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## Research Article

### Antihistaminic and Antispasmodic Potential of *Pongamia pinnata*

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#### Abstract

The present study was undertaken to evaluate antihistaminic and anti-spasmodic potential of *Pongamia pinnata* (L) (PP) seeds by using isolated goat tracheal chain and chicken ileum preparation. In isolated goat tracheal chain preparation and in chicken ileum preparation, dose response curve of histamine in absence and in presence of ethanolic extract of PP was plotted. PP showed significant decrease in contraction induced by histamine (30µg/ml) in isolated goat tracheal chain and chicken ileum preparation respectively. These results suggest the antihistaminic and antispasmodic potential of ethanolic extract of *Pongamia pinnata* seeds (PP).

#### 1. INTRODUCTION

Herbal medicines are being increasingly utilized to treat a wide variety of diseases, though the knowledge about their mode of action is relatively insufficient. Interest regarding the pharmacological evaluation of various plants used in traditional system of medicine is relatively increasing<sup>1</sup>. The use of traditional medicine is expanding to newer horizons and plants still remain as the novel source of structurally important compounds that lead to the development of innovative drugs. Naturally occurring compounds from plants are still used in pharmaceutical preparations in pure or extracted forms<sup>2</sup>.

The *Pongamia pinnata* (L) Pierre belonging to family 'Papilionaceae' is popularly known as Karanj. It is medium sized glabrous tree, found in India and further distributed eastwards mainly in the South Eastern Asia and Australia. Traditionally, the *Pongamia pinnata* (L) Pierre seeds are being used to treat inflammation, cough, whooping cough and allergic disorders such as asthma, bronchitis<sup>3</sup>. The seeds of *Pongamia pinnata* (L) Pierre have been reported for antioxidant<sup>4</sup>, anti-inflammatory<sup>5</sup>, antibacterial<sup>6</sup>, antifungal<sup>7</sup>, anthelmintic activity<sup>8</sup>.

Thus taking into the consideration the traditional claims and reported pharmacological activities of *Pongamia pinnata* (L) Pierre the present study was undertaken to evaluate anti-histaminic and antispasmodic potential of ethanolic extract of *Pongamia pinnata* seeds (PP) which may further contribute in allergic disorders.

#### 2. MATERIALS AND METHODS

##### 2.1 Procurement of Plant

The seeds of *Pongamia pinnata* (L) Pierre were collected from the local area in Kolhapur, Maharashtra and were authenticated from Agharkar Research Institute, Pune, India (Authentication number: Auth 08-009).

##### 2.2 Preparation of ethanolic extract

The extractions of seeds of *Pongamia pinnata* (L) Pierre was carried by using soxhlation method. After 7-days of drying, the dried seeds were powdered by grinding and sieved with a 40# sieve. In this method 1000 gm of seed powder was extracted with 95% ethanol. It was then filtered and concentrated in vacuum under reduced pressure using a rotary evaporator and concentrated to obtain the ethanolic extract of seeds of *Pongamia pinnata* (L) Pierre (PP). The % yield obtained from leaves was 25% w/w.

##### 2.3 Procurement of Animals

Isolated adult goat tracheal and chicken tissue was from slaughter house. Trachea was collected in the ice cold oxygenated Krebs' solution and chicken tissue was collected in the ice cold oxygenated Tyrode solution.

