



Anti-inflammatory Activity of Butanolic Extract of *Albizia lebeck*

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Author's contribution

The sole author designed, analyzed and interprets and prepared the manuscript.

Short Research Article

Received 1st June 2014
Accepted 17th July 2014
Published 30th July 2014

ABSTRACT

Albizia lebeck is a tree widely distributed in India and is also found in South Africa, South America and Australia. As in Indian traditional system as folk medicine, this plant is used to treat several inflammatory pathologies such as asthma, arthritis and burns. Study of other species this same genus has demonstrated an anti-inflammatory activity of crude extract, which, in some work has been attributed to the presence of saponins. In order to confirm these findings a study of phytochemical profile was realized and a rich extract in saponins, butanolic extract, was obtained and its anti-inflammatory activity was evaluated through measured by inhibition of carrageenan-induced mouse paw oedema, using dexamethasone as reference compound. The extract exhibited a moderate control of the both phase of inflammation, provoking an inhibition of edema formation. However, the butanolic extract exhibited lesser activity than reference compound dexamethasone. The results obtained suggest a significant antiinflammatory property of the butanolic extract of *Albizia lebeck*, justifying the use of this plant in the traditional medicine for the treatment of inflammatory conditions and confirm their saponins as bioactive product.

Keywords: *Albizia lebeck*; anti-inflammatory activity; saponins; butanolic extract.

1. INTRODUCTION

Inflammatory conditions are a major cause of morbidity world-wide. Non-steroidal anti-inflammatory drugs and steroids are the most common drugs used to treat inflammation.

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Gastrointestinal side effect is a major side effect associated with the currently available non-steroidal anti-inflammatory drugs which limit their application. This may be contributing to the current move by large proportion of world population towards herbal remedies for the treatment of inflammatory diseases.

Herbal medicine is still the mainstay of about 80% of the whole population, mainly in developing countries, for primary health care because of better cultural acceptability, better compatibility with the human body and fewer side effects. However, the last few years have seen a major increase in their use in the developed world [1]. A number of medicinal plants are used in developing countries for the management of a number of disease conditions including pain and inflammatory conditions. The confirmation of the folkloric claims of these medicinal plants will provide scientific basis for the conservation of tropical medicinal resources, the deployment of the beneficial ones as phytomedicine in the primary health care and the development of potential bioactive constituents as novel lead compounds or precursors in drug design [2].

Saponins are a group of secondary metabolites, nonvolatile surfactants are widely distributed in the plant kingdom and marine animal [3]. They are composed of triterpenoid or steroid aglycone moiety and complex oligosaccharide substituent. This group of natural products have the common characteristic of foaming when shaken with water [4]. The formed foam is a result of the formation of a solution colloidal, being stable to the action of mineral acids diluted and durable, differing from those of ordinary soap. Other important feature of this class of substances is related to the ability of precipitating cholesterol by formation of insoluble complexes. Both of the above characteristics are correlated with amphipathic or amphiphilic nature of these molecules, since they are formed by a portion hydrophilic and lipophilic [5]. These bioactive products have a range of properties due to their extensive diversity structure, which includes features and bitter sweeteners, detergent properties and emulsifiers. In addition of biological, pharmacological and medical properties, such as hemolytic activity, antimicrobial, insecticidal and molluscicidal [3]. Also noteworthy are the jobs in pharmaceutical industries as raw material for synthesis of steroidal drugs such as contraceptives, in addition to heavy use in industry cosmetics [6]. The mounting demand for natural products coupled with their physicochemical properties and numerous biological activities has led to the emergence of saponins as commercially significant compounds with expanding applications in food, cosmetics and pharmaceutical industries [7]. Steroidal saponins are important raw materials for the production of steroidal hormones and drugs [8]. Saponins are used as immunological adjuvants in the formulation of vaccines due to their immune enhancing properties [9]. Information on the biological activities of saponins from variety of sources provide lead for the development and design of new drugs. An example is the chemotherapeutic activity of the ginseng saponins prompted the development of anticancer drugs [10].

Albizia lebbek is a tree widely distributed in tropical areas of some countries, such as India, South Africa and Australia. In traditional medicine it is used as an alternative drug therapy in chronic inflammatory diseases such as arthritis and bronchitis [11]. According to the literature, a significant anti-inflammatory activity was attributed to a mixture of secondary metabolites in its crude extract through of acute and chronic tests models experimentals [12]. Other studies with the same family of *Albizia lebbek* showed antiinflammatory activity correlated with triterpenoidal aglycones, ursolic and equinocistic acid, present in their extracts (Fig. 1). Thus, in order to confirm these results and in attempt to establish a structure-activity relationship an evaluation of the phytochemical profile of the butanolic

extract of this plant was performed and its anti-inflammatory activity against inhibition of carrageenan-induced mouse paw oedema was observed.

