Characterization and Pharmacological Activities of an Isolated Compound from Alcoholic Extract of Desmostachya bipinnata Underground Parts

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Abstract: Desmostachya bipinnata (L.) Stapf [Gramineae (Poaceae)] is used in Indian traditional medicines system for treatment of various diseases such as asthma, kidney stone, diarrhea, wound healing, etc. Some of these traditional claims require authentication by performing animal testing. In present study, D. bipinnata was extracted in alcohol. An alkaloid was isolated from the prepared extract, characterized and evaluated for antidiarrheal and antiasthmatic activity. Isolated compound was characterized by melting point, chemical tests, FT-IR, LC-MS and NMR. Antidiarrheal activity of the isolated compound was evaluated by castor oil and magnesium sulphate induced diarrhea in Wistar rats. Castor oil and magnesium sulphate induced diarrhea in Wistar rats were treated with prepared D. bipinnata alcoholic extract, isolated compound and loperamide. Antiasthmatic activity of the isolated compound was evaluated by isolated guinea pig ileum and histamine induced lethality test. The isolated Compound I was 4-(2-(dimethylamino) ethyl) phenol. Isolated Compound I was 70.16% (P<0.01) effective in reducing feces in castor oil induced diarrheal rats and 70.94% (P<0.01) feces reduction in magnesium sulphate induced diarrheal model. Loperamide, a standard anti diarrheal drug, was effective in reducing number of feces by 70.94% in castor oil induced diarrheal rats and 71.77% reduction in feces in magnesium sulphate induced diarrheal rats. Isolated compound I showed 97.67% contraction, at 0.8 ml dose, where as chlorpheniramine showed 100% contraction, at 0.8 ml dose in isolated guinea pig ileum model. Isolated compound I and cetirizine, both, showed 100% protection against histamine induced lethality with no grasping and difficulty in breathing. The isolated compound was pharmacologically active as anti diarrheal and antiasthmatic in tested animals.

Keywords: Desmostachya bipinnata, diarrhea, asthma, castor oil, magnesium sulphate, rats.

1 Introduction

Desmostachya bipinnata (L.) Stapf [Gramineae (Poaceae)] (Syn: Eragrostis cynosuroides) is commonly known as sacrificial grass, kusha [1], drabh [2] and dab [3]. D. bipinnata contains flavonoids like kaempferol, quercetin, quercetin-3-glucoside, trycin, trycin-7-glucoside; coumarins like scopoletin and umbelliferone; sugars; amino acids; and carbohydrates [4]. Camphene, β-eudesmol, eseroline, and calarene are the main components of the D. bipinnata oil. Other phytoconstituents like diphenylketonum bromide, limonene, 2-cyclohexene-1-one, and 8-nitro-12-tridecanolide have also been isolated in small amounts from D. bipinnata [5]. Leaf paste of D. bipinnata is used to cure cuts and wounds [6]. Roots are used in treatment of asthma, rheumatism [7], carbuncles, piles, cholera, dysuria [2], diuretic, galactagogue, astringent [1], dysentery, leucorrhoea and wounds [1].

Herbal treatment for diarrhea and asthma in natural and traditional medicinal practices includes use of plants or plant extracts such as Semicarpus anacardium, Achyranthus aspera, Rhus semialata [8] D. bipinnata[7], Elytraria acaulis[9, 10]. Antiasthmatic activity of D. bipinnata has been investigated for alcoholic and aqueous extracts of D. bipinnata [11] by castor oil induced diarrheal model in Wistar rats and charcoal meal stimulated gastrointestinal transit in albino mice. Literature review revealed that antiasthmatic activity of D. bipinnata has not yet been investigated. Furthermore, active constituents responsible for antiasthmatic and antiasthmatic activity have not been isolated from D. bipinnata

The present study was carried out to identify active constituents responsible for traditional claims for anti diarrheal and antiasthmatic activity of D. bipinnata. Extract of D. bipinnata were prepared in alcohol. An isolated compound and prepared extract were evaluated for antidiarrheal activity by castor oil induced diarrhea and
magnesium sulphate induced diarrhea models in Wistar rats. Isolated guinea pig ileum and histamine induced lethality tests in guinea pig were conducted to evaluate antiasthmatic activity.

