

SCREENING OF LEAF EXTRACT OF ARGEMONE MEXICANA FOR IT'S ANTI-ASTHMATIC ACTIVITY IN ISOLATED GOAT TRACHEAL CHAIN PREPARATION

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ABSTRACT

The main objective of the work was to evaluate the use of *Argemone mexicana* (AM) leaves for safety and efficacy in the treatment of asthma. The anti-asthmatic activity of the extract by using different concentration of methanolic extract at 500,1000 and 1500 µg/ml was evaluated in isolated goat tracheal chain preparations by using standard drug histamine. In the present study, histamine produced dose dependent contraction of goat tracheal chain preparation, 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 *Argemone mexicana*. The modified physiological salt solution containing methanolic extract of *Argemone mexicana* (1.5 mg/ml) significantly inhibited the contractile effect of histamine thus produces significant bronchodilation. It is concluded that 1500 µg/ml of *Argemone mexicana* showed potent antiasthmatic activity due to bronchodilator activity.

KEYWORDS: Antiasthmatic, *Argemone Mexicana*, Histamine, Goat Tracheal Chain.

INTRODUCTION

Asthma is a chronic inflammatory disease of the air-ways with a wide range of presentations from intermittent to mid symptoms with chronicity. Despite the fact that advancement in the treatment of asthma is on the increase, prevalence and mortality of asthma affect approximately 10% of children and 5% of adults, worldwide.^[1] Asthma is characterized by recurrent episodes of wheezing, breathlessness, chest tightness and cough, reversible airway obstruction and bronchial hyperresponsiveness to a variety of specific and nonspecific stimuli, including: allergen, histamines, chemical irritants, cold air and exercise.^[2] Asthma is a global problem, many synthetic drugs are used to treat acute symptoms of asthma, but they are not completely safe for long term use. Hence search has been started once again to look back to traditional medicine which can be used to treat asthma. Ayurveda and other Indian literature mention the use of plants treatment for various human ailments.^[3] *Argemone mexicana* L., known as Ghamoya (class: Magnoliopsida Dicotyledons; subclass: Magnoliidae; order: Papaverales; family: Papaveraceae; Figure no: 1) is an exotic weed indigenous in South America but has widespread distribution in many tropical and sub-tropical countries including West Africa.^[4] This plant is common everywhere by roadsides and fields in India as well.^[5] The plant has shown Antibacterial activity^[6], Anti-inflammatory activity^[7], Wound healing activity^[8], antidiabetic activity^[9] but its antiasthmatic activity is not yet validated scientifically as on date. Hence in the current dissertation the antiasthmatic activity of methanolic extract of leaves of *Argemone mexicana* in isolated goat tracheal chain preparation was performed.

MATERIALS AND METHODS

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

Plant Material: Leaves of *Argemone mexicana* were collected in the Month of August from the agricultural fields of Kopergaon, Maharashtra, India. The collected plant material was shade dried to retain its vital phytoconstituents and then subjected to size reduction for further extraction process.



Figure no: 1.

Preparation of Methanolic extract

The powder of *Argemone mexicana* leaves was charged in to the thimble of a Soxhlet apparatus and extracted using methanol. Appearance of colourless solvent in the siphon tube was the indication of exhaustive extraction and based on that the further extraction was terminated. The extract was then transferred into the previously weighed empty beaker and evaporated to a thick paste on the water bath, maintained at 50° C to methanol extract. The extract was finally air dried thoroughly to remove all traces of the solvent and the percentage yield was calculated. The perfectly dried extract was then stored in an air tight container in a refrigerator below 10°C.

Storage of Extract: Methanolic extract of *Argemone mexicana* was stored in tightly closed glass bottles in refrigerator at 2-8°C.

Preparation of extract Solutions: Test solutions (T.S) of Methanolic extract of AM were prepared in distilled water in order to make concentration 100 mg/ml.

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 $37 \pm 1^{\circ} \text{C}$ 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 \pm 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 $p < 0.01$.