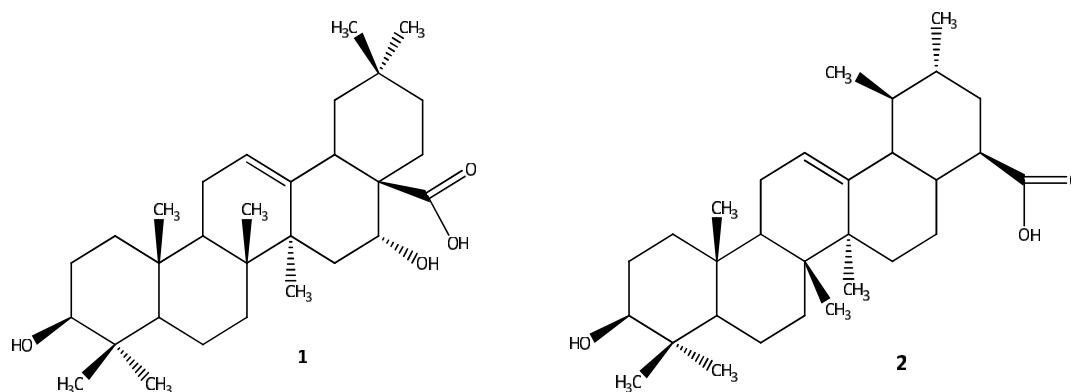


Fig. 1. Structure of equinocistic (1) and do ursolic acid (2)

Martínez et al. [13] identified in extracts of *Albizia lebbek* a variety of secondary metabolites as catechins, kaempferol, quercetin, lupeol, α -amyrine, triterpenoids and, mainly, some saponins (Albiziasaponins A (3), B (4), and C (5)) (Fig. 2).

2. MATERIALS AND METHODS

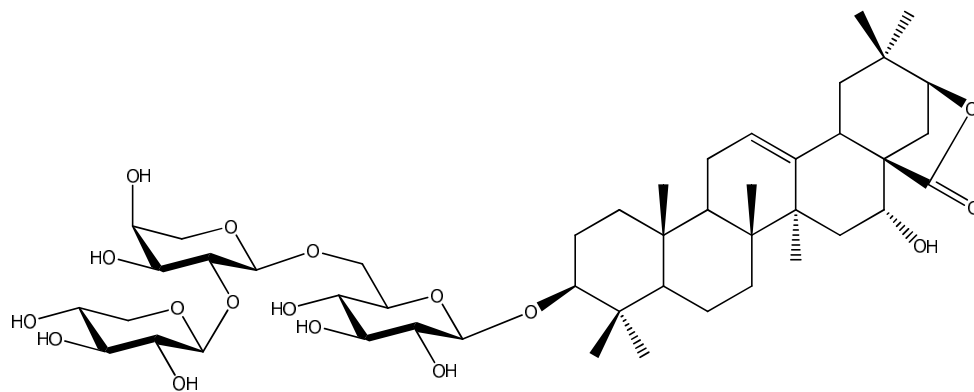
2.1 Plant Materials

Albizia lebbek leaves (100g) were extracted with ethanol for 72 hours. The extract was concentrated under reduced pressure to give a crude extract. Subsequently, this extract was partitioned with n-butanol to remove polar substances and obtain an extract rich in saponins (50mg).