2 Materials and methods

2.1 Plant material: collection, authentication

The whole plants of *D. bipinnata* were collected from Chirawa and Jhunjhunu districts of Rajasthan, India. The collected plant was authenticated by Dr. R. P. Pandey from Botanical Survey of India, Jodhpur, India. A voucher specimen, JNU/PH/2010/D b D2, was deposited in the herbarium of Jodhpur National University, Jodhpur, India.

2.2 Extraction and isolation

Underground parts of *D. bipinnata* were grinded in electric mixer-grinder and screened using BSS standard sieves. Powder which passed through sieve no. 22 (average aperture size 710 μm) and retained on sieve no. 44 (average aperture size 355 μm) was selected and used for extraction. The powdered crude drug was packed in a paper cylinder made from a filter paper and placed in the body of soxhlet extractor. The solvent was poured in soxhlet extractor and allowed to run for 3-4 cycles. After that, the apparatus was fitted in appropriate manner and drug was extracted. The obtained extracts were filtered through Whatman filter paper, concentrated to syrupy mass, acidified with dilute sulphuric acid and filtered. Filtrate was decolorized with charcoal, made slightly alkaline by adding sodium carbonate and extracted with petroleum ether in separating funnel. On evaporation of ethereal extract, needle shaped crystals were obtained. These crystals of the isolated compound were further purified by recrystallization in boiling water.

2.3 Characterization of isolated compound

2.3.1 Melting point determination

Melting point of isolated compound was determined by melting in a glass capillary. Isolated compound was filled in a glass capillary tube fused at one end and heated in a melting point apparatus. The melting point was determined as the temperature when the isolated compound melted in the capillary tube.

2.3.2 Xanthoproteic test

Isolated compound was treated with concentrated nitric acid, a white precipitate was observed. It was boiled and cooled. Further to it, sodium hydroxide (20%) or ammonia was added. Orange color developed in solution indicated the presence of aromatic amino acids [12].

2.3.3 Million’s test

Isolated compound was treated with Millon’s reagent. The formation of white precipitate, which turned to red upon heating, indicated the presence of proteins and amino acids [12].

2.3.4 Ferric chloride test

Isolated compound was treated with ferric chloride. Reddish blue coloration was observed [12].

2.3.5. Thin layer chromatography

Thin layer chromatographic plates of isolated compound were prepared using solvent system, n-butanol:acetic acid:water (60:15:25). Iodine and Dragendorff’s reagent were used as detecting reagents.

2.3.6 IR spectroscopy

KBr disk of Compound I was prepared and IR spectrum of Compound I was recorded using Perkin Elmer 1600 FTIR (USA) spectrophotometer.

2.3.7 LC-MS spectroscopy

The mass spectrum of Compound I was recorded using TOF MS ES micromass spectrometer.

2.3.8. NMR spectroscopy

The $^1$H and $^{13}$C NMR spectrum of Compound I was recorded using Bruker Advance II 400 NMR spectrometer in deuterium substituted water (D$_2$O) using tetramethylsilane (TMS) as internal standard (chemical shifts in δ ppm).

2.4 Pharmacological evaluation of isolated compound

2.4.1 Antidiarrheal activity

Magnesium sulphate induced diarrhea and castor oil induced diarrhea models in rats models were used for evaluation of antidiarrheal activity.

2.4.1.1 Animals

The study was conducted on healthy female Wistar rats of either sex weighing 150-200 g. The animals were randomly distributed to 7 different groups, each consisting of 5 animals. Prior approval by Institutional Animal Ethical Committee (IAEC), registration number 1258/ac/09/CPCSEA, was obtained for conduct of animal experiments. The animals were kept in colony cages at standard husbandry conditions. All animals had free access...
to feed and water *ad libitum*.

