EVALUATION OF ANTIALLERGIC ACTIVITY OF THE VARIOUS EXTRACTS OF THE AERIAL PART OF *ACHYRANTHES ASPERA* VAR. PORPHYRISTACHYA (WALL. EX MOQ.) HOOK.F.

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Summary

Various extracts of the aerial parts of the *Achyranthes aspera* were evaluated for antiallergic activity. Plant may show antiallergic activity as the plant shows potent antiasthmatic activity where allergy is one of the pathological condition observed. Petroleum ether extract (200 mg/kg, i.p.) of the plant shows significant antiallergic activity in both milk induced leukocytosis and milk induced eosinophilia in mice. Thus the antiallergic activity of *A. aspera* may be due to nonpolar constituents. The phytochemical screening of petroleum ether extract shows the presence of steroids. Literature shows the presence of steroids like β-sitosterol, ecdysone and ecdysterone. Thus these steroids present in the plant may be responsible for the antiallergic activity.

Keywords: *Achyranthes aspera*, milk induced leukocytosis, milk induced eosinophilia

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Introduction

Several medicinal properties have been attributed to the plants in the traditional system of medicine. The presence of antistress (adaptogenic) properties in some plant materials is being one of them, as described to be tonics in the Ayurvedic system of medicine[1]. The most important characteristic of an adaptogen (antistress), is that it increases resistance to adverse influences of a wide range of factors of physical, chemical and biological nature; and its normalization action, which reveals itself irrespective of the direction of the previous pathologic shifts[2]. Ayurveda provides a number of herbs for the treatment of asthma and herbal formulations used for the treatment of asthma include some antistress (nervine support) herbs to enable adoption to stress, since excessive stress or nervous debility may aggravate the symptoms of asthma. After parenteral administration of milk there is increase in TLC, and this stressful condition can be normalized by administration of an antistress or adaptogenic drug[2]. Furthermore leukocytes recruited during asthmatic inflammation release the inflammatory mediators like cytokines, histamine, and major basic protein and promote the ongoing inflammation. This model was used to evaluate the protective effect of *A* .aspera extract against milk-induced leukocytosis.

Eosinophilia is an abnormal increase in peripheral eosinophil count to more than 4 % of total leukocytes[3]. In the late phase, especially in the development of allergic asthma, eosinophils play role as an inflammatory cell. Eosinophil secretes mediators such as eosinophil cationic protein (ECP), eosinophil derived neurotoxin (EDNT), granulocyte macrophage colony stimulating factor (GM-CSF), tumor necrosis factor (TNF), and Prostaglandin (PG), which results in epithelial shedding, bronchoconstriction and promotion of inflammation in respiratory tract[3, 4]. Eosinophilia is associated with respiratory disorder, often allergic in nature together with pulmonary infiltrates that are detectable on chest films[5]. Most of the literatures do not include a diagnostic evaluation and precise practical clinical approach to eosinophilia. It was also demonstrated that parental administration of milk produces a marked and significant increase in the leukocytes/eosinophils count after 24hr of its administration[6].

In this model milk in a dose of 4 ml/kg was administered subcutaneously and absolute eosinophil count was taken before and after administration on milk was calculated[7].

Materials and methods

Plant Material

Mature aerial part of *A. aspera* was collected from Ahmednagar district of Maharashtra in June 2008, cleaned and dried at room temperature in shade and away from direct sunlight and authenticated by Mr. P.G. Divakar, Botanical Survey of India, Pune, where a sample specimen (voucher number: AAPSBDPL1) has been deposited.

Extraction

Dried and coarsely powdered aerial part of *A.aspera* was subjected to successive solvent extraction in Soxhlet extractor using petroleum ether, chloroform, ethyl acetate, and ethanol as solvent and the marc left was refluxed with water. All the extracts were vacuum dried to produce PEE (1.28%), CLE (0.716%), EAE (0.58%), ETE (6.94%), and AQE (7.14%), respectively.

Animals

Male albino mice (Swiss strain) weighing 25-28 g were housed under standard laboratory conditions, in groups of five each. The animal had free access to food and water. The ethical committee of the institute approved the protocol of the study.

Drugs and Chemicals

The following drugs and chemicals were used. petroleum ether AR (60-80°C) (PCL, India), chloroform AR (PCL, India), ethyl acetate AR (PCL, India), ethanol AR (PCL, India), and tween 80 AR (PCL, India).

Milk induced leucocytosis[2].

Brekhman and Dardymov, (1969) described this method. Mice were divided into six groups, six animals in each group. Animals belonging to group I, II, III, IV, V and VI received boiled and cooled milk injection in dose of 4ml/kg, (s.c.). Animals belonging to groups II to VI received petroleum ether, chloroform, ethyl acetate, ethanol and aqueous extracts of *A. aspera* (200 mg/kg, i.p.) respectively. Blood samples were collected from each mouse from the retro orbital plexus, under light ether anesthesia. Total leukocyte count was done in each group before drug administration and 24 hr after milk injection. Difference in Total leukocyte count before and 24-hr. after drug administration was calculated.

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Phytochemical screening of active extract.