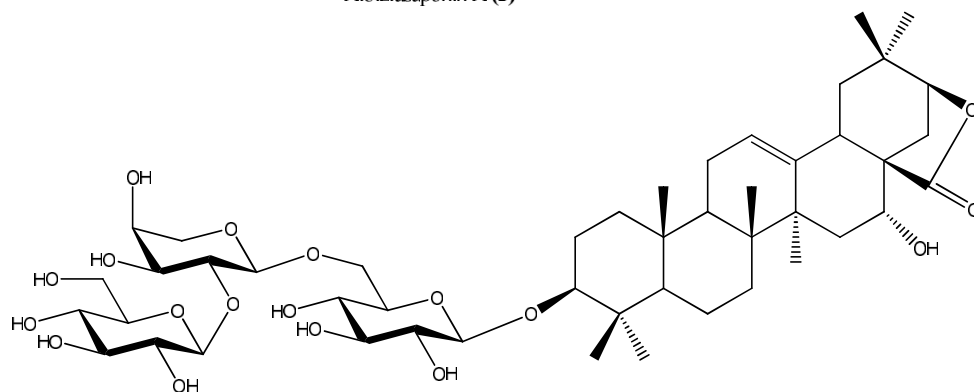
2.2 Antiinflammatory Activity

The anti-inflammatory activity was assessed by measuring inhibition of rat paw edema induced by carrageenan, using dexamethasone as reference compound. Fifteen Swiss, male mice were divided into three groups. The first group was given 1ml of saline solution negative control, the second group was administered the butanolic extract (100mg/kg), and the third group was administered dexamethasone (25mg/kg), a positive control. One hour after oral administration, acute inflammation was induced by injection of 50 μ l of 1% freshly prepared colloidal suspension of carrageenan in physiological saline into the subplantar region of the right hind paw of mice [14]. The paw diameter was measured with the aid of a spring-loaded dial gauge (Mitutoyo Corp., Tokyo, Japan) at 0, 1, 2, 3, and 4 hours after injection of the carrageenan. Percentage inhibition of oedema was calculated using the following formula:

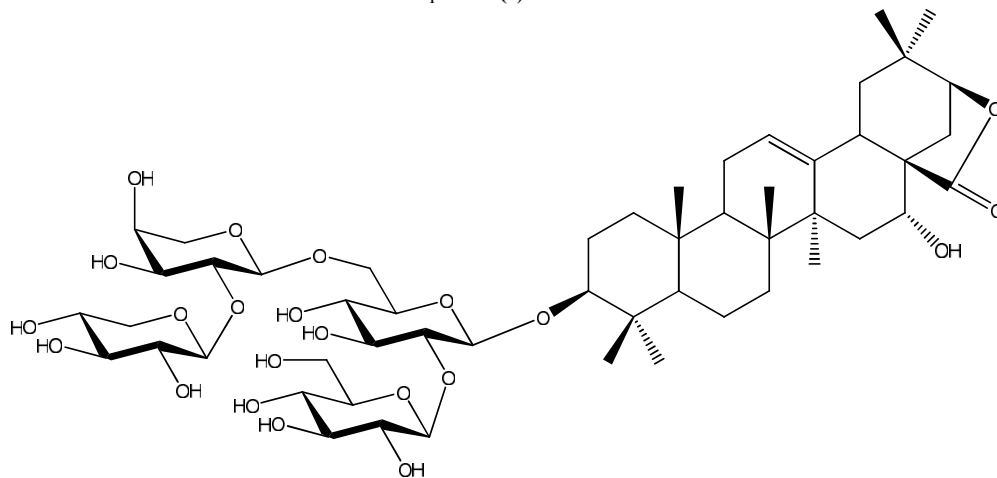
$$\% \text{ inhibition of oedema} = \frac{\text{Mean oedema increase (Ct)} - \text{Mean oedema increase (Tt)}}{\text{Mean oedema increase (Ct)}} \times 100$$



Albiziasaponin A (3)



Albiziasaponin B (4)



Albiziasaponin C (5)

Fig. 2. Structure of albiziasaponins A (3), B (4) and C (5)

Where:

Ct = Control group at time 1, 2, 3 and 4 hours

Tt = Treated groups at time 1, 2, 3 and 4 hours

2.3 Statistical Analyses

The data obtained from the carrageenan-induced paw oedema test were expressed as mean \pm SEM and analyzed using one way ANOVA followed by Student's t-test. P values less than 0.05 and 0.01 ($P < 0.05$ and $P < 0.01$) were considered to be statistically significant.

3. RESULTS AND DISCUSSION

The butanolic extract rich in saponins, produced statistically significant ($P < 0.05$ and $P < 0.01$) decreases in the carrageenan-induced rat paw oedema at 3 hrs which was the point of maximum oedema production in this study as shown in (Fig. 3).

The butanolic extract showed significant anti-inflammatory activity in the carrageenan-induced rat paw oedema test and the presence of saponins in this butanolic extract was confirmed by TLC employing assays using orcinol-H₂SO₄ as reagent cartridge. The development of the carrageenan-induced oedema has been shown to be bi-phasic in nature. The first phase of the development has been attributed to the release of histamine, serotonin and kinins, which promote vasodilation and increased permeability. The second phase has been considered to be due to the release of chemical mediators occur with prolonged action, which are released during the inflammatory process until the agent is totally eliminated from the tissue. In this case, the main mediators are bradykinin and prostaglandins, which promote vasodilation and increased permeability, and promote chemotaxis, which is the attraction of leukocytes to the affected area.

