ANTI-INFLAMMATORY ACTIVITY OF FRUITS OF *ABELMOSCHUS ESCULANTUS* LINN.

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Summary

Anti-inflammatory activity of the methanolic and aqueous extracts of the fruits of *Abelmoschus esculentus* Linn. were studied in wistar rats using the carrageenan induced left hind paw edema. The methanolic and aqueous extracts at a dose of 250 mg/kg body wt shows moderate to significant anti-inflammatory activity. The methanolic and aqueous extracts of *A. esculentus* reduced the edema induced by carrageenan by 48.29 % and 68.85 % respectively on oral administration of 250 mg/kg body wt, as compared to the untreated control group. Diclofenac sodium at 100 mg/kg body wt inhibited the edema volume by 75.08 %. The results indicated that the aqueous extract shows more significant anti-inflammatory activity then methanolic extracts when compared with the standard and untreated control.

Key words: Anti-inflammatory, *Abelmoschus esculentus*, Carragennan.

Introduction

Abelmoschus esculentus L. (or Hibiscus esculentus or Okra) – Malvaceae is used for a long time as an edible vegetable in many countries, and commonly eaten in India and Vietnam because of its nourishing components. Traditionally, it is believed that the plant is useful in the treatment of inflammatory disorders, constipation and retention of urine. On the other hand, a number of previous studies have reported that *Abelmoschus sp.* possessed hypoglycemic effect[1]. However, there is a little study regarding its anti-inflammatory effect.

Prolonged uses of both steroidal and non-steroidal anti-inflammatory drugs are well known to be associated with peptic ulcer formation[2]. Hence, search for new anti-inflammatory agents that retain therapeutic efficacy and yet are devoid of these adverse effects is justified. There is much hope of finding active anti-rheumatic compounds from indigenous plants as these are still used in therapeutics. Herbal drugs are being proved as effective as synthetic drugs with lesser side effects.

The enzyme, phospholipase A2, is known to be responsible for the formation of mediators of inflammation such as prostaglandins and leukotrienes which by attracting polymorphonuclear leucocytes to the site of inflammation would lead to tissue damage probably by the release of free radicals. Phospholipase A2 converts phospholipids in the cell membrane into arachidonic acid, which is highly reactive and is rapidly metabolized by cycloxygenase (prostaglandin synthase) to prostaglandins, which are major components that induce pain and inflammation[3,4]. Arachidonic acid is also converted to leukotrienes via lipoxygenase enzyme.

The aim of this present study is to investigate and evaluate the anti-inflammatory effect of *Abelmoschus esculentus* L. extracts on caragennan induced inflammation in rats and provide scientific evidence for development of *Abelmoschus esculentus* L. as a potential natural oral anti-inflammatory agent or functional food.

Materials and Methods

Plant Material:

The fruits of *Abelmoschus esculentus* were collected from the local areas of Bardoli, identified and authenticated by Prof. B.R. Patel, Dept. of Botany, PG Science College, Bardoli. A specimen voucher of the same is deposited in the college museum. The fruits were shade dried at room temperature for 10 days and coarsely powdered with the help of a hand-grinding mill and the powder was passed through sieve No. 60.

Preparation of the Extract

The powder of fruits of *Abelmoschus esculentus* was extracted by continuous hot extraction process using soxhlet apparatus with methanol and water [5].After extraction, the extracts were concentrated and the extractive values were calculated with reference to the air-dried drug. The dried extracts were subjected to various chemical tests to detect the presence of different phytoconstituents.

Animals:

Wistar rats of either sex and of approximately the same age, weighing about 150-200 g were used for the study. They were housed in polypropylene cages and fed with standard diet and water *ad libitum*. The animals were exposed to alternate cycle of 12 h of darkness and light each. Before each test, the animals were fasted for at least 12 h. The experimental protocols were subjected to the scrutinization of the Institutional Animal Ethics Committee and were cleared by the same.

Acute Toxicity Studies:

The animals were divided into control and test groups containing six animals each. The control group received the vehicle (5% acacia) while the test groups received graded doses of extracts orally (p.o.) and were observed for mortality till 48 h and the LD50 was calculated.[6].

