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Study of the Anti-Inflammatory Activity of *Cassia Fistula* Linn.

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Abstract

Inflammation involving the innate and adaptive immune systems is a normal response to infection. However, when allowed to continue unchecked, inflammation may result in autoimmune or autoin-flammatory disorders, neurodegenerative disease, or cancer. A variety of safe and effective antiinflammatory agents are available, including aspirin and other nonsteroidal anti-inflammatories with many more drugs under development. Other anti-inflammatories currently in use or under development include statins, histone deacetylase inhibitors, PPAR agonists, and small RNAs. Anti-inflammatory and Antioxidant activities of the aqueous (CFA) were assayed in wistar albino rats. The extracts were found to posses significant anti-inflammatory effect in both acute and chronic models. Cassia fistula Linn has many therapeutic uses. Therefore, we aimed to study its anti- inflammatory activity. The aqueous extract of dried fruits of Cassia fistula Linn was prepared. The anti-inflammatory activity of these extracts was investigated using the CFA-induced paw edema model in rats. It was observed that extracts of dried fruits Cassia fistula Linn showed anti-inflammatory activity. This aqueous extract showed maximum anti-inflammatory activity at 400 mg/kg dose. It showed maximum percentage inhibition of 41.15%, which was comparable with the positive control, diclofenac sodium, which showed 47% inhibition. Further, the acute toxicity study with the extracts showed no sign of toxicity up to a dose level of 2000 mg/po. Thus it could be concluded that cassia fistula bark extracts (CFA & CFM) possess significant anti-inflammatory and anti oxidant properties. The present study reveals that the aqueous extracts of fruit of Cassia fistula Linn can be used as anti-arthritic drug.

Key Words

Anti-inflammatory activity, *Cassia fistula* Linn.

Introduction

Cassia fistula Linn (family-caesalpinaceae) commonly has known as the Golden shower Indian Laburnum. It is an Indian medicinal plant. It is 8-15m to 24m in height, with greenish grey smooth bark when young & rough, dark brown when mature. Leaflets are 8.12 pair, flowers yellow, long drooping racemes. Pod cylindrical & pulpy. Seeds light brown, hard & shiny.

Medicinally it has been various pharmacological activities antipyretic, analgesic, larvicidal, anti-

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inflammatory, antioxidant, anti-tumor, hepatoprotective, hypoglycemic.

Taxonomical Classification
Kingdom: Plantae
Division: Magnoliophyta
Class: Magnoliopsida
Subclass: Rosidae
Order: Fabales
Family: Fabaceae
Subfamily: Caesalpinaceae
Genus: Cassia
Species: Cassia Fistula

Besides the above mentioned effects, the plant *Cassia fistula Linn (family-caesalpinaceae)* is not scientifically explored for its anti-inflammatory activity. Hence an effort has been made here to screen the plant for its anti- inflammatory activity.

Materials and Methods

Plant material

The plant materials of *Cassia fistula* were collected from Regional Medical Research Centre, Belgaum, Karnataka. The plant was authenticated by Dr. Harsha Hegde, Scientist B, Regional Medical Research Centre, Belgaum, Karnataka.

Preparation of aqueous extracts

The freshly collected leaves and fruits of this plant was chopped, shad dried and coarsely powdered. The powder was defatted with petroleum ether (60-80 ^{o}c) and soaked in water. The aqueous extracts were dried under reduced pressure using a rotary vacuum evaporator. The percentage yield was 7% w/w for aqueous extract (CFA). The extracts were kept in refrigerator for further use.

Animals

Male Albino rats (Wistar strain) weighing 150-200gm, procured from Sri Venkateshwara Enterprises, Bangalore, were used for the study.

Housing of the Animals

Animals were kept for one week to acclimatize to laboratory conditions before starting the experiment. They were given free access to water and standard rat feed. 12 hrs prior to an experiment, the rats were deprived of food but not water.

Chemicals and drugs

1) Carrageenan (Sigma, St. Louis, USA).
2) Diclofenac sodium (Himedia, Mumbai)
3) Tween 80 (Himedia, Mumbai)
4) Sterile water for injection (Core Health Care Ltd., Mumbai)

Acute oral toxicity

The acute oral toxicity was carried out as per the guidelines set by Organization for Economic Co-operation and Development (OECD), revised draft guidelines 423, received from Committee for the Purpose of Control and Supervision of Experiments on Animals, Ministry of Social Justice and Empowerment, Government of India.

