Assessment of Anti-Inflammatory Activity of *Taxus Baccata Linn*. Bark Extract

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ABSTRACT

Taxus baccata (L) known as Sthauneyaka in Sanskrit[1] has wide range of biological activities including analgesic, anti-malarial, anti-rheumatic, sedative, anti-spasmodic, aphrodisiac and anti-asthmatic. In the present study, the dried and powdered bark of Taxus baccata (L) was extracted with 95% ethanol and ether at room temperature and screened for their anti-inflammatory activity by Carrageenan-induced paw edema method in rat. 95% ethanol extract exhibits potent anti-inflammatory activity at 200mg/kg four hours after administration in comparison with ether extract, as well reference standard, Aspirin. The observed pharmacological activities provide a scientific basis for the folklore use of the plant in treating acute inflammation.

Keywords: *Taxus baccata, Anti-inflammatory, Anti-rheumatic, Carrageenan*

INTRODUCTION

Nature has been a source of medicinal agents for thousands of years and since the beginning of man. In India, almost all plants are medicinal and the application of medicinal plants especially in traditional medicine is currently well acknowledged and established as a viable profession[2]. Extraction of bioactive compounds from medicinal plants permits the demonstration of their physiological activity. It also facilitates pharmacology studies leading to synthesis of a more potent drug with reduced toxicity[3-7]. The inflammatory reaction is a response of the organism against an injury and it involves the action of complex events and mediators through the blood vessels[8]. It is a body defense reaction in order to eliminate or limit the spread of injurious agents[9].

In historical documents from the Roman period, *Taxus baccata (*L) was used as an analgesic, anti-inflammatory, anti-malarial, anti-rheumatic, emmenagoque, sedative, antispasmodic, aphrodisiac and anti-asthmatic[10]. The genus *Taxus (*L) has interested many researchers since the discovery of the anti-cancer agent paclitaxel (Taxol), a diterpenoid alkaloid originally isolated from the bark of the Pacific yew, *Taxus brevifolia*[11] (Nuttall, 1849). It was also listed in Avicenna's cardiac drugs, namely *Zarnab*. So far, the isolation of a large number of taxoids as well as lignans, flavonoids, steroids and sugar derivatives has been reported from different parts of various *Taxus* species[12]. In this paper, we would like to describe the extraction and evaluation of anti-inflammatory activity of bark extract of *Taxus baccata (*L).

MATERIALS AND METHODS

Plant Material

The bark of *Taxus baccata (*L) was collected from Tawang forest Divisions of Arunachal Pradesh in the month of September. The plant was identified and authenticated by the Botanist of Shibpur Botanical Garden, Kolkata, West Bengal (voucher specimen no. SBG/08-005).

Preparation of extracts

The bark was cleaned, dried under shade at temperature 40±2C and powdered by a mechanical grinder. Then the dried and powdered bark of *Taxus baccata (*L) was extracted with 95% ethanol *and ether* at room temperature. Then the extract was concentrated and the concentrate thus obtained was suspended in H_2O and extracted with $CHCl_3$ which left residue. The $CHCl_3$ extract was purified by preparative TLC[13].

Preliminary Phytochemical Screening:

All the extracts were subjected to preliminary phytochemical qualitative screening for the presence or absence of various primary or secondary metabolites following standard procedures[14,15].

Experimental Animals

Adult Wistar albino rats of either sex weighing 150 to 200 g maintained in our college animal house were used for the study. The selected animals were maintained by giving pelleted diet, water *adlibitum* and kept in 12 hrs/12 hrs light/dark cycle. The animals were divided into six groups each containing six rats. All experimental procedures were carried out in strict accordance with the guidelines prescribed by the committee for the Purpose of Control and Supervisions on Experimentation on Animals (CPCSEA) and were approved by the Institutional Animal Ethics Committee (HPI/08/60/IAEC, 0026).

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Acute Toxicity Study

The study was carried out according to OECD (Organization of Economic Co-operation and Development) guidelines 423 (Acute Toxic class method) [16]. Nine female Wistar albino rats weighing 150-200 g were taken and extracts were administered orally to animals at a dose of 2000 mg/kg in 0.3% w/v Carboxy Methyl Cellulose Sodium. Then the animals were observed for mortality and morbidity at 0, 0.5, 1, 2, 4, 6, 8, 12 and 24 hr. Food was given to the animals after 4 hr of dosing and the body weight was checked at 6 hr after dosing. Morbidity like convulsions, tremors, grip strength, lethargy, ptosis and pupil dilation were observed. The animals were observed twice daily for 14 days and body weight was noted.