##### 2.4 Methodology

##### 2.4.1 Histamine induced contraction of isolated goat trachea preparation<sup>9</sup>

The goat tracheal tissue was obtained immediately after slaughter of animals. Pieces of trachea were collected in freshly prepared ice-cold oxygenated Krebs' solution (Composition mM: NaCl, 115; KCl, 4.7; CaCl<sub>2</sub>, 2; NaHCO<sub>3</sub>, 25; KH<sub>2</sub>PO<sub>4</sub>, 1.2; Mg<sub>2</sub>SO<sub>4</sub>, 1.19; glucose, 11.5). Goat trachea was then cut into individual rings and tied together in series to form a chain. It was suspended in bath containing Krebs' solution and maintained at 37 ± 0.5 °C, a stream of air was bubbled through the organ tube (1 bubble/sec). One end of the tracheal muscle was attached to S-shaped aerator and the other attached to isotonic frontal writing lever to a drum. The tissue was allowed to equilibrate for 45 min under a load of 1g. A dose response curve for histamine was recorded at variant molar concentrations by maintaining 15 min time cycle. After obtaining dose response curve of histamine (30 µg/ml) on trachea, the PP (100 µg/ml) was added to reservoir and same doses of histamine were repeated. Graph of percentage of maximum contractile response on ordinate and negative log of molar concentration of histamine on abscissa was plotted to record dose response curve of histamine, in absence and in presence of PP and standard drug Chlorpheniramine maleate (1 µg/ml).

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### 2.4.2 Histamine induced contraction of chicken ileum preparation<sup>10</sup>

Chicken ileum was suspended in bath containing Tyrode solution (Composition mM: NaCl, 136.7; KCl 2.68; CaCl<sub>2</sub>, 1.8; NaHCO<sub>3</sub>, 11.90; NaH<sub>2</sub>PO<sub>4</sub>, 0.42; MgCl<sub>2</sub>, 1.05; glucose, 5.55) maintained at 37±0.5°C. A stream of air was bubbled through the organ tube (1 bubble/sec). One end of the ileum was attached to S-shaped aerator and the other attached to isotonic frontal writing lever to a drum. The tissue was allowed to equilibrate for 45 min under a load of 500 mg. A dose response curve for histamine was recorded at variant molar concentrations by maintaining 15 min time cycle. After obtaining dose response curve of histamine (30 µg/ml) on ileum, the PP (100 µg/ml) was added to reservoir and same doses of histamine were repeated. Graph of percentage of maximum contractile response on ordinate and negative log of molar concentration of histamine on abscissa was plotted to record dose response curve of histamine, in absence and in presence of PP and standard drug Chlorphenaramine maleate (1 µg/ml).

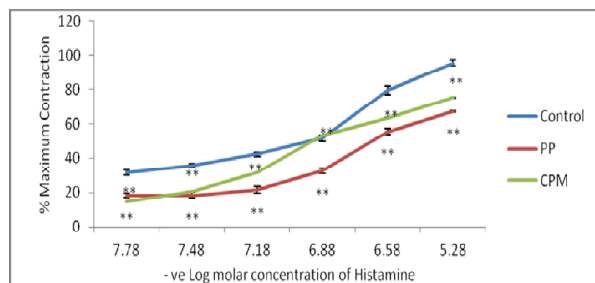
### 2.5 Statistical Analysis

The results have been indicated in terms of mean ± SEM, (n=5). Difference between the groups was statistically determined by one way ANOVA with Dunnett's test. The level of significance was set at \*\*p<0.01.

## 3. RESULTS

### 3.1 Histamine induced contraction of isolated goat trachea preparation

Histamine produced dose dependant contraction in goat tracheal chain preparation at the concentration 30 µg/ml. This was significantly inhibited by modified PSS into which the preparation was incubated with ethanolic extract of seeds of PP at 100 µg/ml as well as with Chlorphenaramine maleate (1 µg/ml). This indicates that there was competitive antagonism between histamine and PP for H<sub>1</sub> receptors present on the smooth muscle. (Figure 1)

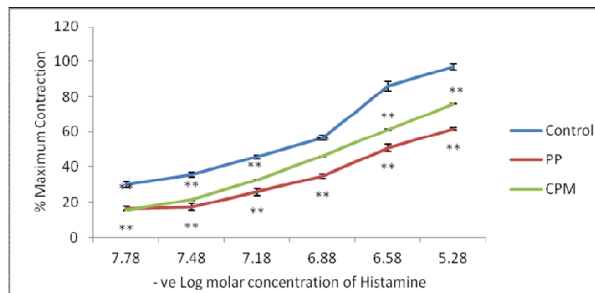


**Figure 1:** Effect of ethanolic extract of *Pongamia pinnata* (L) Pierre on histamine induced contraction of smooth muscle using isolated goat tracheal chain preparation.

