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PRECLINICAL EVALUATION AND ANTIASTHMATIC ACTIVITY OF *CASSIA TORA* LINN. LEAVES

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ABSTRACT
The present study was designed to investigate the efficacy and safety of *Cassia tora* Linn leaves. The anti-asthmatic activity of the extract by using different concentration of aqueous extract as 500, 1000 and1500 µg/ml was evaluated in isolated goat trachea chain, preparations by using standard drugs histamine.
In the present study, histamine produced dose dependent contraction of goat tracheal chain preparation was studied. As number of papers with antiasthmatic activity are reported with use of guinea pig ileum, tracheal chain preparation but yet antiasthmatic activity was not carried out using tracheal chain hence present study was designed using tracheal chain preparation. The actual dose required to produce bronchodilation was not known so comparison was made by testing various concentration of drug extract of *Cassia tora* L. The modified physiological salt solution containing hydroalcoholic extract of *Cassia tora* (1.5 mg/ml) significantly inhibited the contractile effect of histamine thus produces significant bronchodilation.
It is concluded that 1500 µg/ml of *Cassia tora* showed potent antiasthmatic activity due to bronchodilator activity.
Key words: Antiasthmatic, *Cassia Tora* Linn, Histamine, Goat tracheal chain.

INTRODUCTION

Bronchial asthma is a chronic inflammatory disorder of the airways characterized by paroxysmal dyspnoea and wheeze due to increased resistance to the flow of air through the narrowed bronchi. Narrowing of the bronchi is due to spasm of the bronchial smooth muscles, infiltration of inflammatory cells basophil and macrophage), edema of bronchial mucosa, blocking of bronchial mucosa and blocking of bronchial lumen by mucus. It can lead to COPD[1]. Among several respiratory diseases affecting man, bronchial asthma is the most common disabling syndrome.

Despite the wide range of drugs are available in market, the relief offered by them is mainly symptomatic and short lived. Moreover, these drugs produce side effects. Therefore, there is a dire need to identify effective and safe remedies to treat bronchial asthma. Ayurveda, an ancient system of Indian medicine, has recommended a number of drugs from indigenous plant sources for the treatment of bronchial asthma and allergic disorders[2]. *Cassia tora* Linn. A foetid, annual herb up to 1-2m in height leaves per pinnate; leaflets 3 pairs family Caesalpiniaceae. It has been used in the Ayurvedic system for the treatment of asthma, also used as an antifungal, anthelmintic[3], hypolipidemic, hepatoprotective[4], antibacterial activity[5], purgative, jaundice, stomachic, tonic, eye diseases, liver complaints and earache[6].

MATERIAL AND METHOD

Tissue preparation

Goat trachea was obtained from the slaughter house and kept in Krebs's solution.

Collection of plant and Authentication

Fresh leaves of *Cassia tora* Linn were collected from Hadapsar region of Pune district, (Maharashtra) India, authenticated from botanical survey of India, Pune. (Voucher specimen no. – AST-1)

Preparation of Extract

The fresh leaves of *Cassia tora* Linn were collected, and then dried leaves were obtained by sun drying. The dried leaves (80gm) were taken in 250 ml of water and kept for maceration for about 7 days and filtered and concentrated on hot plate. The different preservative like toluene, chloroform was added in it to prevent microbial growth.

METHODS

Isolated Goat Tracheal Chain Preparation

Goat trachea was brought from slaughter house was cut into individual ring and tied together in series to form chain. It was suspended in bath containing Krebs's solution maintained at 37 ± 1^0 C stream of CO^2 in O_2 was bubbled through the organ tube. One end was tied to aerator tube and other attached to isotonic frontal lever to Kymograph paper on Sherrington rotating drum. Tissue was allowed to equilibrate for 45 min under to load of 400mg. A dose response curve for histamine was taken in variant molar concentration. After obtaining a dose curve of histamine on goat trachea aqueous solution of extract (n=4) was added to reservoir and same dose of histamine were repeated[7].

Statistical Analysis

The results of various studies were expressed as mean ± SEM and analyzed statistically using one way ANOVA followed Dunnett's test to find out the level of significance. Data were considered statistically significant at minimum level of $p < 0.01$.

RESULT AND DISCUSSION

Physicochemical and Phytochemical Screening

In the present study *Cassia tora* Linn was used to study anti-asthmatic activity. Preliminary qualitative phytochemical screening of aqueous extract of leaves of *Cassia tora* Linn showed the presence of lignans, flavonoids glycosides, sterols, sugars, amino acids and

triterpenoids. The physical constant / physicochemical parameters were also studied.

Table 1: Results of Physico-Chemical Parameters

Sr.no	Parameters	Inference
1	loss on drying	13% w/w
2	Total ash value	16%
3	Acid insoluble ash value	3%
4	Sulphated ash value	15%
5	Water soluble ash value	6%

Histamine is autacoid is one of the major inflammatory mediators in the immediate phase of asthma, causing airway hyper responsiveness and bronchial airway inflammation. Besides the triple response caused by it, histamine has spasmogenic response on intestinal smooth muscle by acting on H1-histamine receptor that causes the contraction of intestinal smooth muscle. Guinea Pig is highly sensitive to histamine due to presence of histaminergic receptors in ileum and tracheal smooth muscle[8].