Anti-inflammatory Activity

Anti-inflammatory activity was assessed by Carrageenan-induced paw edema method in rat[17]. Wistar albino rats of either sex were weighed and divided into groups (6 rats in each group). In control group simple distilled water, in standard group Aspirin (100 mg/kg) and in test groups 95% ethanol and ether extract of *Taxus baccata (*L) (100 mg/kg and 200 mg/kg) were suspended in 0.5% w/v CMC and administered orally. After 30 minutes Carrageenan solution (1% w/v) was injected in the planter region of the left paw of control as well as standard and test groups. After the administration of Carrageenan solution the paw volume of control, standard and test groups were noted at 1hr, 2hr, 3hr, and 4hr time interval. The percentage of inhibition was calculated by using the formula[18]:

% inhibition = 100 × (1-Vt/Vc)

Where, Vc = Edema volume of Control, and Vt = Edema volume of Test

Statistical Analysis

The results are expressed as mean ± SEM. The statistical analysis was performed by analysis of variance (ANOVA) followed by Dunnet's test. $P<0.05$ was considered as significant.

RESULT

The results of the anti-inflammatory activity of 95% ethanol and ether extract of *Taxus baccata (*L) against Carrageenan-induced paw edema in Wistar albino rats are expressed in **Table 1**. A significant increase in hind paw edema was observed after 3 hrs. The percentage inhibition of edema was 44.44% at a dose of 200 mg/kg of 95% ethanol extract which is higher than the reference standard, Aspirin. The 100 mg/kg of 95% ethanol extract showed a percentage inhibition of 37.04%. The statistical analysis showed a significant value ($P< 0.05$) for the test drugs and reference standard when compared with control.

Table 1: Effect of *Taxus baccata* (L) bark extracts on Carrageenan-induced paw edema method in rats

Group	Dose mg/kg, p.o.	Paw volume in ml, Mean±SEM (% inhibition of paw edema)			
		1.0h	2.0h	3.0h	4.0h
Control	20 ml	0.72±0.034	0.97±0.041	1.02±0.037	1.08±0.039
95% Ethanol extract	100	0.61±0.013* (15.28)	0.56±0.017* (42.27)	0.60±0.007* (41.18)	0.68±0.01* (37.04)
95% Ethanol extract	200	0.63±0.016* (12.50)	0.55±0.018* (43.29)	0.57±0.012* (44.11)	0.60±0.024* (44.44)
Ether extract	100	0.66±0.012* (8.33)	0.56±0.023* (42.27)	0.59±0.017* (42.16)	0.69±0.009* (36.11)
Ether extract	200	0.55±0.051* (23.61)	0.57±0.026* (41.24)	0.60±0.027* (41.17)	0.65±0.017* (39.81)
Standard Aspirin	100	0.63±0.02* (12.50)	0.54±0.019* (44.34)	0.54±0.018* (47.06)	0.64±0.014* (40.74)

n=6 in each group. *$P<0.05$ compared to control.

DISCUSSION

The Carrageenan-induced paw edema was taken as a prototype of exudative phase of acute inflammation. Inflammatory stimuli microbes, chemicals and necrosed cells activate the different mediator systems through a common trigger mechanism. The development of Carrageenan-induced paw edema is believed to be biphasic. The early phase is attributed to the release of histamine, and serotonin and the delayed phase is sustained by the leucotrienes and prostaglandin[19].

The present study focused on the bark of Taxus baccata (L) in rat paw edema. Carrageenan- induced paw edema is the standard experimental model for acute inflammation. Carrageenan is the phlogistic agent of choice for testing anti-inflammatory drugs as it is known to be non-antigenic and is devoid of apparent systemic effects. Moreover, this model shows high degree of reproducibility.

CONCLUSION

For our study, when we compare the anti-inflammatory efficiency, among the two extracts, 95% ethanol extract of Taxus baccata (L) at 200 mg/kg is having potent anti-inflammatory effect may be due to the presence of taxol, a nitrogenous diterpenoid compared with the standard, Aspirin. This data suggest that, the drug may have the antagonist or blocking effects on mediators like histamine, serotonin and kinins. It needs further evaluation to trace the bio-molecular mechanism of *Taxus baccata (*L) bark extract.

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