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SHORT COMMUNICATION

Anti-inflammatory Action of *Ougeinia oojeinensis* (Roxb.) Hochr. Bark by HRBC Membrane Stabilization

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ABSTRACT

The ethanolic and aqueous extracts of the bark of *Ougeinia oojeinensis* (Roxb.) Hochr (Fabaceae) known in Hindi as Tinsa, were screened for anti-inflammatory activity. The dried powdered drug was successively extracted with several solvents and out of these the ethanolic and aqueous extract was selected for present study due to presence of several photochemical in them. The prevention of hypotonicity induced HRBC membrane lysis was taken as a measure of the anti-inflammatory activity. Both the fraction showed a biphasic effect on the membrane stabilization. Their activities are comparable to that of the standard drug diclofenac sodium. However their activities decreased with time.

KEY WORDS *Ougeinia oojeinensis* (Roxb.) Hochr, anti-inflammatory, HRBC

INTRODUCTION:

Since the time in immemorial, our traditional system of medicine and folklore claming that medicinal plants as a whole or their parts are being used in all types of skin diseases successfully including antibacterial and antifungal. As we know very well, nowadays the medicinal preparation available in the market from which most of them either not effective up to the mark or has to develop resistance resulting in reoccurrence again. Plant derived drug serve as a prototype to develop more effective and less toxic medicines[1].

Ougeinia oojeinensis (Roxb.) Hochr (Fabaceae) known in Hindi as Tinsa and in Sanskrit as Ratha is a deciduous trees , found in the outer Himalayas and sub-Himalayan tracts from Jammu to Bhutan up to an altitude of 1500m and extending through the whole of northern and central India into the greater part of Deccan peninsula[2,3]. The extract of the whole plant *Ougeinia oojeinensis* were scientifically evaluated for analgesic activity[4]. The 50% of ethanolic extract of stem bark showed antispasmodic action. Phytochemical investigated of *O. oojeinensis* showed the presence of lupeol, hydroxlupeol, betulin and isoflavanones such as dalbergioidin, homoferreirin and ougenin[5-7]. A survey of literature showed that no systematic approach has been made to study anti-inflammatory activity in this plant by in-vitro method. The present study is an attempt to study the anti-inflammatory activity of *O. oojeinensis* using ethanolic and aqueous extracts.

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Plant material

The bark of *O. oojeinensis* were collected from the Lamber forest Raipur, Chhattisgarh in the month of May. The collected material was authenticated by Dr. P. Jayaraman, Botanist, Plant Anatomy Research Centre (PARC), Chennai. The plant was also compared with herbarium specimen maintained at Minor Forest Produce (Trading and Development) Co-Op. Fed. Ltd., Shankar Nagar, Raipur, Chhattisgarh, by Expert Medicinal Plant, Mr. S. N. Khotele.

Preparation of extract

Dried powder of bark was exhaustively extracted successively in soxhlet apparatus, using petroleum ether, choloroform, ethylacetate, ethanol and distilled water repectively. The extracts were then made to powder by using rotary evaporator under reduced pressure. Bark of *O. oojeinensis* yielded 0.62%, 0.45%, 0.57%, 4.50% and 3.7% w/w powdered extract with petroleum ether, choloroform, ethylacetate, ethanol and distilled water repectively.

Anti-inlammatory activity

The HRBC membrane stabilization has been used as method to study the anti-inflammatory activity. Blood was collected from healthy volunteers. The collected blood was mixed with equal volume of sterilized Alsever solution (2% dextrose, 0.8% sodium citrate, 0.5% citric acid and 0.42% sodium chloride in water).

The blood was centrifuged at 3000 rpm and packed cell were washed with isosaline (0.85%, pH 7.2) and a 10 %(v/v) suspension was made with isosaline.

The assay mixture contained the drug (concentration as mentioned in Table 1), 1 ml of phosphate buffer (0.15M, pH 7.4), 2 ml of hyposaline (0.36%) and 0.5 ml of HRBC suspension. Diclofenac was used as the reference drug.

Instead of hyposaline 2 ml of distilled water was used in the control. All the assay mixture were incubated at 37°C for 30 min and centrifuged. The hemoglobin content in the supernatant solution was estimated using spectrophotometer at 560 nm. The percentage hemolysis was calculated by assuming the hemolysis produced in the presence of distilled water as 100%. The percentage of HRBC membrane stabilization or protection was calculated using the formula [8-11].

$$\% \text{ Protection} = 100 - \frac{\text{O. D. of drug treated sample}}{\text{O. D. of control}} * 100$$

RESULT AND DISCUSSION:

The lysosomal enzymes released during inflammation produced a variety of disorders. The extracellular activity of these enzymes is said to be related to acute or chronic inflammation. The diclofenac drugs act either by inhibiting these lysosomal enzymes or by stabilizing the lysosomal membrane.

Since HRBC membrane is similar to lysosomal membrane components, the prevention of hypotonicity-induced HRBC membrane lysis is taken as a measure of anti-inflammatory activity of drugs. The results are reported in Table 1. Both the extracts of the bark of *O. oojeinensis* showed biphasic effects on HRBC membrane stabilization. They increasing activity at low concentration levels but decreasing activity with high concentrations. They have a critical concentration (50 µg/ml) at which their activities are maximum. The activities of both extracts are comparable to that of Diclofenac at concentration of 50 µg/ml. The variation of activity with time was studied at 10 µg/ml concentration, the activities in general decreased with time (Table 2).

Further work is in progress to isolate and identity the compounds responsible for activity.

Table. 1
Effect of concentration on the activity of both extracts of O. *oojeinensis*

Concentration (µg/ml)	Activity (prevention of lysis %)		
	Ethanolic extract	Aqueous extract	Diclofenac
10	42.12±0.35	38.01±0.51	------
50	51.23±1.01	48.36±0.41	52.22±0.05
100	35.13±0.03	34.65±0.21	------
200	30.32±0.02	31.31±0.18	-----

Data are expressed as mean ± S.E., n = 6

Table. 2
Variation of activity with time (drug concentration 10 µg/ml in all)

Concentration (µg/ml)	Activity (prevention of lysis %)		
	Ethanolic extract	Aqueous extract	Diclofenac
10	54.36±0.23	49.53±0.12	68.25±0.36
20	44.12±0.05	43.25±1.02	64.20±0.04
50	36.34±0.10	38.12±0.24	59.31±0.13
100	34.78±0.03	35.35±0.03	51.63±0.52
200	32.36±1.01	31.46±0.56	48.16±0.02

Data are expressed as mean ± S.E., n = 6

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