

Original Research Article

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Anti-Inflammatory Activity on *Hibiscus sabdariffa* Seeds

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ABSTRACT

Keywords

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Anti inflammatory activities of methanolic extract obtained from the dried extract of *Hibiscus sabdariffa* were investigated in an attempt to evaluate its medicinal potentials. The methanolic extract of the seeds of *Hibiscus sabdariffa* at the doses of 200 and 400 mg/kg b.w and indomethacin at dose 10 mg/kg produced dose dependent significant reduction in carrageenan-induced rat maximal paw oedema by the results suggested that the seeds *Hibiscus sabdariffa* possessing anti-inflammatory activity, but to a lower extent.

Introduction

The word inflammation comes from the latin word 'inflammare', means to burn. Inflammation (Serhan *et al.*, 2005) is a response of the tissue to an injection, irritation or foreign substances. It is a part of host's defense or inflammation, a defensive reaction to injury with classical signs of warmth, reddening, pain, swelling and loss of function, is of acute or chronic type. The characteristics (Seeley *et al.*, 2000) of inflammation are numerous: reddening (visible), swelling (Anonymous, 1975) (oedema), soreness (pain) and the corresponding histological changes. Inflammation serves to destroy, dilute or wall off the injurious agents and the tissue cells that may have been destroyed. In turn, the inflammatory response sets into motion a

complex series of events, which heal and reconstitute the damaged tissue. Repair being doing the active phase of inflammation, but reaches completion usually after repairing the damaged tissue cells had neutralized the injurious response, there. Both inflammation and repair generally serve as useful purposes. Inflammatory reaction underlines the genesis of crippling rheumatoid arthritis, life threatening sensitivity reaction and some forms of glomerular diseases. Inflammation is normally beneficial, being part of a complex protective homeostatic mechanism. The complexity of inflammatory process and the diversities of the drugs that have been found effective in modifying the process have resulted in the development of

numerous methods for detecting anti-inflammatory substances. In the present investigation, the author has tried to test the anti-inflammatory activity of the methanolic extract of seeds of *Hibiscus sabdariffa*. The method that followed was carrageenan induced rat paw oedema model. The plant *Hibiscus sabdariffa* (Malvaceae), claimed to be used traditionally in the treatment of various ailments including rheumatoid arthritis, anti-depressant, parkinson's disease, infertility, stomachic and emollient.

However, Literature survey indicated hepatoprotective activity, anti-inflammatory and anti-ulcer activity of leaves and anti-fertility activity on male rats (Robert, 1965' Nasrin *et al.*, 2005; Winder *et al.*, 1958; Meier *et al.*, 1958; Robert *et al.*, 1957; Jain *et al.*, 1991; Chopra *et al.*, 1958; Winter *et al.*, 1962; D'Arcy *et al.*, 1960). But no published reports on the anti-inflammatory activities on seeds of the plant. In the view of this the author aimed to study the anti-inflammatory activity of the methanolic extracts of the seeds of *Hibiscus sabdariffa*

Extraction process

The collected seeds were dried under shade

and powdered. The powdered materials were reflux extracted.

Reflux extraction

The dried powdered materials of seeds of the plant were extracted successively three times with methanol. The extract thus obtained were concentrated and dried completely, weighed and stored in a dessicator

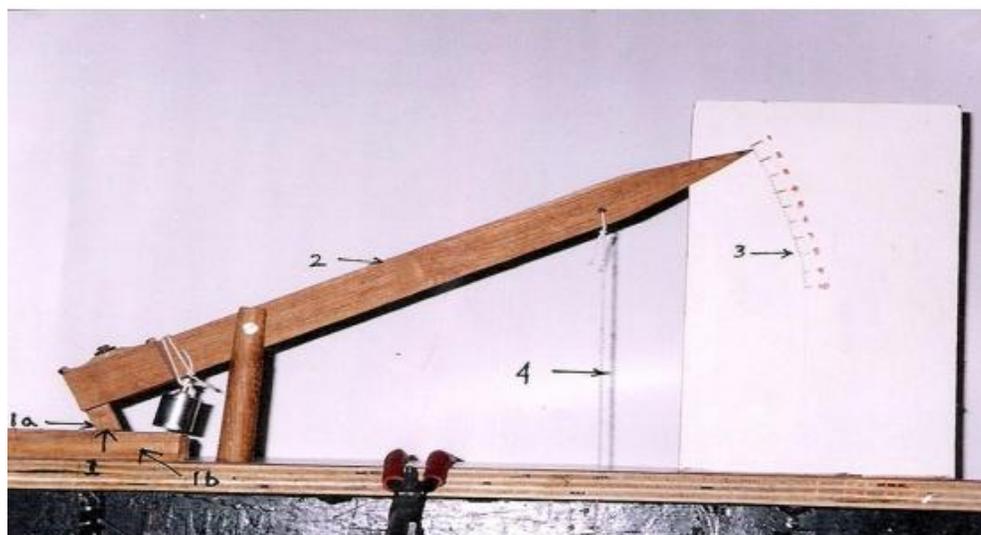
Materials for anti inflammatory activity

