

ANTI-INFLAMMATORY ACTIVITY OF ALCOHOLIC AND AQUEOUS HEARTWOOD EXTRACTS OF *BERBERIS ARISTATA* DC.

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ABSTRACT

Objective – To evaluate the anti-inflammatory activity of alcoholic and aqueous heartwood extracts of *Berberis aristata* DC.

Method- The air dried coarsely powdered plant material (500 gm. each) was extracted with ethanol and distilled water for six hrs. Then the ethanolic extract was distilled off at low temperature under reduced pressure and water extract was completely air dried on water bath. Dried alcoholic and aqueous extracts were screened for the anti-inflammatory activity.

Result- The ethanolic and aqueous extracts of *Berberis aristata* DC. heartwood exhibited significant anti-inflammatory activity with percent inhibition 33.40% & 44.50% at a dose of 25 mg/kg, p.o., and 52.20% & 57.0% at a dose of 50 mg/kg, p.o., respectively. Whereas standard drug, Indomethacin showed an inhibition of 64.80%.

Conclusion- The alcoholic and aqueous extract of *Berberis aristata* showed their maximum effect in 4th hour. Therefore, it can be concluded that both the extracts have potential to inhibit the serotonin, histamine & prostaglandins and can be used as a potent anti-inflammatory agent.

Keywords: *Berberis aristata*, Indomethacin, Carageenan

INTRODUCTION

Berberis aristata DC. (Berberidaceae) is commonly known as Daruharidra in Bengali, Daruhald & Rasaut in Hindi. It is an erect, glabrous, spinescent shrub attaining 3-6 m of height [1]. It is distributed in temperate and subtropical parts of Asia, Europe and America. In India drug is largely collected in Chamba district of U.P. and sold in the markets of Chamba, Dehradun and Haridwar. The chief constituent is berberine, and other reported phytoconstituents are berbamine, armoline, palmatine and oxycanthine [2]. Plant is used as tonic, stomachic, antipyretic, immunogogue and also in treatment of jaundice etc. Fresh berries are useful in piles, sores and eye diseases particularly conjunctivitis. Extract prepared by milk is found useful in stomatitis and leucorrhoea. Decoction of stems mixed with that of *Curcuma longa* is recommended in gonorrhoea [3, 5]. The root extract of other species like *Berberis crataegina* showed potent anti-inflammatory, analgesic and febrifuge effects in mice and rats [6]. Synthetic agents cause many serious ill effects like liver and kidney damage, ulcers, allergic reactions etc [7, 8, 9]. The present study is aimed for evaluating the anti-inflammatory activity of alcoholic and aqueous extract of *Berberis aristata* heartwood in carageenan – induced paw edema in albino wistar rats [10].

Materials and Methods

Plant Material

The plant material was procured from AIMIL Pharmaceuticals, New Delhi and authenticated by Prof. M.P. Sharma, Taxonomist, Department of Botany, Jamia Hamdard, New Delhi. A voucher specimen was preserved in the Phytochemistry Research Lab, Jamia Hamdard (Hamdard University), New Delhi.

Extraction

The air dried coarsely powdered plant material (500 gm. each) was extracted with ethanol and distilled water for six hrs. Then the ethanolic extract was distilled off at low temperature under reduced pressure and water extract was completely air dried on water bath. Dried alcoholic and aqueous extracts were screened for the anti-inflammatory activity.

BIOLOGICAL ACTIVITY

Animals

Wistar strains of albino rats (180-230 gm each), maintained under standard animal housing conditions, were used for all sets of experiments performed on 6 rats in each groups. The rats were allowed to take standard laboratory feed with water *ad libitum*. Study was performed according to guidelines of CPCSEA & approved by Institute Animals Ethics Committee (IAEC) R.I.T., greater Noida, U.P.

Preparation of Solutions

Normal Saline Solution: Mixture of normal saline and Tween 20 was prepared in the ratio of 95:5 and administered orally.

Carageenan: 1% w/v Solution was prepared in sterile water and injected 0.1 ml sub-plantary in the right hind paw of rats.

Standard drug: Indomethacin (10 mg/kg body weight) solution was prepared with normal saline solution and administered orally.

Preparation of alcoholic extract:

Both the doses, 25 mg/ml & 50 mg/ml were prepared with normal saline solution and administered orally according to their weights.

Preparation of aqueous extract

Both the doses, 25 mg/ml & 50 mg/ml were prepared with normal saline solution and administered orally according to their weights.

Screening of anti-inflammatory activity

All the animals were weighed and a mark was made on the left hind paw just beyond tibio- tarsal junction so that every time the paw was dipped in the mercury column up to the fixed mark to ensure constant paw volume. The initial paw volume of the left hind paw was noted by mercury displacement method. The paw was dipped in the reservoir 'a' to the predetermined mark. This raised the level of

mercury in the column 'b'. The rise of mercury in the scale was measured by reading the division marked on the scale and this represented the volume of foot.

The animals were divided into six groups each consisting of six albino rats.

