PHYTOCHEMICAL SCREENING AND EVALUATION OF IN VITRO HAEMOLYTIC, THROMBOLYTIC AND ANTIINFLAMMATORY ACTIVITIES OF AERVA LANATA (L.)

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ABSTRACT

The present study was aimed to screen the phytochemicals and to determine the in vitro hemolytic, thrombolytic and anti-inflammatory activities of aqueous extracts of leaf, flower and root of Aerva lanata. The in vitro haemolytic activity of root, flower and leaf of A. lanata was determined against human erythrocytes. Thrombolytic activity of root, flower and leaf of A. lanata was evaluated against human blood clot (thrombosis). The anti-inflammatory activity of root, flower and leaf of A. lanata was studied by membrane stabilization method. The phytochemical analysis of aqueous extracts of A. lanata showed that the presence of bioactive secondary metabolites. The aqueous extracts of root possess minimum haemolytic activity against RBC membrane. So, it can be considered as safe for the human erythrocytes. The maximum level of lyses of clot by root extract of A. lanata was observed, which may be due to the presence of tannins, alkaloids and saponins. The highest protection / stabilization of Human Red Blood Cell (HRBC) membrane by the flower extract of A. lanata were observed. The results of this study confirmed that the medicinal plant A. lanata possess bioactive compounds and in vitro haemolytic, thrombolytic and anti-inflammatory properties. So, we concluded that this study may be helpful in future for the identification of new pharmaceutical drugs from leaf, flower and root of Aerva lanata to treat inflammatory disease, myocardial infarction and other heart diseases.

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INTRODUCTION

Herbal medicines are the major remedy in traditional system of medicine which has been used in medical practices since antiquity. The practices continue today because of its biomedical benefits as well as place in cultural beliefs in many parts of world and have made a great contribution towards maintaining human health [1]. Plants are one of the major groups which support a new drug production. In the native system of medicine, plant plays an important role for the human health care in India and also suggested to increase the natural resistance of the body to diseases. Plants are used to treat various diseases from ancient times. Medicinal plants are known for their extensive production of bioactive compounds [2].

Haemolysis is the breakage of the red blood cell (RBC’s) membrane causing the release of haemoglobin and other internal components into the surrounding fluid. In vivo haemolysis may be due to pathological conditions, such as autoimmune haemolytic anaemia or blood transfusion reaction. In vitro haemolysis may be due to improper specimen collection, specimen processing, or specimen transport [3]. In vitro haemolytic activities are becoming a new area of research in drug discovery. Researchers are exploring ethnobotanically important plants to find out potential natural products with antiaggregant action. These studies are important because some patients have become resistant to the already existing drug e.g. aspirin [4] and/or conventional medication in association with medicinal plant formulations. Plants constitute a major component of diet and traditional medicine. However, many plants are reported to have serious adverse effects, which include the induction of haemolytic anaemia [5]. Patients and health care providers need to be provided with lists of commonly used plants that could worsen the haemolytic conditions of patients with haemolytic disorders. Therefore, many of the commonly used plants need to be evaluated for their potential haemolytic activity [6]. Various models exist for evaluation of membrane toxicity of surfactants including single cell models using erythrocytes, erythrocyte ghosts and liposomes. The erythrocyte model has been widely used technique and direct indication of toxicity of injectable formulations as well as general indicator of membrane toxicity. Another advantage of erythrocyte model is readily available and the red blood cells are easy to isolate from the blood; moreover, its membrane has similarities with other cells [7].

A blood clot (thrombus) developed in the circulatory system due to failure of haemostasis causes vascular blockage and while recovering leads to serious consequences in atherothrombotic diseases such as myocardial or cerebral infarction, at times leading to death [8]. In India, though Streptokinase (SK) and Urokinase (UK) are widely used due to lowering cost [9] as compared to other thrombolytic drugs. Thrombolytic agents are associated with hyper risk of haemorrhage [10], severe anaphylactic reaction and lacks specificity. As a result of immunogenicity multiple treatments with SK are restricted [11], because of the shortcomings of the available thrombolytic drugs. The attempts are underway to develop improved recombinant variants of these drugs in this modernised world. Nowadays, we are facing major threats against many stress related disease. Thrombosis is a pathological condition, which plays a vital role to causes many diseases like cardiovascular diseases and stroke.