Animals	: Albino wistar rats of either sex (175-225g)
Carrageenan	: 1% suspension in saline.
Test drugs	: Methanolic extracts of seeds of <i>Hibiscus sabdariffa</i> (At the doses of 200, 400mg/kg b.w)
Standard drug	: Indomethacin (10mg/kg b.w)
Drug vehicle	: sodium CMC in water.

Apparatus available for measurement of oedema (Paw thickness/ volume)

1. Zeitlin's constant loaded lever
2. Plethymograph (13)

Zeitlin's apparatus was used to measure the paw thickness.



Zeitlin's constant loaded liver (paw thickness measuring devices)

1. Place, where the paws use to be kept to measure the thickness.
2. Constant load lever.
3. Graduated scale numbered between 1-10 and divided by 0.5cms
4. Thread to pull down the lever with right leg in order to facilitate to keep the paw in between pointer 1a and basement 1b.

Preparation of carrageenan suspension

Suspension of carrageenan sodium salt 1% was prepared by sprinkling 100mg of carrageenan powder in 10 ml of saline (0.9% sodium chloride) and set aside to soak for 1 hour and then the suspension was mixed thoroughly using magnetic stirrer.

Preparation of sodium CMC suspension

Stock suspension of sodium CMC was prepared by triturating the powder sodium CMC (1 g) finely in 2.5ml of water containing tween 20. A 1:10 dilution of this stock solution made in distilled water was used for suspending the test and standard drugs.

Experimental procedure

Inflammation was induced in the right hind paw of each rat by sub plantar injection of 1% carrageenan suspension (0.1 ml). The left hind paw of the rat was injected 0.1 ml of saline.

Table.1

Plant material	Solvent used	No. of cycles	Weight of the extract
Dried seeds 1 kg	Methanol	3	45.2gr

Table.2

Treatment	% inhibition of maximal paw oedema During 6 th hour
Group A Drug vehicle	0.0±0.99
Group B Standard (indomethacin)	64.00±4.96
Group C Dose 200 mg/kg	9.09±1.52
Group D Dose 400 mg/kg	4.54±1.32

Fig.1 Effect of crude extract of *Hibiscus sabdariffa* 200, 400 mg/kg along with indomethacin (10 mg/kg) on the total paw oedema in Carrageenan induced rats