### 2.4.1.2. Experimental procedure

Healthy Wistar rats were marked and randomly distributed to 4 groups, each consisting of 5 animals. The groups were given treatments as follows:

- Group I: Normal control (1% CMC; 10 ml/kg, body weight)
- Group II: Standard drug (loperamide 3 mg/kg body weight)
- Group III: Diarrheal rats, treated with alcoholic extract of *D. bipinnata*
- Group IV: Diarrheal rats, treated with isolated Compound I

All animals were initially screened for induction of diarrhea by administering 1 ml of castor oil. Only animals which developed diarrhea were selected for antidiarrheal studies.

### 2.4.1.3. Castor oil induced and magnesium sulphate induced diarrhea in rats

Wistar rats weighing 150-200 g were selected and kept for overnight fasting. Loperamide, *D. bipinnata* extract (500 mg/kg body weight) and isolated Compound I were administered orally by gavage.

For castor oil induced diarrhea, 1 ml of castor oil was administered orally to each animal after one hour after administration of drug/extract. For magnesium sulphate induced diarrhea, magnesium sulphate was administered at dose 2 g/kg orally to each animal, thirty minutes after administration of drug/extract. All animals were placed in cages, where floor was lined with non-wetting paper sheets of uniform weight. Non-wetting paper sheets were changed per hour up to 6 hour. Characteristic diarrheal droppings per hour up to 6th hour were recorded after draining the urine by gravity.

A numerical score based on stool consistency was assigned. Normal stools were assigned a score 1, semi solid stools as 2 and watery stools as 3. Mean of diarrheal droppings passed by treated animals in various groups were compared to control group [13-15].

### 2.4.1.4. Statistical analysis

The data obtained in the studies was analyzed by one way analysis of variance (ANOVA) followed by Dunnett’s ‘*t*’ test using GraphPad Prism Version 5.01 (Graph Pad Software Inc., U.S.A) to compare different groups with control. P-value <0.01 was considered significant and results were expressed as mean ± SD.

### 2.4.2. Antiasthmatic activity

#### 2.4.2.1. Isolated guinea pig ileum

Isolated guinea pig ileum was used for evaluating contractile responses of test compound and extract. Concentration response curves of various drugs have been prepared and compared to evaluate antiasthmatic activity [15, 16]. Overnight fasted guinea pigs were sacrificed, dissected to cut and remove ileum. Ileum was mounted in an organ bath containing tyrode solution continuously aerated at 37±0.5°C. Concentration response curves for histamine, chlorpheniramine with histamine, drug extract with histamine and isolated compound with histamine in Tyrode solution were plotted.

#### 2.4.2.2. Histamine induced lethality in guinea pig

##### 2.4.2.2.1. Animals

The study was conducted on healthy guinea pigs of either sex weighing 200-300 g. Prior approval by IAEC (registration number 1258/ac/09/CPCSEA) was obtained for conduct of experiments. The animals were kept in colony cages at standard husbandry conditions and had free access to feed and water *ad libitum*.

##### 2.4.2.2.2. Experimental procedure

Healthy guinea pigs were marked and randomly distributed to 4 groups, each consisting of 5 animals. These animal groups were given treatments as follows:

- Group I: Normal control
- Group II: Standard drug (Cetirizine 0.5 mg/kg)
- Group III: Asthmatic guinea pigs, treated with alcoholic extract of *D. bipinnata*.
- Group IV: Asthmatic guinea pigs, treated with isolated Compound I

#### 2.4.2.2.3. Induction of asthma and its treatment

Asthma was induced in guinea pigs as per standard procedure [17]. The plant extracts were dissolved in distilled water and administered by oral feeding needle. After one hour of treatment, fur was removed from either left or right leg of guinea pigs with help of blade and area was cleaned with warm cotton swab to dilate pedal vein. After that histamine hydrochloride solution (1.25 mg/kg body weight) was injected intravenously to each animal. Animals in each group were observed for convulsions and lethality up to one hour after histamine injection.
3 Results and discussion

Our previous studies reported the presence of alkaloids, flavonoids, steroids, glycosides, and coumarins in alcoholic extract of *D. bipinnata* [18].