Inflammation is the reaction of living tissues to redness, swelling, injury, infection or irritation. Lysosomal enzymes released during inflammation produce a variety of disorders which leads to the tissue injury by damaging the macromolecules and lipid peroxidation in membranes. They are assumed to be responsible for certain pathological conditions like heart attacks, septic shocks, rheumatoid arthritis etc. The extra cellular activity of these enzymes is said to be related to acute or chronic inflammation. Stabilization of lysosomal membrane from the inflammatory response inhibits the release of lysosomal constituents of activated neutrophil such as bactericidal enzymes and proteases, which cause further tissue inflammation and damage upon extra cellular release [12]. In order to overcome this problem, mainly the Non Steroidal Anti-inflammatory Drugs (NSAIDs) are prescribed. The activity of NSAIDs in rheumatoid arthritis and other inflammatory diseases does not seem to be only due to the inhibition of the production of endogenous prostaglandins, but also by preventing the denaturation of proteins which could be affected at much lower doses than those required in these conditions [13]. These anti-inflammatory agents in spite of their potency in relieving pain and other consequences of inflammatory responses are also associated with some serious side effects, especially in elderly. Prolong usage of NSAIDs may cause gastric bleeding, ulceration, bone marrow disturbance, kidney and liver dysfunction [14]. However plant derived drugs is used to treat most of the diseases wherever difficult to treat with allopathic medicines. Even today 80% of the world population depends on plant derived medicines for the first line of primary health care because of least / no side effects [15, 16].

*Aerva lanata* Linn. (Family: Amaranthaceae) is a woody, prostrate or succulent, perennial herb. It is a common weed which grows wild everywhere in plains of India and Bangladesh. The whole plant, especially the leaves, is edible. The plant is claimed to be useful as diuretics, anthelmintic, antidiabetic, and hepatoprotective drug in traditional system of Indian medicine. Previous studies have also shown that it possess antibacterial [17], antimicrobial and cytotoxicity [18], diuretics [19], antidiabetic [20], hepatoprotective [21, 22] and anti inflammatory [23] activities. It is being commonly prescribed by Ayurvedic doctors, alone or in combination for the treatment of urinary infections. It also possesses analgesic, anthelmintic, anti-inflammatory, anti-malarial, anti-venin, diuretic and sedative properties [24, 25]. It has been reported that Canthin-6-one and β-Carboline alkaloids were isolated from the leaves of *A. lanata* [26]. *In vitro* anti-platelet and haemolytic effects of methanolic extract of *A. lanata* were reported [27]. *In vitro* anti-inflammatory activity of petroleum ether, chloroform, methanol and aqueous extracts of *A. lanata* was also reported [28]. So, there are many pharmacological studies are available using *A. lanata*. But there is no detailed study on phytochemical profile and *in vitro* haemolytic, thrombolytic and anti-inflammatory activities of aqueous extracts of leaf, flower and root of *A. lanata* (L.). So, the present study was aimed to screen the phytochemicals and to determine the *in vitro* haemolytic, thrombolytic and anti-inflammatory activities of aqueous extracts of leaf, flower and root of *A. lanata*.
MATERIALS AND METHODS

Collection of Plant Material
The medicinal plant *A. lanata* was collected from in and around Mayiladuthurai at Nagapattinam District, Tamilnadu, India. The plant was identified by Rev. Dr. S. John Britto, Director, Rapinat Herbarium and Centre for Molecular Systematics, Department of Botany, St. Joseph’s College, Tiruchirappalli, Tamilnadu, India.

Preparation of Crude Extracts
The leaf, flower and root of *A. lanata* were separated from the collected plants and washed thoroughly in tap water followed by distilled water. The plant materials were shade dried at room temperature and ground well using mechanical grinder. 30 g powder of leaf, flower and root were soaked separately in distilled water for 12 to 16 hours and boiled, and then it was filtered through muslin cloth and Whatmann no. 1 filter paper. The aqueous extracts were concentrated and made the final volume to one-fifth of the original volume [29]. The paste form of extract was obtained and used for further phytochemical and *in vitro* studies.

Phytochemical Analysis
Qualitative phytochemical analysis was carried out for the determination of alkaloids, carbohydrates, glycosides, proteins, phytosterols, tannins, phenols, flavanoids, coumansins, saponins, quinones, cardiac glycosides, terpenoids, pholabatannins and anthraquinones in aqueous extracts of leaf, flower and root of *A. lanata* by standard procedures [30, 31, 32].

