EFFECTS OF METHANOL EXTRACT OF *PIPER CHABA* STEM BARK ON CHRONIC INFLAMMATION IN RATS

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Abstract

Piper chaba Hunter (*Piperaceae*), a climbing glabrous shrub grows in plenty in southern Bangladesh. Popularly known as 'Choi' it is used as spices and believed to have medicinal value in a wide variety of disease conditions including arthritis, asthma, bronchitis and piles. Earlier studies on methanol extract of *Piper chaba* stem bark have reported anti-inflammatory activities against acute inflammation. In the present study, effect of methanol extract of *Piper chaba* stem bark on chronic inflammation has been reported. The anti-inflammatory effect was studied in rats using cotton pellet implantation method, where granuloma formation was used as an index of chronic inflammation.

Methanol extracts of *Piper chaba* stem bark given orally for 14 days daily at doses of 125 and 250 mg/kg body weight produced statistically significant ($p < 0.05$ and $p < 0.01$) anti-inflammatory effect compared to control. The percent inhibition of granuloma formation was 25% and 28% respectively, which however was less compared to aspirin (41%) and hydrocortisone (58%). The results suggest that in case of chronic inflammation, *Piper chaba* stem bark possess mild to moderate anti-inflammatory effect compared to that of aspirin and hydrocortisone.

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Introduction

The plant *Piper chaba* Hunter (*Piperaceae*) is a climbing, glabrous shrub available in various parts of India and Malay Islands.[1] In Bangladesh it is grown in plenty in the southern part particularly in Jessore, Khulna, Satkhira and Bagerhat areas. Popularly known as choi, it is used as spices in meat curry and other dishes and believed to have medicinal value in a wide variety of disease conditions including arthritis, asthma, bronchitis and piles. The crude extract was found to possess antibacterial, carminative, expectorant, analgesic, hypotensive and smooth muscle relaxant properties.[2-4] Recently, 80% aqueous acetone extract from the fruit of *Piper chaba* as well as some isolated alkamides were found to be protective against ethanol and indomethacin induced gastric lesions in rats.[5] Methanol extract of stem bark of *Piper chaba* was found to possess anti inflammatory activity in case of acute inflammation.[6]

Considering its reported anti-inflammatory properties and availability in our country, the present study was undertaken to evaluate the anti-inflammatory effect of methanol extract of *Piper chaba* stem bark, compared to steroidal and non-steroidal anti-inflammatory agents in case of chronic inflammation in rats.

Materials and Methods

The stem bark of *Piper chaba* Hunter was cut into pieces, shade-dried and grounded to coarse powder, which was then extracted with methanol at room temperature for 3 days. The filtrate concentrated in vaccuo (50°C) yielding the crude methanol extract was stored at 4°C, weighed to appropriate dosages form and diluted with normal saline prior to use.[7]

30 Long Evan Norwegian rats of either sex, weighing between 150-200g were kept under standard conditions

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of light and temperature, fed with animal pellets and allowed to drink water ad libitum.

Animals were divided into five groups each consisting of 6 rats. Chronic inflammation (granuloma) was induced by implanting one autoclaved cotton pellet (20mg) subcutaneously to each rat on 1st day.[8] They were treated as follows for 14 days: Group I served as control that received normal saline, Group II and Group III were given methanol extract of *Piper chaba* Hunter at a dose of 125 and 250mg/kg body weight respectively. Group IV were given aspirin at a dose of 100 mg/kg body weight orally and Group V were given hydrocortisone at a dose of 2 mg/kg body weight subcutaneously. On the 15th day rats were anaesthetized with ether and sacrificed. The implanted cotton pellets were dissected out, dried in the hot air oven at 60°C and then weighed. The dry weight of the granuloma was difference between the final and initial weights of the cotton pellets i.e. CPW_2–CPW_1. Percent inhibition of granuloma formation was calculated in each case by using the formula: (1-T/C)x100, where 'T' was the dry weight of granuloma for test drugs and 'C' for control.[8]

Results

The dry weight of the granuloma as measured by the formula $CPW_2 - CPW_1$ was 24.2 mg ± 0.08 mg for the control group, while those of methanol extract of *Piper chaba* (125 mg/kg b.w.), methanol extract of *Piper chaba* (250 mg/kg b.w.), aspirin (100 mg/kg b.w.) and hydrocortisone (2 mg/kg b.w.) treated groups were 18.3 ± 0.20, 17.5 ± 0.28, 14.3 ± 0.18, 10.49 ± 0.17 mg respectively. The differences compared to the control group in the dry weight of granuloma were statistically significant (Table 1). Compared to control group, the percent inhibition of granuloma formation with methanol extracts of *Piper chaba* (125 mg and 250 mg/kg b.w.), aspirin and hydrocortisone treated groups were 25, 28, 41 and 58 respectively. The percent inhibition with methanol extracts of *Piper chaba* (25% and 28%) was comparatively less than that of aspirin (41%) and hydrocortisone (58%) (Table 1).

Discussion

Methanol extracts of *Piper chaba* stem bark, given p.o. (per oral) daily for 14 days at doses of 125 and 250 mg/kg b.w. produced statistically significant ($p < 0.05$ and $p < 0.01$) anti-inflammatory effect compared to control. The percent inhibition of granuloma formation was 25% and 28% respectively, which however was less compared to aspirin (41%) and hydrocortisone (58%).

Table-1: *Anti-inflammatory effects of Piper chaba extracts, aspirin and hydrocortisone on cotton pellet induced granuloma in rat.*

Groups	Initial weight of cotton pellet(CPW_1 in mg) mean ± s.e.m.	Final weight of cotton pellet(CPW_2 in mg) mean ± s.e.m.	Increment of cotton pellet(CPW_2-CPW_1 in mg) mean ± s.e.m.	% Inhibition of granuloma formation
Group-I (Control)	20.00 ± 0.018	46.22 ± 0.18	24.2 ± 0.08	
Group-II (*Piper chaba* extract 125 mg/kg b.w.)	20.00 ± 0.018	38.34 ± 0.15	18.3 ± 0.20*	25
Group-III (*Piper chaba* extract 250 mg/kg b.w.)	20.00 ± 0.018	37.53 ± 0.20	17.5 ± 0.28**	28
Group- IV (Aspirin 100 mg/kg b.w.)	20.00 ± 0.018	34.27 ± 0.16	14.3 ± 0.18***	41
Group- V (hydrocortisone 2 mg/kg b.w.)	20.00 ± 0.018	30.37 ± 0.18	10.49 ± 0.17***	58

* $P < 0.05$ in a test of significance difference from control.
** $P < 0.01$ in a test of significance difference from control.
*** $P < 0.001$ in a test of significance difference from control.

Earlier studies showed a significant anti-inflammatory effect of methanol extract of *Piper chaba* stem bark in case of acute inflammation.[6] However, no report is available in case of chronic inflammation.

Chronic inflammation is the result of a balance between progressive tissue damage on the one hand and eradication of the damaging stimulus, followed by healing and scar formation, on the other. Granulomatous inflammation is a special type of chronic inflammation, characterized by the presence of granulomata. A granuloma consists of mononuclear cell infiltrate composed of macrophages, lymphocytes and plasma cells. The main features of chronic inflammation are- tissue destruction, granulation tissue formation, i.e. proliferation of fibroblasts, small blood vessels and fibrosis.[9]

The results suggest that *Piper chaba* stem bark possess mild to moderate anti-inflammatory effect compared to aspirin and hydrocortisone in case of chronic inflammation. However, further studies should be carried out to isolate the active principle and to evaluate further its dynamics, kinetics and safety profile before it can be recommended for clinical use.

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