3.1. Extraction and isolation

*D. bipinnata* underground parts (480 g) were extracted with alcohol from which 115 mg of alkaloid was obtained after isolation.

3.2. Characterization of isolated compound

Melting point of the isolated Compound I was 117-118°C as determined by melting a glass capillary tube. It gave positive xanthoproteic test, Millon’s test and a reddish blue color with ferric chloride. TLC of isolated Compound I in solvent system n-butanol: acetic acid: water (60:15:25) gave Rvvalue 0.44 (Figure 1). IR spectrum is presented in Figure 2. IR spectrum had shown characteristic peaks at 1313 cm⁻¹ (C-N stretch); 2797 and 2856 cm⁻¹ (C-H stretch); 1464, 1514, 1589, and 1612 cm⁻¹ (C=C stretch); 818 and 841 cm⁻¹ (C-H out of plane bending, p-di-substituted benzene).

![Figure 1](image1.png)

Figure 1. TLC of alcoholic extract of *D. bipinnata* and isolated Compound I

![Figure 2](image2.png)

Figure 2. IR spectrum of isolated Compound I

The mass spectrum of compound I was recorded on TOF MS ES micromass spectrometer. The m/z for compound I was found to be 165 (*C_{10}H_{14}ON*), 121(*C_{6}H_{9}O*), 103 (*C_{6}H_{7}*), 93(*C_{6}H_{10}O*), and 77(*C_{6}H_{5}*). The mass spectrum is shown in Figure 3(a) and 3(b).

![Figure 3(a)](image3a.png)

Figure 3(a). Mass spectrum of isolated Compound I

![Figure 3(b)](image3b.png)

Figure 3(b). Mass spectrum of isolated Compound I

¹H NMR and ¹³C NMR spectrum of compound I was recorded on Bruker Avance II 400 NMR spectrometer in deuterium substituted water (D₂O) using TMS as internal standard (Chemical shifts in δ ppm).

The observed NMR bands for different functional groups in the Compound I in ¹H NMR and ¹³C NMR spectra are shown in Figure 4 and Figure 5, respectively. ¹H NMR bands for different functional groups in the isolated compound I were δ 4.70 (s, 1H, -OH group), δ 7.01- 6.99 (d, J 8.4, 2H, -Ar-H group), δ 6.64- 6.62 (d, J 8.8, 2H, -Ar-H group), δ 2.91- 2.71 (m, 4H, -N-CH₂ group), δ 2.5 (s, 6H, -CH₃ group). ¹³C NMR bands for different functional groups in the isolated compound I were 158.26 δ (s, -Ar-H group), 132.26 δ (s, -Ar-H group), 129.97 – 126.77 δ (m, -Ar-H group), 116.88 δ (s, -Ar-H group), 59.35 δ (s, -CH₂-N group), 42.99 δ (s, N-CH₃ group), 30.18 δ (s, -C-CH₂
Compound I showed positive Xanthoproteic test and Millon's test. Also, water soluble phenols were indicated by positive ferric chloride test. IR, $^1$H and $^{13}$C NMR, mass interpretation results confirmed that isolated Compound I was 4-(2-dimethylamino) ethyl phenol having molecular weight 165.23 and molecular formula C$_{10}$H$_{15}$NO.

3.3. Pharmacological evaluation of isolated compound

3.3.1. Antidiarrheal activity

Alcoholic extract successfully inhibited diarrhea in both models, i.e., the castor oil and magnesium sulphate induced diarrhea. The antidiarrheal effect was evident from the reduction of total number of feces in the test groups, which may be due to flavonoids, glycosides, tannins and alkaloids present in alcoholic extract [18]

3.3.1.1. Castor oil induced diarrhea in rats

In castor oil induced diarrhea model, the D. bipinnata extract in alcohol and isolated Compound I showed antidiarrheal effect in Wistar rats. Loperamide, being standard anti-diarrheal drug, was most effective in reducing number of feces by 70.94%, while among studied extracts alcoholic extract was found most effective which reduced the number of feces by 61.54%. Isolated Compound I was found effective as antidiarrheal agent and reduced the number of feces by 69.23%. All the tested extracts significantly (P<0.01) reduced the total number of feces when compared to control group by ANOVA followed to Dunnett’s ‘t’ test (Table 1).