Determination of safety of the extract

Liver function tests (GPT, GOT) and kidney function tests (serum urea and creatinine concentrations) were determined for the treated and untreated control groups (except

groups 5 and 6) 30 days after Carageenan induced oedema administration and before sacrifice of the rats to evaluate subacute toxicity of the extract. In addition, acute toxicity was determined by subjecting 7 groups of mice (7 mice each) to incremental doses of the extract and determining 24- hour lethality.

Dose selection

The doses of 200 mg/kg and 400 mg/kg b.w of aqueous extracts of fruits of *Cassia fistula,* was chosen for Carrageenan induced in rats. Diclofenac sodium 100mg/Kg body weight was used as standard drug.

Preparation of dose of selected extracts

Dose of extracts of *Cassia fistula,* was prepared as a suspension by triturating extract with water and 1 % Tween 80.

Preparation of Diclofenac sodium dose

Diclofenac sodium dosage was prepared 30 mins before administration as a suspension by triturating with water and 1 % Tween 80.

Procedure

Male Wistar albino rats weighing between 150 and 200 g were randomly selected. Animals were divided into seven groups containing six in each group. Inflammation was induced in all animals by injecting 0.1 ml of Carrageenan in sub-plantar region of left hind paw. Control group was treated with vehicle and the standard group was treated with Diclofenac Sodium (13.5mg/kg BW). The other groups were treated with aqueous extracts of the selected drugs at the dose of 200 mg/kg BW and 400 mg/kg BW. Paw oedema was induced by injecting 0.1ml of 1% Carrageenan in physiological saline into the subplantar tissues of the left hind paw of each rat. The extracts CFA (200 & 400 mg/kg) was administered orally 30 min prior to Carrageenan administration. The paw volume was measured at 60, 120, 180, 240 minutes by the mercury displacement method using a plethysmograph. The percentage inhibition of paw volume in drug treated group was compared with the control group. Diclofenac sodium (100 mg/kg) was used as reference standard. The assessment of inflammation activity was done by measuring the mean changes in paw edema on 60, 120, 180, 240 minutes .Secondary lesion (non-injected paw volume) was also measured on 60, 120, 180, 240 minutes. The changes in paw volume were recorded and % inhibition of paw edema was calculated.

Observations and Calculations

The paw edema (injected and non-injected paw) was measured on 60, 120, 180, 240 minutes after induction of Carageenan induced oedema using plethysmometer. The mean changes in injected paw edema, with respect to initial paw volume, were calculated on respective days and percentage inhibition of paw edema with respect to untreated group was calculated using following formula.

$$i = [1-(?V_{Treated} / ?V_{Control})] * 100$$

i = % inhibition of paw edema

$\Delta V_{Treated}$ =Mean change in paw volume of treated rat.

$\Delta V_{Control}$ = Mean change in paw volume of Control rat

Statistical analysis

The mean paw volume was expressed in terms of mean ± SEM and evaluated for statistical significance by ANOVA followed by dunnet t test, $p<0.05$ was considered as statistically significant.

Results and Discussion

In the present study, fruits extract of Cassia fistula, was studied for anti-inflammatory activity by Carageenan induced oedema. The extracts was found to possess significant activity at both selected doses i.e. 200 and 400 mg/kg b.w., Cassia fistula showed comparable results with the standard drug. Aqueous extracts showed significant anti- inflammatory activity. This aqueous extract showed maximum anti- inflammatory activity at 400 mg/kg dose. It showed maximum percentage inhibition of 44.15%, which was comparable with the positive control, diclofenac sodium, which showed 47.02% inhibition.

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Table 1: Percentage Inhibition of CFA Induced Paw Edema.

Group	**% Inhibition of Paw Edema**							
	60 min		**120 min**		**180 min**		**240 min**	
	Dose (mg/kg body weight)							
	200	**400**	**200**	**400**	**200**	**400**	**200**	**400**
Cassia fistula	14.13	28.56	30.26	31.26	33.99	38.99	42.61	44.15
Diclofenac Sodium 100mg/kg b.w	29.56	2956	42.90	42.90	43.99	45.99	46.08	47.02

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