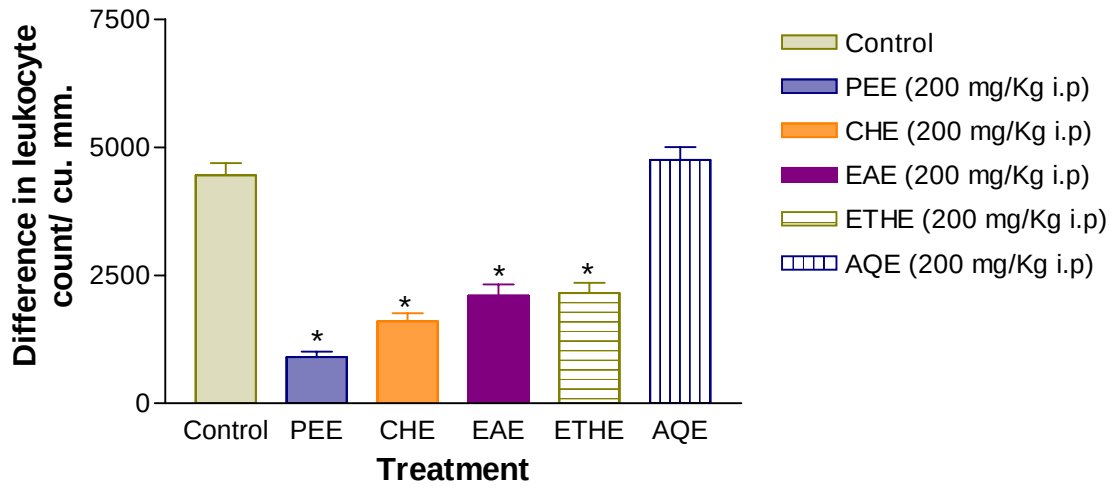
Various phytochemical studies including test for carbohydrates, proteins, alkaloids, glycosides, steroids, flavonoids, tannins and phenolic compounds were carried out[8].

Results

Milk Induced Leukocytosis in Mice

Subcutaneous injection of milk in dose of 4 ml/kg, produced a significant ($p < 0.01$) increase in the leukocyte count after 24 hr of its administration. Mice pretreated with petroleum ether, chloroform, ethyl acetate, ethanol and aqueous extract of *A. aspera* have exhibited significant difference in total leukocytes before and after drug treatment. Petroleum ether extract of *A. aspera* has inhibited the milk induced leukocytosis ($p < 0.05$) with dose of 200 mg/kg, (i.p.).

Graph 1. Effect of various extracts of aerial parts of *A. aspera* on milk induced leukocytosis in mice (200 mg/kg)

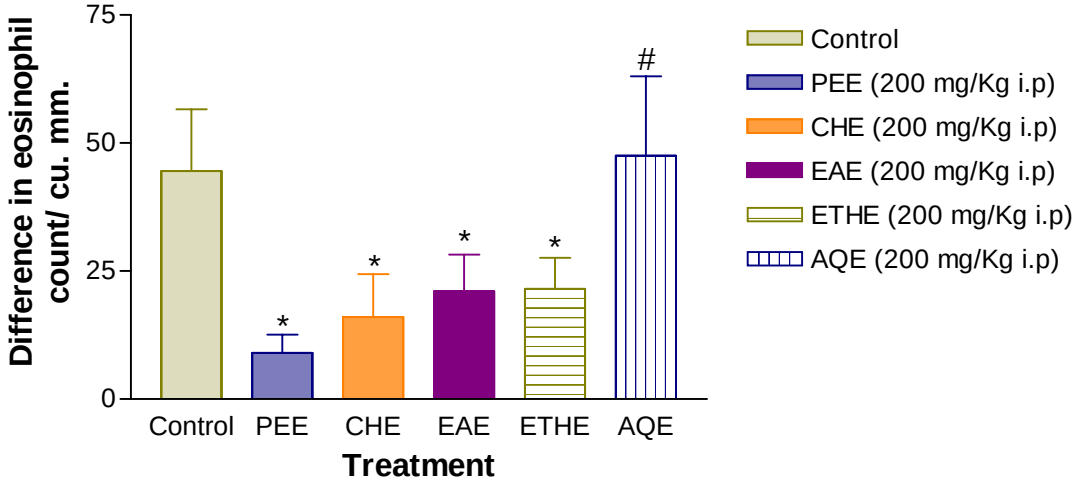


*P<0.01 compared to vehicle treated group (One way ANOVA followed by Dunnett's test

Milk Induced Eosinophilia in Mice

Subcutaneous injection of milk in dose of 4 ml/kg, produced a significant ($p < 0.01$) increase in the eosinophil count after 24 hr of its administration. Mice pretreated with petroleum ether, chloroform, ethyl acetate, ethanol and aqueous extract of *A. aspera* have exhibited significant difference in eosinophil count before and after drug treatment. Petroleum ether extract of *A. aspera* has inhibited the milk induced eosinophilia ($p < 0.05$) with dose of 200 mg/kg, (i.p..).

Graph 2. Effect of various extracts of aerial parts of *A. aspera* on milk induced eosinophilia in mice (200 mg/kg)



*P<0.01 compared to vehicle treated group (One way ANOVA followed by Dunnett's test
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Conclusions

Phytochemical screening of petroleum ether extract shows the presence of steroids. Literature shows the presence of steroids like β-sitosterol, ecdysone and ecdysterone. Thus these steroids present in the plant may be responsible for the antiallergic activity. Literature shows that β-sitosterol is having antianaphylactic and antiasthmatic activity[9].

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