Table 1. Effect of D. bipinnata underground parts alcoholic extract and isolated Compound I on castor oil induced diarrhea in rats (n=5)

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Dose (mg/kg, p.o.)</th>
<th>Mean no. of feces in 6 h</th>
<th>Feces Reduction (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Alcoholic extract</td>
<td>500</td>
<td>9.0±1.00**</td>
<td>61.54</td>
</tr>
<tr>
<td>Compound I</td>
<td>3</td>
<td>7.2±0.84**</td>
<td>69.23</td>
</tr>
<tr>
<td>Loperamide</td>
<td>3</td>
<td>6.8±0.84**</td>
<td>70.94</td>
</tr>
<tr>
<td>Control</td>
<td>10#</td>
<td>23.4±2.07</td>
<td></td>
</tr>
</tbody>
</table>

** Significant difference at P<0.01 vs. control and P<0.001 vs. control; one-way ANOVA followed by Dunnett’s ‘t’ test
*Significant difference at P<0.01 vs control; No significant difference at P<0.001 vs. control; one-way ANOVA followed by Dunnett’s ‘t’ test

Castor oil hydrolysis produces ricinoleic acid which induces diarrhea as hypersecretory response due to changes in the transport of water and electrolytes [19, 20]. Ricinoleic acid causes irritation and inflammation of gastric mucosa resulting in release of prostaglandins causing stimulation of secretion [21, 22]. Furthermore, ricinoleic acid also sensitizes intramural neurons of the gut. Several other mechanisms have also been reported to explain the diarrheal effect of castor oil which are adenylate cyclase activation or cAMP mediated active secretion [23] and inhibition of Na+, K+-ATPase activity [24].

3.3.1.2. Magnesium sulphate induced diarrhea model

In magnesium sulphate induced diarrhea model, the extract of D. bipinnata and isolated compound I showed antidiarrheal effect in Wistar rats (Table 2). Alcoholic extract of D. bipinnata showed 64.52% reduction in feces, which outperform slightly to the effect of standard antidiarrheal drug loperamide with 71.77% reduction in
feces. Isolated Compound I was effective in preventing diarrhea and reduced the number of feces by 70.16%. All extracts showed significant (P<0.01) antidiarrheal effect [18] in reducing the total number of feces, when compared to control using by ANOVA and followed to Dunnett ‘t’ test.

Table 2. Effect of D. bipinnata underground parts alcoholic extract and isolated Compound I on magnesium sulphate induced diarrhea in rats (n=5)

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Dose (mg/kg, p.o.)</th>
<th>Mean no. of feces in 6 h</th>
<th>Feces Reduction (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Alcohol</td>
<td>500</td>
<td>8.8±1.10**</td>
<td>64.52</td>
</tr>
<tr>
<td>Compound I</td>
<td>3</td>
<td>7.4 ±0.55**</td>
<td>70.16</td>
</tr>
<tr>
<td>Loperamide</td>
<td>3</td>
<td>7.0±0.71**</td>
<td>71.77</td>
</tr>
<tr>
<td>Control</td>
<td>10*</td>
<td>24±1.92</td>
<td></td>
</tr>
</tbody>
</table>

** Significant difference at P<0.01 vs. control and P<0.001 vs. control; one-way ANOVA followed by Dunnett’s ‘t’ test

*Significant difference at P<0.01 vs control; No significant difference at P<0.001 vs. control; one-way ANOVA followed by Dunnett’s ‘t’ test

# In ml/kg

Diarrhea in rats is also induced by administration of oral magnesium sulphate which increases the accumulation of fluid in the intestinal lumen and enhances flow from the proximal to distal intestine. This mechanism also involves release of NO, probably through stimulation of the constitutive form of NO synthase [25]. Magnesium sulphate has also been reported to liberate cholecystokinin from duodenal mucosa resulting in increase of small intestine secretions and motility and thus preventing the reabsorption of water and sodium chloride [26, 27].