RESULTS AND DISCUSSION

Table 1: Effect of Methanolic Extract of Leaves of *Argemone mexicana* on Histamine Induced Contraction of Isolated Goat Tracheal Chain Preparation.

Sr. No.	Dose (ml)	Concentration ($\mu\text{g/ml}$)		Log dose		%Response		Height (cm)	
		H	D	H	D	H	D	H	D
1	0.1	5	50	0.6989	1.6989	72.72	30	0.8	0.3
2	0.2	10	100	1	1	81.81	50	0.9	0.5
3	0.4	20	200	1.3010	1.3010	90.90	40	1.0	0.4
4	0.8	40	400	1.6020	1.6020	90.90		1.0	-
5	1.6	80	800	1.9030	1.9030	100		1.1	-

Histamine-50 $\mu\text{g/ml}$, Drug-500 $\mu\text{g/ml}$

H-Histamine, D- Drug extract of *Argemone mexicana*

Table. 2: Effect of Methanolic Extract of Leaves of *Argemone mexicana* on Histamine Induced Contraction of Isolated Goat Tracheal Chain Preparation.

Sr. No.	Dose (ml)	Concentration ($\mu\text{g/ml}$)		Log dose		% Response		Height (cm)	
		H	D	H	D	H	D	H	D
1	0.1	5	100	0.6989	2	28.57	50	0.2	0.2
2	0.2	10	200	1	2.3010	57.14	25	0.4	0.1
3	0.4	20	400	1.3010	2.6020	71.42	-	0.5	-
4	0.8	40	800	1.6020	2.9030	85.71	-	0.6	-
5	1.6	80	1600	1.9030	3.2041	100	-	0.7	-

Histamine-50 $\mu\text{g/ml}$, Drug-1mg/ml

H-Histamine, D-Drug extract of *Argemone mexicana*

Table. 3: Effect of Methanolic Extract of Leaves of *Argemone mexicana* on Histamine Induced Contraction of Isolated Goat Tracheal Chain Preparation.

Sr. No.	Dose (ml)	Concentration ($\mu\text{g/ml}$)		Log dose		%Response		Height (cm)	
		H	D	H	D	H	D	H	D
1	0.1	5	150	0.6989	2.1760	62.5	50	0.5	0.3
2	0.2	10	300	1	2.4771	75	16.66	0.6	0.1
3	0.4	20	600	1.3010	2.7781	87.5	-	0.7	-
4	0.8	40	1200	1.6020	3.0791	87.5	-	0.7	-
5	1.6	80	2400	1.9030	3.3802	100	-	0.8	-

Histamine-50 $\mu\text{g/ml}$, Drug-1.5 mg/ml

H-Histamine, D- Drug extract of *Argemone Mexicana*.

DISCUSSION

Pharmacotherapy using plant-derived substances can be currently regarded as a very promising future alternative to current synthetic drug therapy. The advanced techniques and technologies available today enable to investigate chemically well-defined bioactive plant components as sources of novel drugs.^[10] Histamine is an autacoids, 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.^[11]

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.^[12] 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.^[13] 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 have used isolated goat tracheal chain preparation. The similar response exhibited by the extract in case of goat tracheal chain preparation, which support the above statement that methanolic extract of *Argemone mexicana* leaves was acting on H1 receptor as antagonists.

CONCLUSION

Asthma is a disease of the lung's airways. It represents wide range of symptoms from intermittent mild to severe chronicity. In the present study anti-asthmatic activity of the extract by using different concentraton of methanolic extract at 500,1000, and 1500 µg/ml was evaluated in isolated goat tracheal chain preparations by using standard drug histamine. Histamine produced dose dependent contraction of goat tracheal chain preparation, The

modified physiological salt solution containing methanolic extract of *Argemone mexicana* (1.5 mg/ml) significantly inhibited the contractile effect of histamine thus produces significant bronchodilation. It is concluded that 1500 µg/ml of *Argemone mexicana* showed potent antiasthmatic activity due to bronchodilator activity.

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