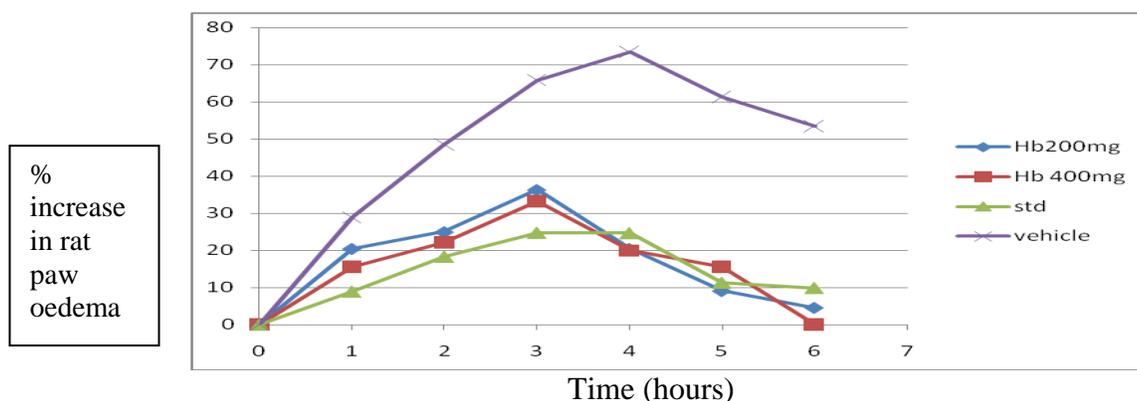
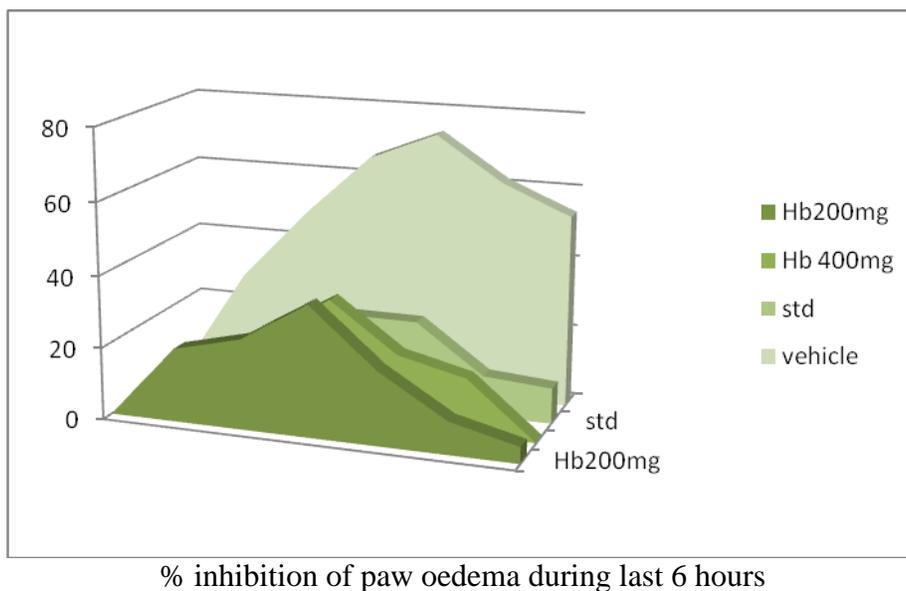


Fig.2 Percentage inhibition of carrageenan induced paw oedema in rats by prophylactic treatment with the methanolic extract of *Hibiscus sabdariffa*



Animals were divided into different groups (each contains 4 rats) as follows:

Group A received drug vehicle 1% sodium CMC. Group B received standard drug Indomethacin at the dose 10mg/kg. Group C and D received methanolic extract of *Hibiscus sabdariffa* at the doses 200 and 400 mg/kg body weight, respectively. Two hours after administration of doses, each rat was injected with saline subcutaneously into sub-plantar tissue of the left hind paw. The paw thickness of each rat was measured using Zeitlin's apparatus before carrageenan injection and every hour up to 6hrs after carrageenan injection. The percentage inhibition of paw oedema was calculated by using the following formula.

$$\% \text{ Increase in paw thickness} = \frac{Y_t - Y_0}{Y_0} \times 100$$

Y_t = Paw thickness at time (1, 2, 3, 4, 5 and 6th) after injection.

Y_0 = Paw thickness at 0 hour (before injection).

Results and Discussion

Sub-plantar injection of 1% carrageenan (0.1ml) produced marked, sustained and time related increase and decrease in the rat hind paw oedema of the control group. Paw swelling and or oedema was reached peak level at 4 hour after the injection of carrageenan and gradually decreased in the following hours.

The methanolic extract of seeds of *Hibiscus sabdariffa* at the doses of 200 and 400 mg/kg b.w and indomethacin at dose 10 mg/kg produced dose dependent significant reduction in carrageenan-induced rat maximal paw oedema by the results suggested that the seeds *Hibiscus sabdariffa* possessing anti-inflammatory activity, but to a lower extent. The preliminary phytochemical examination suggested that the seeds having sterols, saponins and flavonoids. It is therefore assuming that since the plant possessing sterols. The anti-inflammatory activity of the plant maybe due to the presence of the above said

category of compounds. It is therefore worth study further to isolate the pure molecules responsible for anti-inflammatory activity.

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