Alkaloids, flavonoids, and steroids, in combination, exists in various plant extracts which possess anti-diarrhoeal activity, viz. root extract of Asparagus racemosus (Liliaceae), leaf extract of Clerodendrum phlomidis (Verbenaceae), stem bark extract of Cylicodiscus gabunensis (Mimosaceae), leaf extract of Emilia coccinea (Asteraceae) and fruit extract of Momordica cymbalaria (Cucurbitaceae) [28]. However, it was also observed that root extracts of Guiera senegalensis (Combretaceae) containing alkaloid, flavonoid and glycosides are effective in controlling diarrhea [28]. Stem bark extracts of Butea monosperma (Fabaceae) contains glycosides, flavonoids and steroids, which are effective in curing diarrhea [28]. It was also observed that plants containing alkaloids, flavonoids, glycosides, and steroids, viz. stem bark extract of Annona senegalensis (Annonaceae), root extract of Combretum sericeum (Combretaceae) and leaf extract of Dalbergia sissoo (Fabaceae), are also effective in diarrheal conditions [28].

Isolated Compound I, 4-(2-(dimethylamino) ethyl) phenol, is an alkaloid which showed antidiarrheal effect comparable to standard loperamide in both castor oil induced diarrhea and magnesium induced diarrhea model. Flavonoids and alkaloids have been reported to inhibit prostaglandins and autacoids release resulting in reduction of motility and secretion [28, 29]. Therefore, it is proposed that isolated Compound I acts by inhibition of prostaglandins and autacoids release resulting in reduction of motility and secretion.

3.3.2 Antiasthmatic activity

Standard drug chlorpheniramine showed 100% and alcoholic extract showed 97.72% contraction, at 0.8 ml dose (Figure 6 and Table 3). Preliminary phytochemical screening revealed the presence of alkaloids, flavonoids, steroids, glycosides, and coumarins in alcoholic extract of D. bipinnata [18]. Chlorpheniramine and alcoholic extract showed similar contractile responses. But, isolated compound I showed 97.67% contraction, at 0.4 ml dose (Figure 7), whereas chlorpheniramine showed 100% contraction, at 0.8 ml dose.
Compound I showed 100% protection against histamine induced lethality with no grasping (Table 4). Therefore, it may be concluded that isolated Compound I, isolated from alcoholic extract, possess anti-histaminic activity and thus isolated Compound I may be pharmacologically active and potentially useful in humans for prevention of asthma.

Table 3. Concentration response curve of D. bipinnata underground parts alcoholic extract and isolated Compound I in isolated guinea pig ileum.

<table>
<thead>
<tr>
<th>Histamine dose (ml)</th>
<th>Chlorpheniramine (%)</th>
<th>Alcoholic extract (%)</th>
<th>Isolated Compound I (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.05</td>
<td>31.7</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>0.1 ml</td>
<td>63.41</td>
<td>-</td>
<td>30.23</td>
</tr>
<tr>
<td>0.2 ml</td>
<td>75.6</td>
<td>-</td>
<td>55.81</td>
</tr>
<tr>
<td>0.3 ml</td>
<td>-</td>
<td>-</td>
<td>83.72</td>
</tr>
<tr>
<td>0.4 ml</td>
<td>95.12</td>
<td>27.27</td>
<td>97.67</td>
</tr>
<tr>
<td>0.6 ml</td>
<td>97.56</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>0.8 ml</td>
<td>100</td>
<td>97.72</td>
<td>-</td>
</tr>
</tbody>
</table>

Table 4. D. bipinnata underground parts alcoholic extract and isolated Compound I effect in histamine induced lethality in guinea pigs (n=5).

