

Research Article

Membrane Stabilizing Activity of *Xanthium strumarium* Leaves Extracts

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ABSTRACT

Xanthium strumarium L. is commonly known as burweed plant commonly found as a weed, is widely distributed in North America, Brazil, China, Malaysia and hotter parts of India. The present study deals with the evaluation of membrane stabilizing activity of *Xanthium strumarium* Linn extracts. Among ancient civilizations, India has been known to be rich repository of medicinal plants. The traditional medicine involves the use of different plant extracts or the bioactive constituents. This type of study provides the health application at affordable cost. The study such as ethno medicine keenly represents one of the best avenues in searching new economic plants for medicine. The extracts were prepared with different selected solvents for the plant to adjudge the major active principles in the solvent that have value in rational drug design. The qualitative analysis of phytochemicals of each extracts was analysed. The results of this study showed that methanol was the richest extract for phytoconstituents and it showed maximum membrane stabilizing activity as compare to other tested extracts. These findings suggested that methanol extract of leaf of *Xanthium strumarium* L. has potent membrane stabilizing ability which could be the reason for its use in inflammation related diseases.

Keywords: *Xanthium strumarium*, anti-inflammatory, erythrocyte membrane stabilization, Aspirin, hypotonic solution.

INTRODUCTION

In the last decades, plant have been used for treatment of disease and maintaining health of human being. Medicinal plants are richest source of natural phytoconstituents. There is huge demand of phytoconstituents for medicinal purposes has increases in all over the world. The plant phytoconstituents are the best source of variety of medicinal agents¹. All over the world, medicinal plants are used as a folklore traditional medicine for treatment of diseases². Inflammatory diseases are increasing all over the world. These diseases involves complex array of enzyme activation, mediator release, fluid extravasations, cell migration, tissue breakdown and repair which are aimed at host defence and usually activated in disease condition. currently there is huge demand in the searching of medicinal plants with anti-inflammatory activity which may lead to the discovery of new therapeutic agent that is not only used to suppress the inflammation but also used in diverse disease conditions where the inflammation response in amplifying the disease process³. *Xanthium strumarium* L. belongs to family asteraceae and also known as cocklebur or burweed. *Xanthium strumarium* L. commonly found

throughout tropical part of India⁴. *X. strumarium* grows in waste places, roadsides and also river bank in tropical parts. The herb is annual and upto 1 m in height. The whole plant is used as medicine. There are many more medicinal properties of the plant like cooling, laxative anthelmintic antipyretic fattening, improves appetite, complexation and memory which is reported in ayurveda. The plant is also used for the treatment of leucoderma, biliousness, epilepsy. The leaves and roots are used for the treatment of anodyne, antirheumatic, antisiphilitic, appetiser, diaphoretic, diuretic, emollient, laxative and sedative activities. The fruits has anodyne, antibacterial, antifungal, antimalarial, antirheumatic, antispasmodic, antitussive, cytotoxic, hypoglycaemic and stomachic properties⁵. The plant has been reported several medicinal properties like anti-inflammatory⁶, antinociceptive⁶, antimitotic⁷, diuretic⁸, repellent and insecticidal⁹, antitumour¹⁰, antimicrobial¹¹, antifungal¹², antitrypanosomal¹³, antioxidant¹⁴, anticancer¹⁵, hypoglycaemic¹⁶, antitussive¹⁷, antiplasmodial¹⁸ activities. In present study the selection of plant for evaluation was based on its traditional uses and evaluated for the its membrane stabilizing activity.

Table 1. Effect of different extracts of *Xanthium strumarium* on stability of erythrocyte membrane:

S. No.	Concentration (µg/ml)	% Inhibition of hemolysis of extracts				Standard drug	
		Hexane	Chloroform	Acetone	Methanol	Aspirin	Concentration (µg/ml)
1	Control	-	-	-	-	-	-
2	1000	03.42±0.88	07.94±1.6	16.14±1.3	39.68±1.2	50.77±1.13	100
3	1500	10.08±1.1	21.18±1.8	29.14±0.7	53.19±0.99	55.01±1.4	150
4	2000	15.90±0.7	30.24±1.67	45.07±1.5	57.12±0.97	59.10±1.4	200

MATERIALS AND METHODS

Collection & Identification of leaves of *Xanthium strumarium*

Leaves of *Xanthium strumarium* were collected from locality of Dehradun (India). Plant material was authenticated by S. K. Srivastava (Scientist D/HOD), in Botanical Survey of India, Northern regional centre, Dehradun (BSI). Authenticated specimen no is – 114541.