Carrageenan Induced Rat Paw Edema:

Anti-inflammatory activity was assessed by the method described by Winter *et al*[7]. The rats were divided into four groups of six animals each. First group (control) received 5 ml/kg body wt of normal saline; second group (standard) received 100 mg/kg body wt (i.p) diclofenac sodium, third group received methanolic extract (250 mg/kg body wt, p.o.) and fourth group received aqueous extract (250 mg/kg body wt, p.o.) of *A. esculentus*, respectively. After 1 h, the rats were challenged with subcutaneous injection of 0.1 ml of 1 % w/v solution of carrageenan (Sigma chemical co, St. Louis MO, USA) into the plantar side of the left hind paw. The paw was marked with ink at the level of lateral malleolus and immersed in solution up to the mark. The plethysmograph apparatus used for the measurement of rat paw volume was og UGO Basil company. The paw volume was measured immediately after injection (0 h) and then every hour till 4 h after injection of carrageenan to each group. The difference between the initial and subsequent reading gave the actual edema volume. Percent inhibition of inflammation was calculated using the formula,

$$\% \text{ inhibition} = 100\ (1-Vt/Vc)$$

Where 'Vc' represents edema volume in control and 'Vt' edema volume in group treated with test extracts.

Statistical Analysis:

All values were expressed as mean. The data were statistically analyzed using one way ANOVA followed by Students t test and differences below $p < 0.05$ are considered as significant.

Results

The average percentage yield of the methanol and aqueous extracts of *A. esculentus* was found to be 0.36 % w/w and 1.04 % w/w respectively. Preliminary phytochemical screening of the leaves of *A. esculentus* revealed the presence of alkaloids, flavanoids, saponins and tannins. The LD50 was found to be 2500 mg/kg for methanolic and aqueous extract of *A. esculentus*. So the 1/10 of LD50 dose was considered as an effective dose.

The effect of methanolic and aqueous extract of *A. esculentus* on carrageenan induced edema in rats is shown in Table 1 and Figure 1. The results obtained indicate that the methanolic extract had significant anti-inflammatory activity in rats, while aqueous

extract had more significant anti-inflammatory activity. The methanolic and aqueous extracts of *A. esculentus* reduced the edema induced by carrageenan by 48.29 % and 68.85 % respectively on oral administration of 250 mg/kg body wt, as compared to the untreated control group. Diclofenac sodium at 100 mg/kg body wt inhibited the edema volume by 75.08 %.

Table 1: Effect of various extracts of *A. esculantus* on carragennan induced rat paw edema.

Groups	**0 hr**	**1 hr**	**2 hr**	**3 hr**	**4 hr**	**Difference**	**% inhibition in paw edema at 4th hour**
Control	0.97	1.08	1.28	1.10	1.13	0.16	--
Standard	1.09	1.24	1.22	1.15	1.13	0.04	75.08
Methanol extract of *A. esculantus*	1.09	1.21	1.07	1.02	1.17	0.08	48.29
Aqueous extract of *A. esculantus*	1.07	1.42	1.15	1.14	1.12	0.05	68.85

Conclusions

Due to the increasing frequency of intake of NSAID's and their reported common side effects, there is a need to focus on the scientific exploration of herbal drugs having fewer side effects. So, there is a continuous search for indigenous drugs, which can provide relief to inflammation. To give a scientific validation to this plant, an attempt was made to study the anti-inflammatory activity.

Carrageenan induced inflammation is a biphasic phenomenon[8]. The first phase of edema is attributed to release of histamine and 5-hydroxy-tryptamine. Plateau phase is maintained by kinin like substances and second accelerating phase of swelling is attributed to prostaglandin like substances. The knowledge of these mediators involved in different phases is important for interpreting mode of drug action.

Thus it can be concluded that the fruits of the plant *A. esculentus* possess significant anti-inflammatory activity in rats. Further studies involving the purification of the chemical constituents of the plant and the investigations in the biochemical pathways may result in the development of a potent anti-inflammatory agent with low toxicity and better therapeutic index.

