.62 ± 0.93** |
| 2 | 0.2 | 25.91 ± 1.95 | 12.22 ± 1.37** |
| 3 | 0.4 | 43.33 ± 1.62 | 21.11 ± 1.24** |
| 4 | 0.8 | 55.17 ± 2.10 | 27.77 ± 1.69** |
| 5 | 1.6 | 81.46 ± 1.95 | 39.24 ± 1.09** |
| 6 | 3.2 | 94.06 ± 1.87 | 45.51 ± 1.48** |

n = 6
Values are in Mean ± SEM.
Control = D.R.C. of Histamine in absence of *Azadirachta indica* extract.
Test = D.R.C. of Histamine in presence of *Azadirachta indica* extract.(80µg/ml)
Statistical analysis done by using Students't'-test.
**p<0.01, significantly different from control.

**Table 2. Effect of *Azadirachta indica* extract on passive paw anaphylaxis in rats**

| Groups | | Paw Edema Volume (ml) Mean ± SEM | | | | |
|---|---|---|---|---|---|---|
| Sr.no. | Dose | 1/2hr | 1hr | 2hr | 3hr | 4hr |
| 1. | Control | 0.73±0.04 | 0.68±0.02 | 0.65±0.01 | 0.64±0.03 | 0.61±0.02 |
| 2. | Dexamethasone | 0.30±0.02** | 0.26±0.02** | 0.27±0.01** | 0.27±0.02** | 0.24±0.02** |
| 3. | 25 | 0.47±0.02** | 0.45±0.03** | 0.42±0.03** | 0.41±0.03** | 0.41±0.03** |
| 4. | 50 | 0.45±0.03* | 0.44±0.04** | 0.40±0.02** | 0.39±0.03** | 0.38±0.03** |
| 5. | 100 | 0.48±0.02** | 0.47±0.03** | 0.43±0.02** | 0.42±0.03** | 0.39±0.02** |

n = 5; *p<0.05, **p<0.01,compared with control group (ANOVA followed by Dunnett's test)

## DISCUSSION

Histamine contracts the tracheo-bronchial muscle of guinea pig, goat, horse, dog and man. Goat tracheal chain is easier to handle and to prepare; it is also much more sensitive than guinea pig tracheal chain. In the present study the isolated goat tracheal chain preparation; there is right side shift of Dose Response Curve (DRC) of histamine in the presence of *Azadirachta indica L.* ethanolic extract indicating antiashmatic action [15]. In passive paw edema models, extract showed the dose dependent responses. Thus *Azadirachta indica L.* can prevent the release of inflammatory mediators or inflammation in asthma.

In conclusion the present study confirmed that the ethanolic extract of *Azadirachta indica L.* exhibits significant dose dependent antiasthamatic activity in various in-vitro and in-vivo animal models and further supports the traditional claim of plant in the treatment of asthma. Further studies are infact underway to isolate and characterize the active principle responsible for the antiasthamatic activity.

## REFERENCES

1. Ponglux D, et al., Medicinal Plants, Bangkok, Thailand, 1987, 41.
2. Cambie RC. and Ash J. Fijian Medicinal Plants, CSIRO, Australia, 1994, 204-206.
3. Bokel M. et al., *Tetrahedron*, 46 (3), 1990, 775-782.
4. Govindachari et al., *J. Nat. Prod*., 55 (5), 1992, 596-601.
5. Siddiqui S. et al., *Phytochemistry*, 31 (12), 1992, 4275-4278.
6. Siddiqui S. et al., *J. Nat. Prod*., 55 (3), 1992, 303-310.
7. Balandrin MF. et al., *J. Agr. Food Chem*., 36 (5), 1988, 1048-1054.
8. Khalid SA. et al., *J. Nat. Prod*., 52 (5), 1989, 922-926.
9. Isman MB. et al., *J. Agr. Food Chem.*, 38 (6), 1990, 1406-1411.
10. Okpanyi SN. and Ezeukwu GC. *Planta Med.*, 41, 1981, 34-39.
11. Castillo JC, De-Beer EJ. The Tracheal Chain-l - A Preparation for the Study of Antispasmodics with Particular Reference to Bronchodilator Drugs. *J. Pharmacol. Exp. Ther*., 90, 1947, 104-109.

**e-ISSN 2248 – 9142**
**print-ISSN 2248 – 9134**

12. Chaudhari KN, Lahiri SC. Role of Goat Trachea for an Isolated Tracheal Chain Preparation . *Indian J. Pharmacology*, 6(3), 1974, 149-151.
13. Mitra SK. Antiasthmatic and Antianaphylactic effect of E- 721B : A Herbal Formulation . *Indian J. Pharmacol*., 31, 1999, 133-137.
14. Perdue MH, Gall DG. Intestinal Anaphylaxis in the Rat : Jejunal response to In-vitro Antigen Exposure. *AJP - Gastrointestinal and Liver Physiology*, 250 (4), 1998, 427-431.
15. Chaudhari KN, Lahiri SC. Role of Goat Trachea for an Isolated Tracheal Chain Preparation. *Indian J. Pharmacology,* 6(3): 1974, 149-151.