Extraction of leaves of *Xanthium strumarium* in different solvents

The collected plant material was washed with water to removed other undesirable material and dried under shade. The air-dried leaves (125gm) of *Xanthium strumarium* were crushed. The crushed leaves extracted with different solvents of increasing polarity viz. Hexane, Chloroform, Acetone, Methanol by hot percolation method using Soxhlet Apparatus. The extract was evaporated till dryness to obtain residue. These extracts were concentrated under reduced pressure¹⁹.

Phytochemical Analysis

All the extracts were analysed for the presence of phytoconstituents. The test for carbohydrates, alkaloids, steroids, terpenoids, phenolic compounds, saponins, protein and amino acids was done for each extract¹⁹.

Invitro Membrane Stabilizing activity of extract

Invitro Membrane Stabilizing activity was done by hypotonic induced haemolysis²⁰⁻²³.

Effect on haemolysis

Erythrocyte suspension

Blood was collected from goat under ether anesthesia. To prevent clotting in blood, add NIH-ACD (National Institute of Health-Acid Citrate Dextrose) solution. After that blood was washed three times with 0.9% saline. The volume of saline was measured and reconstituted as a 40% (v/v) suspension with isotonic buffer solution (pH 7.4). Which contained in 100 ml of distilled water: NaH₂PO₄.2H₂O, 0.26 gm, NaH₂PO₄.2H₂O, 1.15 gm; NaCl, 9 gm (10 Mm sodium phosphate buffer (pH 7.4).

Hypotonic solution-induced haemolysis

Stock erythrocyte suspension (30 µl) was mixed with 5 ml of the hypotonic solution containing the *Xanthium strumarium* leaf extract at concentration of 1000, 1500, 2000 µg/ml, while the control sample was mixed with drug free solution. The mixtures were incubated for 10 minute at room temperature, and centrifuged at 3000 rpm for 10 minutes. All the experiments were performed in triplicates and the absorbance (O.D.) of the supernatant was measured at 560 nm. Acetyl Salicylic Acid (Aspirin) was used as a reference standard of concentration 100, 150 and 200 µg/ml for comparison.

Calculation

The percentage inhibition or acceleration of hemolysis in test (b) and (c) was calculated according to the equation:

$$\% \text{ Inhibition of hemolysis} = 100 \times (OD_1 - OD_2 / OD_1)$$

Where, OD₁= Optical density of hypotonic saline solution + blood (control) and OD₂= Optical density of test sample in hypotonic saline solution + blood

RESULTS AND DISCUSSION

The percentage yield of leaves extracts in different solvents system are Hexane (7.23 %), chloroform (20.67%), Acetone (1.241%), Methanol (6.80%). The extract of leaves of *Xanthium strumarium* undergoes various qualitative chemical tests. we found out that methanol extract was the highly active extract for phytoconstituents. It contains some tested phytoconstituents viz. Alkaloids, carbohydrates, and Phenolic compounds. Proteins and amino acid, triterpenoids of sterols, fats and fixed oil and saponins was absent in methanol. Acetone extract showed the presence of carbohydrates and Phenolic compounds only. Hexane and chloroform extracts both did not show any phytoconstituents.

Membrane Stabilizing activity

The Membrane Stabilizing activity of the different sample extracts was compared with activity of standard drug Aspirin at 560 nm. From the comparison with standard drug it was observed that the concentration of 2000µg/ml of methanol extract showed maximum activity (57.10%). While other extract such as acetone showed 45.07% activity, chloroform showed 30.24% activity and hexane showed 15.90% activity (Table 1). Membrane stabilizing activity is related to anti-inflammatory activities preliminary²¹. Inflammation is due to release of lysosomal constituents causes cell death also. Rupturing of lysosomal membrane releases lysosomal constituents and the stabilization of lysosomal membrane inhibits the release of lysosomal constituents³. Erythrocyte membrane is resemblance to lysosomal membrane, and by stabilize erythrocyte membrane by plant extract may also stabilize lysosomal membrane². Hypotonic solution induced erythrocyte membrane lysis can be taken for the invitro determination of anti-inflammatory activity of drugs or plant extracts by stabilize the erythrocyte membrane²¹.

CONCLUSION

From the above studies, it could be concluded that, the methanol extract of plant showed the increase in the protection of the erythrocyte membrane against hypotonic hemolysis which could be reason for its use in

inflammatory diseases. Further study is needed for the study mechanism of action and isolation of active principle from active extract.

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