Histamine is synthesized, store and released by mast cells in the airway wall. In blood, histamine is stored in basophils, The non-mast cell histamine is stored in histaminocytes in the stomach and in histaminergic neurons in the brain apart from this number of mediators releases on antigen antibody reaction like Kinins and others[9]. Although, airway mast cells are likely to be the major cellular source of histamine in asthma there is increasing evidence that basophiles may be recruited to asthmatic airways and may release histamine in response to cytokine histamine-releasing factors hence, Histamine has multiple effects on airway function that are mediated by specific surface receptors on target cells.

H1 receptors mediate most of the effects of histamine that are relevant to asthma[10]. The published paper suggested that H1 receptors have been demonstrated in animal and human lung , guinea pig are responsible to produce bronchial, smooth muscle contraction. Smooth muscle contraction, thus in the present study we use isolated goat treachial chain preparation. The similar response exhibited by the extract in case of goat tracheal chain preparation, which support the above statement that extract of *Cassia tora* Linn leaves was acting on H1 receptor as antagonists.

In the present study actual dose required to produce bronchodilation was not known so comparison was made by testing various concentration of drug extract of *Cassia tora* Linn The modified physiological salt solution containing hydro alcoholic extract of *Cassia tora* Linn (1.5 mg/ml) significantly inhibited the contractile effect of histamine thus produces significant bronchodilation. (table 2-4) .

Table 2: Effect of Aqueous Extract of Leaves of *Cassia tora* L. on Histamine Induced Contraction of Isolated Goat Tracheal Chain Preparation

Sr. no	Dose (ml)	Concentration (µg/ml)		Log dose		% Response		Height (cm)	
		H	**D**	**H**	**D**	**H**	**D**	**H**	**D**
1	0.1	5	50	0.6989	1.6989	53.84	30.76	0.7	0.4
2	0.2	10	100	1	1	61.53	46.15	0.8	0.6
3	0.4	20	200	1.3010	1.3010	92.30	30.76	1.2	0.4
4	0.8	40	400	1.6020	1.6020	92.30	-	1.2	-
5	1.6	80	800	1.9030	1.9030	92.30	-	1.2	-

Histamine-50µg/ml, Drug-500 µg/ml
H-Histamine, D-Drug extract of *Cassia tora* Linn

Table 3: Effect of Aqueous Extract of Leaves of *Cassia tora* L. on Histamine Induced Contraction of Isolated Goat Tracheal Chain Preparation

SN	Dose(ml)	Concentration (µg/ml)		Log dose		% response		Height (cm)	
		H	**D**	**H**	**D**	**H**	**D**	**H**	**D**
1	0.1	5	100	0.6989	2	33.33	66.66	0.2	0.2
2	0.2	10	200	1	2.3010	50	66.66	0.3	0.2
3	0.4	20	400	1.3010	2.6020	83.33	-	0.5	-
4	0.8	40	800	1.6020	2.9030	83.33	-	0.5	-
5	1.6	80	1600	1.9030	3.2041	100	-	0.6	-

Histamine-50µg/ml, Drug-1mg/ml
H-Histamine, D-Drug extract of *Cassia tora* Linn

Table 4: Effect of Aqueous Extract of Leaves of *Cassia tora* L. on Histamine Induced Contraction of Isolated Goat Tracheal Chain Preparation

SN	Dose(ml)	Concentration (µg/ml)		Log dose		% response		Height (cm)	
		H	**D**	**H**	**D**	**H**	**D**	**H**	**D**
1	0.1	5	150	0.6989	2.1760	57.14	30	0.4	0.3
2	0.2	10	300	1	2.4771	71.42	20	0.5	0.2
3	0.4	20	600	1.3010	2.7781	85.71	-	0.6	-
4	0.8	40	1200	1.6020	3.0791	85.71	-	0.6	-
5	1.6	80	2400	1.9030	3.3802	100	-	0.7	-

Histamine-50µg/ml, Drug-1.5mg/ml
H-Histamine, D-Drug extract of *Cassia tora* Linn

CONCLUSION

Asthma is a chronic inflammatory disease of the air-ways with a wide range of presentations from intermittent to mid symptoms with chronicity. To control such disorders the methanolic extract of *Cassia tora* Linn leaves exhibited antagonistic activity on histaminergic receptors in the present study.

As the dose of *Cassia tora* Linn was not mentioned for *in vitro* study, so we have carried out *in vitro* study of antiasthmatic activity of *Cassia tora* L. by using different concentration of aqueous extract as 500 , 1000 and 1500 µg/ml and it is concluded that 1500 µg/ml showed potent bronchodilator activity.

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