The result showed that butanolic extract of *Albizia lebbek*, (100mg/kg), exhibited significant inhibitory activity against the carrageenan-induced oedema at all the phases of inflammation which was observed to be comparable to that produced by dexamethasone (25mg/kg). Generally the results obtained in this work suggest that the butanolic extract tested have significant anti-inflammatory activity that might be mediated through the inhibition of the release and synthesis of the agents that produce inflammations.

Evidence for the anti-inflammatory properties of saponins has been provided by several studies using different models of inflammation [15-19]. Saponins isolated from the leaves and root extract of *Camellia sinensis* have been shown to inhibit carrageenan-induced rat paw oedema in a dose dependant manner [20]. Other mode of actions have been used to explain the anti-inflammatory activity of saponins; an example is the anti-inflammatory activity of platycodin D isolated from the roots of *Platycodon grandiflorum* shown to be due to the inhibition of 12-O-Tetradecanoyl-phorbol-13-acetate (TPA)-induced PGE2 production through the inhibition of COX-2, secondly the hederagenin glycoside from *Kalopanax pictus* was reported to inhibit lipopolysaccharide (LPS)-induced iNOS and COX-2 protein expression which led to the inhibition of nitric oxide (NO), PGE2 and tumor necrosis factor- α (TNF- α) [21]. It has been suggested that the numerous biological activities of saponins were linked to their amphiphilic nature which help in the accomplishment of these activities through their ability to intercalate into the plasma membrane resulting in changes in membrane fluidity that in turn affect membrane function thus eliciting a cellular response.

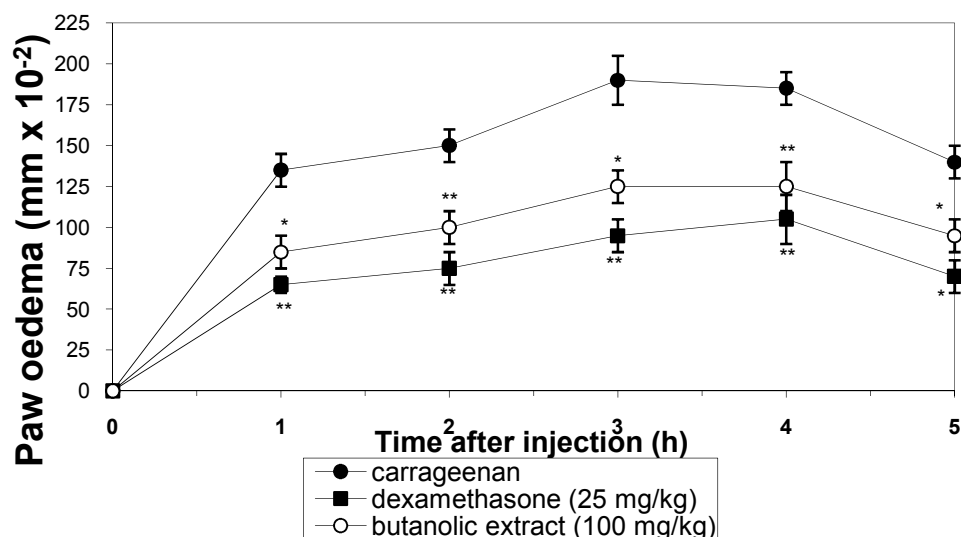


Fig. 3. Anti-inflammatory activity of the butanolic extract (100mg/kg, n=5) and reference compound dexamethasone (25mg/kg, n=5) against mouse paw edema induced by carrageenan. * P<0.05, ** P<0.01

4. CONCLUSION

The results of this study demonstrated that butanolic extract of *Albizia lebeck*, rich in saponins, has anti-inflammatory properties.

CONSENT

Not applicable.

ETHICAL APPROVAL

All authors hereby declare that "Principles of laboratory animal care" (NIH publication No. 85-23, revised 1985) were followed, as well as specific national laws where applicable. All experiments have been examined and approved by the appropriate ethics committee.

ACKNOWLEDGEMENTS

This work was financially supported by FUJB, CAPES, FINEP, and CNPq.

COMPETING INTERESTS

Author has declared that no competing interests exist.

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