In-vitro Antihistaminic and Antispasmodic Study of Aqueous Extract of Leaves of Bryophyllum pinnatum L.

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Abstract: The present study was undertaken to evaluate in-vitro antihistaminic and anti-spasmodic activity of aqueous extract of leaves of Bryophyllum pinnatum L. by using isolated goat tracheal chain and chicken ileum preparation. The cumulative concentration response to histamine and Acetyl choline in the absence and presence of aqueous extract were recorded with a kymograph. The aqueous extract decreases the goat tracheal contraction and ileum contractions induced by histamine and Acetyl choline in dose dependent manner as compared with standard drug Chlorpheniramine Maleate and Atropine sulphate respectively. This revealed that aqueous extract of leaves of Bryophyllum pinnatum L possess a high degree of antihistaminic and spasmyloytic activity by blocking histaminic and cholinergic receptors.

Key words: Anti-histaminic, Anti-spasmodic, Bryophyllum pinnatum, Histamine.

1. Introduction

Histamine is an organic nitrogenous compound involved in local immune responses and has a central role as a mediator of pruritus. Despite of an improvement in the general health of the population, the prevalence of allergy and asthma has risen in the recent years [1]. As part of an immune response to foreign pathogens, histamine is produced by basophils and by mast cells found in nearby connective tissues. Release of histamine increases the permeability of the capillaries to white blood cells and some proteins, to allow them to engage pathogens in the infected tissues [2]. Various epidemiological studies have identified that histamine increase in the prevalence of upper and lower respiratory tract allergic diseases including asthma [3]. The antihistaminic drugs were extensively used to combat the effects produced by histamine. Similarly, different spasmyloytics have been used for the treatment of various gastrointestinal disorders including diarrhea and irritable bowel syndrome [4]. Moreover effects of these drugs are quite disturbing and having serious side effects. Hence a continuous search is on going to identify effective and safe remedies to treat bronchial asthma and gastrointestinal disorders [5]. Ayurveda, an Indian system of medicine, has described several medicinal plants as a source of drugs for treatment of bronchial asthma and allergic disorders. Thus, the Indigenous plant Bryophyllum pinnatum Linn is an environmental weed from the family Crassulaceae commonly known as Parnabeeja [6] was evaluated for its antihistaminic and antispasmodic activity.

2. Materials and Methods

2.1 Chemicals and Drugs

Kreb’s-Henseleit solution, Tyrode solution Histamine (Johan Baker Inc. Colorado, USA), Atropine sulphate & Acetyl choline (Ach) (S.D. Lab Chem., Mumbai) and Chlorpheniramine Maleate (CPM) (Research Lab Fine Chemicals). All the ingredients of solutions of analytical grade and prepared freshly before the experiment.

2.2 Collection of Plant Material and Preparation of Extract

The fresh leaves of Bryophyllum pinnatum L were collected locally from Sangli District of Maharashtra, India and are authentified by Dr. S. M. Shendage, Balwant College, Vita, India. A voucher specimen has been kept in the herbarium (SBN 001-2016-17) at Department of Botany. The aqueous extract was prepared by blending and macerating 500
gm of fresh dried leaves of *Bryophyllum pinnatum* with 100 ml of distilled water at 40°C for 24 hrs. The resulting mixture was filtered & kept for evaporation. The crystals were obtained are dissolve in distilled water to prepare its dose.

2.3 Preliminary Phytochemical Testing

Various phytochemical tests for tannins, saponins, carbohydrates, glycosides, phenols and flavonoids were carried for aqueous extract according to procedure [7].

2.4 Goat Tracheal Strip Preparation

The antihistaminic assay of aqueous extract on goat tracheal strip preparation was carried out according to method described by Kulkarni with slight modification [8, 9]. In this method, fresh goat trachea brought from local slaughter house was cut into zigzag fashion. It was suspended in an organ bath of 20 mL Krebs-Henseleit solution maintained at 37 ± 1°C with continue aeration. One end was tied to aerator tube and other attached to isotonic frontal writing lever to Kymograph paper on Sherrington rotating drum. Tissue was allowed to equilibrate for 45 min during which, the bathing solution was changed frequently after every 5 minute. The contractile responses of tracheal strip to histamine (10 µg/ml) with doses of 0.1ml, 0.2ml, 0.4ml, 0.8ml and 1.6ml were recorded in absence and presence of aqueous extract of *Bryophyllum pinnatum* (ABP)(100 µg/ml) by using Sherrington’s Recording Drum with a frontal writing lever. The similar concentration-effect curve was taken in presence of standard drug Chlorpheniramine Maleate (1 µg/ml). The height of response curve was measured to express percentage inhibition. The graph was plotted by taking log dose verses height of response curve.