*In vitro* Haemolytic Assay
Haemolytic assay was carried out by the method of Bulmus *et al.* [33]. Freshly collected human red blood cells were taken and allowed to clot for 30 minutes. The serum was removed by centrifugation at 2500 rpm for 15 minutes, and then the pellet was washed thrice by 150 mM NaCl (2500 rpm for 10 minutes). 0.5% RBC cells were suspended in 100 mM sodium phosphate buffer. The different concentrations (200μg, 400μg, 600μg, 800μg and 1000μg) of extracts of leaf, flower and root of *A. lanata* were mixed with 200μl of RBC and then the final reaction mixture volume was made up to 2 ml by adding sodium phosphate buffer. The reaction mixture was placed in a water bath for 1 hour at 37°C. After incubation, the reaction mixture was centrifuged again at 1500 rpm for 10 minutes. The supernatant was collected and the optical density was measured at 541 nm. The sodium phosphate buffer was used as blank and aspirin was used as positive control. The experiment was done in triplicates. The result was calculated by the following formula. The results were represented as mean ± SE.

\[
\text{Percentage haemolysis} = \frac{(\text{Absorbance of sample} - \text{Absorbance of blank})}{\text{Absorbance of positive control}} \times 100
\]

Thrombolytic Activity
Thrombolytic assay was carried out by the method of Prasad *et al.* [34]. Venous blood samples were drawn from three healthy human volunteers (3 ml each). 200μl of blood was transferred to previously weighed eppendorff tubes for each subject. In the first series, the transferred blood was allowed to clot at 37 ºC for 45 minutes. After clot formation, serum was completely removed and each tube having clot was again weighed to determine the clot weight (clot weight = weight of clot containing tube - weight of the tube alone). To each eppendorff tube containing pre-weighed clot added 1 ml of different concentrations (200μg, 400μg, 600μg, 800μg and 1000μg) of plant extracts or 1000μL of distilled water as a negative control or 1000 μl of streptokinase (30, 000 IU) as a positive control. All the tubes were then incubated at 37 ºC for 90 min and observed the clot lysis. After incubation, the fluid released and it was removed and then the tubes were again weighed to observe the difference in weight after clot stabilization. The obtained difference in weight was expressed as percentage of stabled or lysed clot, which was calculated by the following formula.

\[
\% \text{ of clot lysis} = \frac{(\text{W}2 - \text{W}3)}{(\text{W}2 - \text{W}1)} \times 100
\]

W1=Empty weight of eppendorff tube
W2=Weight of eppendorff tube + clot
W3=Weight of clot release after addition of plant extract
In vitro Anti-Inflammatory Activity

In vitro anti-inflammatory activity was determined by the human red blood cell (HRBC) membrane stabilization method [35]. The blood was collected from healthy human volunteer who had not taken any NSAIDS for 2 weeks prior to the experiment and mixed with equal volume of Alsever solution (2% dextrose, 0.8% sodium citrate, 0.5% citric acid and 0.42% NaCl) and centrifuged at 3,000 rpm. The packed cells were washed with saline (0.85% NaCl) and a 10% suspension was made. Various concentrations of extracts were prepared (200μg, 400μg, 600μg, 800μg and 1000μg/ml) using distilled water. To 1 ml of different concentrations of extracts, added 1 ml of phosphate buffer, 2 ml of hypo saline (0.35% NaCl) and 0.5 ml of HRBC suspension. All the tubes were incubated at room temperature (37 ºC) for 30 min and centrifuged at 3,000 rpm for 20 min and then haemoglobin contents of the supernatant solution was estimated by UV spectrophotometer at 560 nm. Diclofenac (1000µg/ml) was used as reference standard and a control was prepared by omitting the extracts.

\[
\text{% protection} = \left( {\frac{{100 \times \text{Optical density of drug treated sample}}}{{\text{Optical density of control}}}} \right) \times 100
\]

RESULTS

The medicinal value of the plant can be correlated due to the presence of various bioactive chemical constituents. The phytocompounds such as alkaloids, carbohydrates, proteins, steroids, tannins, phenols, flavonoids, saponins, quinones, cardiac glycosides and terpenoids were found in the aqueous extracts of leaf, flower and root of A. lanata and the results are given in Table 1. Whereas phlobatannins and anthraquinones were absent in all three parts of A. lanata (L.). The higher level of bioactive compounds was observed in root when compared to flower and leaf extracts of A. lanata. Alkaloids, carbohydrates and terpenoids are observed higher concentration in the leaf than flower and root extracts of A. lanata.