Control = D.R.C. of histamine (30 µg/ml) in absence of test drug  
 PP = D.R.C. of histamine (30 µg/ml) in presence ethanolic extract of PP (100µg/ml).  
 CPM = D.R.C. of histamine (30 µg/ml) in presence Chlorphenaramine maleate (1 µg/ml).

### 3.2 Histamine induced contraction of chicken ileum preparation

Histamine produced dose dependant contraction in chicken ileum preparation at the concentration 30 µg/ml. This was significantly inhibited by modified PSS into which the preparation was incubated with ethanolic extract of seeds of PP at 100 µg/ml as well with Chlorphenaramine maleate (1 µg/ml). This indicates that there was competitive antagonism between Histamine and PP. (Figure 2)



**Figure 2:** Effect of ethanolic extract of *Pongamia pinnata* (L) Pierre on histamine induced contraction of chicken ileum preparation.

Control = D.R.C. of histamine (30 µg/ml) in absence of test drug  
 PP = D.R.C. of histamine (30 µg/ml) in presence ethanolic extract of PP (100µg/ml).  
 CPM = D.R.C. of histamine (30 µg/ml) in presence Chlorphenaramine maleate (1 µg/ml).

## 4. DISCUSSION

For screening the activity of a drug on respiratory smooth muscles, both goat tracheal chain and strip preparations can be used<sup>9</sup>. Goat tracheal chain is easier to acquire, handle and easier to dissect and has the same reactions to spasmogenic and spasmolytic drugs<sup>11</sup>. Different agonists like ACh, Histamine, 5- Hydroxytryptamine and Bradykinin show contractile response in isolated goat trachea. H<sub>1</sub> receptor stimulation causes contraction of bronchial smooth muscle<sup>12</sup>. It is reported that activation of α-adrenergic and H<sub>1</sub>-histaminergic receptors causes activation of VIP (Vasoactive Intestinal Polypeptide) in cerebral cortex, which is responsible for release of histamine from sensory neurons<sup>13</sup>. This leads to activation of IP<sub>3</sub> and DAG pathway. This increased IP<sub>3</sub> is responsible for releasing the microsomal calcium, leads to phosphorylation of actin-myosin fibers of goat trachea causing the contraction. Thus, the contraction of tracheal or bronchial smooth muscle in vitro has often been utilized for the study of contractile / dilator responses of agonists as well as antagonist<sup>14</sup>.

In the present study, histamine showed maximum contraction while PP significantly inhibited the histamine induced contraction of isolated goat tracheal chain preparation. The parallel shift of graph towards right side in histamine concentration-response curves in the presence of increasing concentrations of PP indicating that there was competitive antagonism between histamine and PP for H<sub>1</sub> receptors present on the smooth muscle. This effect may be due to its antihistaminic or antispasmodic activity.

Histamine elicited contractions in ileum are believed to be mediated through H<sub>1</sub> receptors present on ileum<sup>15&16</sup>. It was found that PP produced dose dependent inhibition of ileum contractions induced by histamine. The parallel rightward shift in agonist concentration-response curves of histamine in the presence of increasing concentrations of PP was indicating ant-histaminic or antispasmodic activity. The inhibition may be due to the antagonism of histaminergic receptors or nonspecific spasmolytic action.

## 5. CONCLUSION

It can be concluded from present study that the ethanolic extract of *Pongamia pinnata* (L) Pierre seeds possesses significant antihistaminic and anti-spasmodic activity that may be attributed due to H<sub>1</sub>-antagonistic. This may further contribute in the management of allergic disorders like asthma.

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