EVALUATION OF ANTI-INFLAMMATORY PROPERTIES OF *Albizia lebbeck* (Suriya mara) AND *Albizia odoratissima* (Huri mara)

S.H. Punchihewa[1], A.P.P.R. Amarasinghe[1], S.P. Senanayake*[1], P.A. Paranagama[2]
[1]Department of Botany, University of Kelaniya
[2]Department of Chemistry, University of Kelaniya
**Corresponding author (email: priyangi@kln.ac.lk)*

Introduction

***Albizia lebbeck* (L.) Benth (Vern. Suriya mara) and *Albizia odoratissima* (L.F) Benth (Vern. Huri mara) are two major species with high medicinal values belong to genus *Albizia* (Family: Fabaceae). Parts of these plants are used as ingredients in many Ayurveda preparations and other indigenous medicine systems for treatment of diseases related to the respiratory tract, skin, cancer and inflammatory diseases. Due to the increasing demand of these species, adulteration has been reported in many instances. Therefore, this study aims to screen the phytochemicals of these two species and to investigate the efficacy of anti-inflammatory properties of *A. lebbeck* and *A. odoratissima,* which would be useful in standardization of the herbal formulations.**

Methodology

a) Preliminary phytochemical screening

Leaves, barks, seeds and flowers of the two *Albizia* species were collected, air dried and ground separately to obtain fine powder. Samples were subjected to sequential solvent extraction using hexane, chloroform, ethyl acetate, methanol and water in increasing polarity order. Each extract was filtered, concentrated and stored at 4 °C in clean airtight glass containers. One gram of each extract was dissolved in the Dimethyl Sulphoxide (DMSO) and original solvents to obtain a stock of 1% (v/v) concentration. The extracts were subjected to preliminary phytochemical screening.

b) Screening of *in vitro* anti-inflammatory activity

***In vitro* anti- inflammatory activity of *A. lebbeck* and *A. odoratissima* extracts were assessed by Human red blood cell (HRBC) membrane stabilizing method with slight modification. Fresh blood was collected from healthy volunteers and centrifuged at 3000 rpm for 15 minutes. Serum was removed and red blood pellets were washed with normal saline. Amount of red blood cells were measured and reconstituted to get 10% (v/v) suspension with normal saline. By using Aspirin in normal saline as the stock solution, concentration series was prepared (1.0, 0.5, 0.125, 0.0625, 0.03125 mg/mL). RBC suspension was added to each solution. Mixtures were incubated in a water bath (56 ^{o}C) for 30 minutes. They were cooled to room temperature and centrifuged at 3000 rpm for 15 minutes. Absorbance of the supernatants was measured at 560 nm. Normal saline was used as the blank solvent. The plant extract was dissolved in Dimethyl sulphoxide (0.2 mL) and normal saline (14.8 mL) was added to it. By using it as the stock solution, concentration series was prepared. RBC suspension was added to each solution. Mixtures were incubated in a water bath (56 ^{o}C) for 30 minutes. They were cooled to room temperature and centrifuged at 3000 rpm for 15 minutes. Absorbance of the supernatants was measured at 560 nm. Normal saline was used as the blank solvent.**

Solution with normal saline (14.8 mL), DMSO (0.2 mL) and RBC suspension (0.5 mL) was used as the control.

Inhibition percentage for standard and test samples was calculated using the following equation;

$$\text{Inhibition of haemolysis} = \frac{100*(\text{Absorbance of control} - \text{Absorbance of test})}{\text{Absorbance of control}}$$

Results and Discussion

According to the findings all leaf extractions of *A. lebbeck* were rich in glycosides, flavonoids, tannins and saponins while in barks of *A. lebbeck* glycosides, flavonoids, tannins, saponins and steroids were present. Methanol extract showed the highest number of phytochemicals while hexane extract showed less numbers. Alkaloids, flavonoids and glycosides were present in seeds of *A. lebbeck* (Table 1).