2.5 Isolated Chicken Ileum Preparation

The antispasmodic assay of aqueous extract was performed according to procedure [8, 10]. The fresh chicken ileum was collected from local slaughter house in Tyrode solution and cleaned off the mesentery. The segment of 2cm long was mounted in a 20ml tissue organ bath and maintained at 37°C. The issue was allowed to equilibrate for 30 min, during which, the bathing solution was changed at every 10 min. Contact time of 60 sec, and base line of 30sec time cycle were opted for proper recording. Cumulative concentration-effect curves were recorded on kymograph for Acetyl choline (1 µg/ml) in absence and presence of aqueous extract of *Bryophyllum pinnatum* (100 µg/ml) on Kymograph by using Sherrington’s Recording Drum. The same procedure was carried for concentration-effect curve of Ach in presence of Atropine sulphate as a standard drug. The percentage inhibition of extract and standard drug was calculated and graph was plotted by taking log dose verses height of response curve.

3. Results and Discussion

Preliminary phytochemical evaluation of extract showed presence of saponins, flavonoids, tannins, glycosides and carbohydrates etc. The percentage yield was obtained about 6.7% w/w. The antihistaminic effect of aqueous extract of *Bryophyllum pinnatum* was carried out using histamine induced contraction on goat tracheal strip preparation. Histamine contracts the tracheo-bronchial muscle of guinea pig, goat, horse, dog and man. Goat tracheal chain is much more sensitive and easier to handle [5]. Histamine produced dose dependent increase in contraction of goat tracheal chain and thus subsequent increase in height response curve which was significantly inhibited by aqueous extract of *Bryophyllum pinnatum* as compared to standard drug Chlorpheniramine Maleate (Table 1). The antagonistic effect of aqueous extract towards histamine was indicated from the right shift of cumulative dose response curve of histamine in presence of aqueous extract (Figure 1) which may be due to blocking H1 receptors.

<table>
<thead>
<tr>
<th>Dose of Histamine (ml)</th>
<th>Height of DRC (mm)</th>
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<tbody>
<tr>
<td></td>
<td>Histamine</td>
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<td>4</td>
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<td>0.2</td>
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<td>0.4</td>
<td>11</td>
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<td>0.8</td>
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Figure 1. Antagonistic effect of ABP to Histamine

Similarly, spasmolytic (antispasmodic) effect of aqueous extract of *Bryophyllum pinnatum* was also evaluated by observing its decrease in Ach induced ileal contractions (Table 2) and shift the dose response curve of Atropine (Figure 2) to right side. These indicate that *Bryophyllum pinnatum* has spasmolytic activity on gastrointestinal smooth muscles. This effect may be contributes by anticholinergic action.

Table 2. Antispasmodic effect of aqueous extract of *Bryophyllum pinnatum* on chicken ileum

<table>
<thead>
<tr>
<th>Dose of Ach (ml)</th>
<th>Height of DRC (mm)</th>
<th>Ach</th>
<th>CPM + Ach</th>
<th>ABP+ Ach</th>
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</thead>
<tbody>
<tr>
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<td>4</td>
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<tr>
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4. Conclusion

Our present study showed that, aqueous extract of *Bryophyllum pinnatum* is capable of inhibiting the response of wide range of contractile stimuli such as Histamine and Ach on tracheal and intestinal smooth muscles. Therefore possessing wide range of antihistaminic and spasmylocytic action which may be useful in the treatment of respiratory and GIT disorders. Further study regards to isolation, purification, mechanisms and pharmacological screening of the active principles responsible for the activity is to be carried out.

5. Acknowledgement

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6. References