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Body weight (Avg. ± Std. dev.)</th>
<th>Dose (mg/kg body weight)</th>
<th>Recovery/Death</th>
<th>Lethality (%)</th>
<th>Grasping/Asthma</th>
</tr>
</thead>
<tbody>
<tr>
<td>Standard (Cetirizine)</td>
<td>314.86±47.07</td>
<td>0.5</td>
<td>Recovered</td>
<td>0</td>
<td>No grasping observed</td>
</tr>
<tr>
<td>Control</td>
<td>270.90±34.90</td>
<td>0.5</td>
<td>Death</td>
<td>100</td>
<td>Grasping observed</td>
</tr>
<tr>
<td>Alcoholic</td>
<td>496.3±10.87</td>
<td>500</td>
<td>Recovered</td>
<td>0</td>
<td>No grasping observed</td>
</tr>
<tr>
<td>Isolated Compound I</td>
<td>378.21±18.15</td>
<td>0.5</td>
<td>Recovered</td>
<td>0</td>
<td>No grasping observed</td>
</tr>
</tbody>
</table>

Histamine is an important chemical indicator involved in many allergic reactions. Histamine causes contraction of airway smooth muscle. Consequently, the muscles surrounding the airways constrict causing bronchospasm and shortness of breath, a life threatening condition [17]. Guinea pigs are highly sensitive to histamine induced bronchospasm. Present study deals with screening of antiasthmatic activity of D. bipinnata underground parts extract and isolated Compound I. Different agonists like acetyl choline, histamine, 5-hydroxytryptamine and bradykinin are responsible for contractile responses [30].

Bronchial asthma is characterized by increased airway reactivity to spasmogens. An initial event in asthma appears to be the release of inflammatory mediators (e.g. histamine, tryptase, leukotrienes and prostaglandins). Some of these mediators directly cause acute bronchoconstriction, airway hyper responsiveness and bronchial airway inflammation. Spasmolytic drugs like β adrenergic agonists, xanthine derivatives and anticholinergics relax the airway smooth muscles and are thus used as quick relief medications in acute asthmatic attacks. β adrenergic agonists promote bronchodilation by direct stimulation of β adrenergic receptors in the airway smooth muscle, that lead to relaxation of bronchial smooth muscle by rapid decrease in airway resistance in vivo [31, 32].

β adrenergic agonists activates adenylate cyclase resulting an increase in concentration of intracellular cyclic adenosine 3′, 5′-monophosphaste (cAMP) leading to activation of specific cAMP-dependent protein kinases that cause relaxation [33, 34]. Relaxation may also be due to inhibition of myosin phosphorylation. β adrenergic agonists reverse bronchoconstriction irrespective of the contractile agent. β adrenergic agonists prevent release of mediators from a number of inflammatory cells in vitro [33, 34].

Leaf extract of Adhatoda vasica (Acanthaceae), Fagopyrum esculaentum (Polygonaceae), seed oil of Hordeum vulgare (Poaceae), and whole plant extract of Passiflora incarnate (Passifloraceae), Passiflora foetida (Passifloraceae), Lepidium sativum, Solanum brevistigma (Solanaceae), Solanum xanthocarpum (Solanaceae), Solanum trilobatum (Solanaceae), Tylorhophora indica (Asclepiadaceae) are different plant extracts in which active constituents such as alkaloids have been proven for their antiasthmatic action and mechanism of action in these drugs is bronchodilation [8, 35, 36].

Isolated compound, an alkaloid, is pharmacologically active as antiasthmatic agent. The proposed mechanism for isolated Compound I appear similar to β agonist as it antagonizes histamine contractile responses of airway smooth muscle, resulting in relaxation in smooth muscles.

4 Conclusion

An isolated Compound I was isolated from alcoholic extract of D. bipinnata. The isolated Compound I was proteinaceus and aromatic in nature. The isolated compound I was confirmed by IR, 1H and 13C NMR, and mass as 4-(2-(dimethylamino) ethyl phenol with molecular weight 165.23 and molecular formula C10H15NO. Animal studies with isolated compound had proved the antiasthmatic and antidiarrheal activity of D. bipinnata.

Conflicts of Interest

Authors declare no conflicts of interest.

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