Group I : Control normal saline solution (0.9% w/v)
 Group II : 25 mg/kg test dose of alcoholic extract
 Group III : 50 mg/kg test dose of alcoholic extract
 Group IV : 25 mg/kg test dose of aqueous extract
 Group V : 50 mg/kg test dose of aqueous extract
 Group VI : Standard (10 mg/kg body weight)

All the groups of animals received their respective treatments and after 60 minutes, 0.1 ml of 1 % (w/v) carrageenan was injected into the sub-plantar region of the left hind paw of the rats of each groups. Paw volume was again measured at the interval of 1 hour, 2 hour, 3 hour and 4 hour. Percent inhibition of oedema volume between treated and control was calculated as follows:

$$\% \text{ inhibition} = (1 - V_t / V_c) \times 100$$

Table 1: Evaluation of anti-inflammatory activity of alcoholic & aqueous extracts of *Berberis aristata* heartwood

Groups	Paw edema					% edema Inhibition					
	1 hr	Mean ± SEM		3 hr	4 hr	5 hr	1 hr	2 hr	3 hr	4 hr	5 hr
I	0.508 ± 0.0254	0.657 ± 0.0329	0.813 ± 0.0406	0.728 ± 0.0364	0.687 ± 0.0364	-	-	-	-	-	-
II	0.411 ± 0.0205**	0.508 ± 0.0254**	0.575 ± 0.0287***	0.485 ± 0.0242***	0.480 ± 0.0240***	19.0	22.6	29.3	33.4	30.1	
III	0.374 ± 0.0187**	0.441 ± 0.0221***	0.485 ± 0.0242***	0.348 ± 0.0174***	0.387 ± 0.0193***	26.4	32.8	40.4	52.2	43.7	
IV	0.385 ± 0.0192**	0.456 ± 0.0228***	0.475 ± 0.0237***	0.404 ± 0.0202***	0.393 ± 0.0196***	24.2	30.6	38.7	44.5	40.5	
V	0.337 ± 0.0169***	0.378 ± 0.0189***	0.409 ± 0.0204***	0.313 ± 0.0156***	0.322 ± 0.0161***	33.7	42.5	49.7	57.0	53.2	
VI	0.294 ± 0.0147***	0.331 ± 0.0165***	0.340 ± 0.0170***	0.256 ± 0.0128***	0.266 ± 0.0133***	42.2	49.6	58.2	64.8	61.3	

SEM= Standard Error Mean **P < 0.01 Vs Control ; *** P < 0.001 Vs Control

Group-I (Control): Received normal saline solution
 Group-II (Low dose of Test drug): Received dried alcoholic extract of *B. aristata*
 Group-III (High dose of Test drug): Received dried alcoholic extract of *B. aristata*
 Group-IV (Low dose of Test drug): Received dried aqueous extract of *B. aristata*
 Group-V (High dose of Test drug): Received dried aqueous extract of *B. aristata*
 Group-VI (Standard): Received Indomethacin

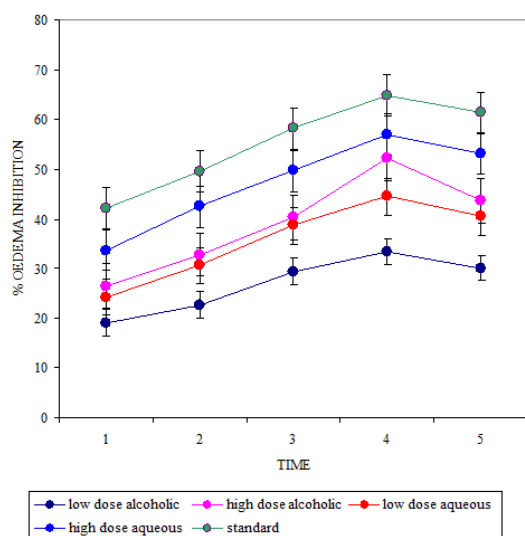


Figure1: It shows Comparable Dose Response Graph

Where: V_t is edema vol. in drug treated group & V_c in the control group.

Statistical Analysis

Significance of the results of biological estimations was calculated by ANOVA followed by Dunnett's multiple comparison t -test. The experimental results were expressed as mean ± SEM.

RESULTS

The ethanolic and aqueous extracts of *Berberis aristata* DC. heartwood exhibited significant anti-inflammatory activity with percent inhibition 33.40% & 44.50% at a dose of 25 mg/kg, p.o., and 52.20% & 57.0% at a dose of 50 mg/kg, p.o., respectively. Whereas standard drug, Indomethacin showed an inhibition of 64.80%. On comparison with standard drug, the alcoholic and aqueous extracts showed significant ($p < 0.5$) activity. The present result suggest that both the extracts possess a dose dependent significant anti-inflammatory activity while the effect of aqueous extract at dose of 50 mg/kg is more significant and comparable to that of standard drug, Indomethacin (Table-1 and comparable dose response graph)

DISCUSSION

Carrageenan induced edema in a biphasic response. The first phase was mediated through the release of histamine, serotonin and kinins, whereas the second phase is related to the release of prostaglandins and slow reacting substances which peak at 3 hr¹⁰. The alcoholic and aqueous extract of *Berberis aristata* showed their maximum effect in 4th hour.

CONCLUSION

In conclusion, this study provides evidences for the anti inflammatory activity of *Berberis aristata* which could partly contribute to its ethno medical use. However, further investigation is required to isolate the active constituents responsible for these activities and to elucidate the exact mechanisms of action. Therefore, it can be concluded that the both extracts have the potential to inhibit the serotonin, histamine & prostaglandins and can be used as a potent anti-inflammatory agent.

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