Table 1. Phytochemical screening of leaf, flower and root of A. lanata.

<table>
<thead>
<tr>
<th>Name of the phytocompounds</th>
<th>Aqueous extract</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Leaf</td>
</tr>
<tr>
<td>Alkaloids</td>
<td>+++</td>
</tr>
<tr>
<td>Carbohydrates</td>
<td>+++</td>
</tr>
<tr>
<td>Glycosides</td>
<td>-</td>
</tr>
<tr>
<td>Proteins</td>
<td>+</td>
</tr>
<tr>
<td>Phytosteroids</td>
<td>+</td>
</tr>
<tr>
<td>Tannins</td>
<td>-</td>
</tr>
<tr>
<td>Phenols</td>
<td>+</td>
</tr>
<tr>
<td>Flavonoids</td>
<td>+</td>
</tr>
<tr>
<td>Coumarins</td>
<td>+</td>
</tr>
<tr>
<td>Saponins</td>
<td>+</td>
</tr>
<tr>
<td>Quinones</td>
<td>++</td>
</tr>
<tr>
<td>Cardiac glycosides</td>
<td>++</td>
</tr>
<tr>
<td>Terpenoids</td>
<td>++</td>
</tr>
<tr>
<td>Phlobatannins</td>
<td>-</td>
</tr>
<tr>
<td>Anthraquinone</td>
<td>-</td>
</tr>
</tbody>
</table>

+ Low amount; ++ moderate amount; +++ high amount; + present; - Absent

In vitro Haemolytic Activity

Haemolytic activity could be used as a primary tool for studying the toxicity of the drugs and it provides primary information of the interaction between molecules and biological entities at cellular level. The in vitro haemolysis test was used as a method for substance toxicity screening and estimating any in vivo damage on animal model. In vitro haemolytic activity of aqueous extracts of leaf, flower and root of A. lanata at different concentrations (200μg, 400μg, 600μg, 800 μg and 1000μg/ml) was performed on human erythrocytes. The aqueous extracts of leaf, flower and root of A. lanata exhibited low to high haemolytic effect on human erythrocytes. Haemolytic activity of the plant extract was expressed as % haemolysis and the results were reported in Table 2. Maximum lysis was observed in 1000 µg/ml (40.7±0.38%) of aqueous extract of flower of A. lanata. The minimum red blood cell lysis was observed in root than leaf and flower extracts of A. lanata.
Table 2. *In vitro* haemolytic activity of leaf, flower and root of *A. lanata*.

<table>
<thead>
<tr>
<th>Name of the part of plant</th>
<th>% of haemolysis</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Concentration of plant extracts (µg/ml)</td>
</tr>
<tr>
<td></td>
<td>200</td>
</tr>
<tr>
<td>Leaf</td>
<td>4.65±0.75</td>
</tr>
<tr>
<td>Flower</td>
<td>15.65±0.25</td>
</tr>
<tr>
<td>Root</td>
<td>5.83±1.60</td>
</tr>
<tr>
<td>Negative control (buffer)</td>
<td>-</td>
</tr>
<tr>
<td>Positive control (Aspirin 1mg/ml)</td>
<td>-</td>
</tr>
</tbody>
</table>

Values are expressed as mean ± SE of triplicates

In *Vitro* Thrombolytic Activity of *A. lanata*

As a part of discovery of cardioprotective drugs from natural sources, the aqueous extracts of leaf, flower and root of *A. lanata* was assessed their thrombolytic activity and the results were represented in Table 3. Addition of 1000µl of streptokinase as positive control (30,000 IU/ml) to the clots and subsequent incubation for 90 minutes at 37 ºC showed 74.22±1.22% lysis of clot. At the same time distilled water was treated as negative control which exhibited negligible percentage of lysis of clot (3.75±0.51%). In this study, the aqueous extract of root exhibited highest thrombolytic activity in different concentrations. Flower extract showed significant clot lysis activity against dissolute of thrombosis. *A. lanata* leaf extract has moderate thrombolytic activity when compared with streptokinase (positive control).

Table 3. *In vitro* thrombolytic activity of leaf, flower and root of *A. lanata*.