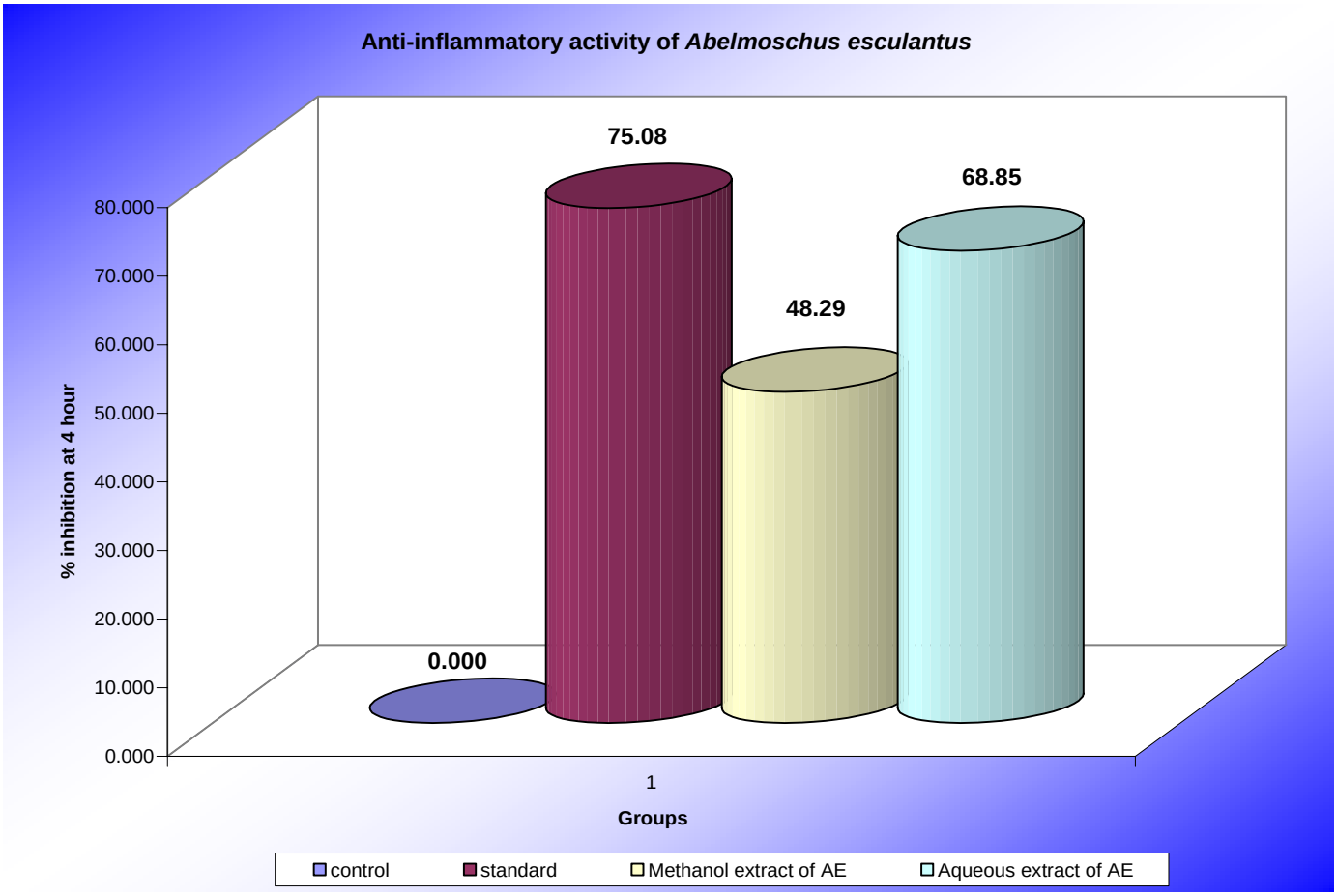

Figure 1: % Inhibition of rat paw edema at 4th hour by various extracts of *A. esculantus*.

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