

IMMUNOPHARMACOLOGICAL STUDIES ON PICRORHIZA KURROA ROYLE-EX-BENTH PART V : ANTI-INFLAMMATORY ACTION : RELATION WITH CELL TYPES INVOLVED IN INFLAMMATION

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Summary : Relative importance of various cells involved in inflammation and in anti-inflammatory action of *P. kurroa* extract (PK) was investigated in albino rats. Effects of chemical depletion of macrophages and polymorphs and a functional deprivation of mast cells and platelets were examined on carrageenin induced pedal inflammation as well as on anti-inflammatory effect of PK treatment in this test. Such depletions/functional deprivements altered the inflammatory response in conformity with the known role of these cells. The anti-inflammatory effect of PK treatment was counteracted at 1 hr, 3 hr and 5 hr post-insult intervals by mast cell, neutrophil and macrophage depletion respectively. Manipulation of platelets was without effect.

Involvement of multiple target cells favours a nonspecific mechanism as basis of anti-inflammatory action of *P. kurroa*.

Key words : anti-inflammatory effect picrorhiza kurroa leukocytes

INTRODUCTION

Earlier studies on rhizome extract of picrorhiza kurroa (PK) (Fam. Scrofulariaceae) revealed a delayed anti-inflammatory effect suggesting to be through changes in structure/function of inflammatory cell-membranes (7). A variety of cell types are involved as effectors/modulators of inflammatory responses (3). Observations in the present study seek information on the role of various proinflammatory cell types in anti-inflammatory action of PK.

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MATERIAL AND METHODS

Preparation of water soluble fraction of the alcoholic extract of *P. kurroa* (PK) was described in earlier reports (5, 6). A 3-day once daily oral treatment schedule with PK (extractives from 100 mg of parent alcoholic extract/kg) or equivalent volumes of distilled water (DW) in male Wistar rats (100-150 g) was adopted (7). Anti-inflammatory effects of PK treatment against carrageenin-induced pedal oedema (10), were compared in normal rats and rats depleted or functionally deprived of proinflammatory cell types as under :

Rats were depleted of macrophages by carrageenin (300 mg/kg, ip; 9) and 'treatment' were given 8 hr later. Neutrophils were depleted by vinblastin sulphate (0.5 mg/kg, ip; 4) and treatments given 3 days later. Mast cells were functionally depleted by compound 48/80 (1 mg/kg, ip; 4 injections at 6 hr intervals) and treatments started a day later. Sulphinpyrazone was given (50 mg/kg, ip) 2 hr after completion of treatments. In first three types of experiments carrageenin pedal oedema test was done 2 hr after last dose of a 'treatment'. In experiments with platelet, the pedal oedema test followed 1 hr after sulphinpyrazone.

Group differences were analysed with students 't' test. Wherever necessary, the group values were subjected to preliminary screening for differences in variance with F values more than 1.

RESULTS

Carrageenin induced pedal oedema attained peaks at 3 hr with subsequent decline in both DW control and PK treated rats (Table I, group I). PK-treatment suppressed inflammatory oedema at each of the 1 hr, 3 hr and 5 hr assay period postinsult.

Inflammatory reaction in macrophage depleted animals did not significantly differ from that exhibited by normal ones, though the magnitude in the former animals were somewhat lower in either of the 3 hr and 5 hr assay postinsult. Such depletion did not affect the anti-inflammatory effect of PK (Table I, gr. II. i) Neutrophil depletion significantly inhibited carrageenin inflammation assayed 3 hr, after insult. PK treatment failed to render any additional inhibition of inflammatory oedema in such animals (Table I, gr. II. ii).

Mast cell depletion particularly reduced oedematous inflammatory reaction developed at 1 hr postinsult; the influence was not however, statistically significant. Such depletion counteracted anti-inflammatory effect of PK at 1 hr postinsult; but significant anti-inflammatory effect was apparent at 3 hr. Pretreatment with platelet antiaggregatory drug sulphinpyrazone neither affected carrageenin inflammation nor the anti-inflammatory effect of PK treatment in the singular assay at 3 hr postinsult (Table I, gr. II. iv).

TABLE I : Effect of Picrorhiza kurroa extract (PK)-treatment on carrageenin-induced paw oedema in albino rats with deprived function of major inflammatory effector cells.

Group	n	% Increment in paw volume (Mean±SE)		
		Post insult time of assay		
		1 hr	3 hr	5 hr
I. Normal rats				
Control	5	18.4±2.1	33.4±3.2	29.2±2.8
PK-treated	8	10.3±1.8**	17.3±3.8***	15.2±2.6***
II. Rats with deprived function of effector cells				
i) Macrophage				
Control	6		28.2±3.3	23.1±3.2
PK-treated	6		16.4±3.1***	12.7±3.6*
ii) Neutrophil				
Control	6		15.4±1.2	
PK-treated	7		13.2±2.9	
iii) Mast cell				
Control	5	12.6±3.0	29.9±2.7	
PK-treated	6	8.4±1.1	18.3±4.7**	
iv) Platelets				
Control	7		37.1±5.2	
PK-treated	6		15.1±3.9**	

Asterics indicate P value with respective controls in the same group as :

*<0.1, **<0.02 and <0.01 respectively

F values, when VARIANCE of any of the PK treatment groups in II are examined with both the control and treated group values in I, are uniformly above '1'

DISCUSSION

Mononuclear cell participation in carrageenin induced acute inflammation commences primarily after 4 hr (3). Somewhat lower oedema magnitudes in macrophage depleted animals at 5 hr postinsult assay, agree with the said literature. The decline in oedema from 3 hr to 5 hr may indicate that events preceding macrophage recruitment are more important in carrageenin inflammation. Anti-inflammatory effect of PK at 5 hr in macrophage depleted animals is considerable (i.e. 45%) which locates macrophage as a minor target of PK action. Results at 3 hr indicate involvement of other cell types in anti-inflammatory action of PK.

Marked inhibition of carrageenin oedema observed in neutrophil depleted animals is in agreement with suggested major role of neutrophils in carrageenin inflammation over 3 hr (3). Very low (14%), anti-inflammatory effect of PK after neutrophil depletion, confirms neutrophils as major target for anti-inflammatory action of PK.

Removal of mast cell influence leads to some reductions (33% and 39% at 1 hr and 3 hr respectively) in inflammatory oedema. Results obtained after mast cell depletion agree with minor role ascribed to mast cells in initial phases of inflammation (2). Such depletion overtly prevents anti-inflammatory effect of PK treatment in 1 hr and reduction at 3 hr. Mast cells

may thus appear as one target for PK action at early period. The finding is also in conformity with reported action of PK, i.e. stabilizing the mast cell membrane (6).

Platelets are capable of serving a proinflammatory role by releasing potent inflammatory mediators when aggregated by injury or other insult (8). Lack of effect of sulphinyprazone on carrageenin oedema or the anti-inflammatory effect of PK, negates any role of platelets in either of the phenomena.

Anti-inflammatory action of PK thus seems related to membranes of two cells responsible for inflammatory reaction (5). Receptor types on cell membrane vary from cell to cell. The present observations thus, favour mechanism of anti-inflammatory PK effect to be a non-specific membrane action.

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