<table>
<thead>
<tr>
<th>Name of the part of plant</th>
<th>% of thrombolysis</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Concentration of plant extracts (µg/ml)</td>
</tr>
<tr>
<td></td>
<td>200</td>
</tr>
<tr>
<td>Leaf</td>
<td>7.48±2.14</td>
</tr>
<tr>
<td>Flower</td>
<td>5.14±1.41</td>
</tr>
<tr>
<td>Root</td>
<td>29.08±2.97</td>
</tr>
<tr>
<td>Negative control (water)</td>
<td>-</td>
</tr>
<tr>
<td>Positive control Streptokinase (30,000IU/ml)</td>
<td>-</td>
</tr>
</tbody>
</table>

Values are expressed as mean ± SE of triplicates

In *Vitro* Anti-inflammatory Activity

The investigation is based on the need for newer anti-inflammatory agents from natural source with potent activity and lesser side effects as substitutes for chemical therapeutics. *In vitro* anti-inflammatory activity was assessed by HRBC membrane stabilization. Aqueous extract of flower of *A. lanata* showed the highest membrane protection (anti-inflammatory activity) in all concentrations, but the maximum protection was observed at the concentration of 800µg/ml (80.14±0.52%) and 1000µg/ml (87.9±0.28%) of plant extracts. Whereas root extract possess higher inhibition of RBC membrane lysis, when compared to leaf extract of *A. lanata*. Diclofenac as a standard drug showed the maximum protection (82.74±0.91%) at the concentration of 1000 µg/ml. *A. lanata* has significant anti-inflammatory activity, which may be due to the presence of chemical profile such as flavonoids, triterpenoids and phenolic compounds.

Table 4. *In vitro* anti-inflammatory activity of leaf, flower and root of *A. lanata*.

<table>
<thead>
<tr>
<th>Name of the part of plant</th>
<th>% of membrane protection</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Concentration of plant extracts (µg/ml)</td>
</tr>
<tr>
<td></td>
<td>200</td>
</tr>
<tr>
<td>Leaf</td>
<td>38.36±0.55</td>
</tr>
<tr>
<td>Flower</td>
<td>60.28±0.63</td>
</tr>
<tr>
<td>Root</td>
<td>51.55±0.76</td>
</tr>
<tr>
<td>Diclofenac sodium (1mg/ml)</td>
<td>-</td>
</tr>
</tbody>
</table>

Values are expressed as mean ± SE of triplicates
DISCUSSION

Since ancient times, herbal preparations have been used for the treatment of several diseases. Herbal products are often perceived as safe because they are “natural” [36]. As previously reported study shows that the phytochemicals are divided into number of groups such as alkaloids, volatile essential oils, phenols and phenolics, flavonoids, glycosides, resins, steroids, tannins and terpenes. The secondary metabolites are important plant source, safer and valuable substitute for the synthetic drugs created antimicrobial agents. [37]. The previous report showed that the presence of alkaloids, tannins, saponins, flavonoids, carbohydrates, glycosides, phenols, steroids, phlobatannins, cardiac glycosides, proteins and resins in the methanol extract of A. lanata [38]. Yamunadevi [39] was also reported that the presence of various phytocompounds in the methanol extract of A. lanata. Rajendra Prasad et al. [40] reported that the phytochemical analysis of hexane, chloroform, acetone, ethyl acetate and methanol extracts of root of Aerva lanata showed that the presence of secondary metabolites. Vjiyalakshmi et al. [41] was also reported the presence phytoconstituents in the aqueous extract of root of A. lanata. The phenolic components are of importance and interest in pharmacy due to their relationship with anticancer activity [42].

The available reports are showed that the aqueous extracts of leaves of A. lanata, Calotropis gigantean and Elaeocarpus ganitrus alone and in combination with each other, is non/less toxic to the human erythrocytes [43]. The researcher reported that haemolytic activity of aqueous extract of stem of A. lanata was screened against normal human erythrocytes and the percentage of activity is increased with increase the concentration of extracts [44]. Ralph et al. [45] reported the haemolytic activity rated to the degree of in vitro toxicity according to the observed mortality rates such as 0 to 9% - non-toxic, 10 to 49% - slightly toxic, 50 to 89% - toxic and 90 to 100% - highly toxic. So, the haemolytic activity is an important measurement and the indicator for cell toxicity. The in vitro haemolysis test has been employed by many different groups for the toxicological evaluation of different plants [7]. Mechanical stability of erythrocytes membrane is a good indicator of in vitro cytotoxicity [46]. This assay is an important technique to determine toxicity of drug and if a drug possesses antioxidant and other bioactivities and it can be used in pharmacological applications [47]. The erythrocyte membranes are susceptible to peroxidation, because they are rich in polyunsaturated fatty acids. RBC membrane contains haemoglobin, which may catalyze the oxidation process and they are continuously exposed to high concentration of oxygen. The oxidation of erythrocytes serves as good models for the oxidative damage of biological membranes [48]. It has been found that certain chemicals are having ability to generate radicals, which can attack the erythrocyte membrane and lead to haemolysis [49].

Epidemiologic studies have provided evidence that plant food materials possess antithrombotic activity which can reduce risk of thrombosis. A number studies have been carried out by researchers to determine the herbs and natural food sources and their supplements having antithrombotic (anticoagulant and antiplatelet) effect and it provides evidence for consuming of such food leads to prevention of coronary events and stroke [50].

Plasmin is a natural fibrinolytic agent lyse clot by breaking down the fibrinogen and fibrin in the clot. Streptokinase reacts with plasminogen form a complex that can convert additional plasminogen to plasmin [51]. Moreover, phlorotannin, isolated from marine brown algae, have a unique property to lyse blood clot in intravascular blood vessel via antiplasmin inhibition [34]. Considerable efforts have been directed towards the discovery and development of natural products from various plant and animal sources which have antiplatelet [36, 52], anticoagulant [53, 54], antithrombotic and thrombolytic activities [55]. Medicinal plants showed thrombolytic activity was studied and some significant observations have been reported [56]. Advances of phytochemistry and identification of plant compounds, which are effective in curing certain diseases has renewed the herbal medicines.

The inflammation is a body response to injury or infection or destruction characterized by heat, redness, pain, swelling and disturbed physiological functions. It is triggered by the release of chemical mediators from injured tissue and migrating cells [57]. The in vitro and in vivo anti-inflammatory studies were performed based on folklore information [58]. The lysosomal enzymes released during inflammatory conditions and produce a variety of disorders. The extracellular activity of these enzymes is said to be related to acute or chronic inflammation. The non steroidal drugs act either by inhibiting these lysosomal enzymes or by stabilizing the lysosomal membrane [59].

The erythrocyte membrane is similar to the lysosomal membrane and its stabilization implies that the extract may be well stabilizing lysosomal membranes. Mechanism of stabilization of lysosomal membrane is an important in the inflammatory response by preventing the release of lysosomal constituents of activated neutrophil such as bactericidal enzymes and proteases, which cause tissue inflammation and damage extracellular release [60]. The plant extracts possess maximum stabilization of RBC membrane. These results provide evidence for membrane stabilization as an additional mechanism of anti-inflammatory activity. The phenolics and flavonoids of medicinal herbs are act as anti-inflammatory agents [61]. The anti-inflammatory activity of A. lanata may be due to inhibiting either release of lysosomal enzymes or by stabilizing the lysosomal membrane, which is one of the major events responsible for the inflammatory process [62]. The anti-inflammatory activity of the flower and root extracts of plant may be due to their content of flavonoids, tannins and anthocyanins which inhibits the cyclooxygenase activity [63].
CONCLUSION

The phytochemical investigation of aqueous extracts of leaf, flower and root of A. lanata (L.) showed that the presence of secondary metabolites. The maximum concentration of phytochemicals was found in aqueous extract of root of A. lanata. In vitro haemolytic, thrombolytic and anti-inflammatory activities of aqueous extract of leaf, flower and root of A. lanata were screened. The results revealed that the root extract possess less haemolytic activity against human erythrocytes and it can be considered as safe for the human erythrocytes. All parts of plant extract showed significant thrombolytic activity against human blood clot (thrombosis), which may be due to the presence of tannins, alkaloids and saponins. The maximum lysis of clot was observed in root extract of A. lanata. In future, it may be incorporated as a thrombolytic agent for the improvement of the patients suffering from Atherothrombotic disease. The anti-inflammatory activity of leaf, flower and root of A. lanata was observed using human red blood cells and it may be due to inhibition of the mediators of inflammation such as histamine, serotonin and prostaglandins. In this study no toxicity towards erythrocyte membrane was observed. So, we recommend for further research to find a novel and effective anti-inflammatory drug from A. lanata to treat skin diseases.

ABBREVIATIONS
SK – Streptokinase
UK - Urokinase
HRBC - Human red blood cell

COMPETING INTERESTS
The authors declared that there is no conflict of interests exists.

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