Table 1: Qualitative phytochemical constituents of *A. lebbeck* (+) detected, (-) undetected

	Leaves					Barks					Seeds						Flowers			
	Hexane	Chloroform	Ethyl Acetate	Methanol	Aqueous	Hexane	Chloroform	Ethyl Acetate	Methanol	Aqueous	Hexane	Chloroform	Ethyl Acetate	Methanol	Aqueous	Hexane	Chloroform	Ethyl Acetate	Methanol	Aqueous
Alkaloids	+	-	-	+	+	-	+	-	+	+	-	-	+	-	+	-	+	+	+	-
Glycosides	+	+	+	-	-	+	+	+	+	-	-	+	-	+	+	-	+	-	+	+
Saponins	+	+	+	+	+	+	+	+	+	+	-	-	-	-	-	-	-	+	+	+
Steroids	+	-	-	-	-	+	-	-	+	-	+	-	-	-	+	-	+	+	+	-
Tannins	+	+	+	+	+	+	+	+	+	+	+	-	-	+	-	-	-	-	+	+
Flavonoids	+	+	+	+	+	+	+	+	+	+	-	+	+	+	+	-	-	-	+	+
Terpenoids	+	+	-	-	-	-	+	-	-	-	-	-	-	-	-	+	-	+	-	-

Considering the *A. odoratissima,* leaves showed the presence of glycosides, saponins, steroids, tannins and flavonoids. Barks contain steroids, terpenoids, flavonoids, alkaloids, glycosides and saponins. The flowers of *A. odoratissima* were rich in glycosides, saponins, alkaloids, flavonoids, steroids and tannins. Terpenoids could not be observed in any of the extracts. Results revealed that mainly the barks and seeds of both species exhibited membrane stabilizing activity (Table 2).

Table 2: Qualitative phytochemical constituents of *A. Odoratissim,* (+) detected (-) undetected

	Leaves					Barks					Seeds					Flowers				
	Hexane	Chloroform	Ethyl Acetate	Methanol	Aqueous	Hexane	Chloroform	Ethyl Acetate	Methanol	Aqueous	Hexane	Chloroform	Ethyl Acetate	Methanol	Aqueous	Hexane	Chloroform	Ethyl Acetate	Methanol	Aqueous
Alkaloids	+	-	-	-	+	-	+	-	+	-	-	-	-	+	-	-	-	-	+	+
Glycosides	+	+	+	+	-	-	+	+	+	-	+	+	+	+	+	+	+	-	+	+
Saponins	+	-	-	-	+	-	-	-	+	+	+	-	-	+	-	+	-	+	+	+
Steroids	+	+	+	+	-	+	+	-	-	-	+	+	+	+	+	-	+	+	+	-
Tannins	+	+	+	+	-	-	-	+	+	+	+	+	+	+	+	-	-	-	+	+

Flavonoids	+	+	+	+	+	-	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Terpenoids	+	+	-	-	-	+	+	+	+	+	-	-	-	-	-	-	-	-	-	-

Mean inhibition percentage values of the extractions of leaves of both species were significantly different from the standard (Asprin, 85.9% ± 4.5). Hexane extractions showed the highest percentage of mean inhibition in both *A. lebbeck* and *A. odoratissima* leaves. Leaf extraction (Hexane) of *A. lebbeck* showed comparatively higher inhibition percentage (56.12^{b} ± 3.81) than *A. odoratissima* leaves (49.23^{b} ± 3.56). The mean inhibition values obtained for the extraction of barks in methanol of both species revealed similar anti-inflammatory activity compared to the standard. Comparatively *A. lebbeck* obtained the highest inhibition percentage (83.58% ± 5.14) whereas *A. odoratissima* obtained the lowest inhibition percentage (81.3% ± 5.33). According to these results, barks of *A. odoratissima* can be accepted as adulterate for *A. lebbeck* bark in traditional medicine. Flower extractions of both species showed lower inhibition percentages (less than 50%) compared to the standard. Therefore, it signifies that the flowers of both species do not have considerable inflammatory properties. Methanolic extractions of seeds of both species were not significantly different from the standard. That means seeds of both species have a higher anti-inflammatory property. *A. odaratissima* showed higher inhibition percentage (81.92% ± 4.19) than *A. lebbeck* (79.82% ± 4.21). Flavonoids and steroids have remarkable anti-inflammatory activity and have a correlation with the membrane stabilizing ability. The high anti-inflammatory effect of barks and seeds of *A. lebbeck* and *A. odoratissima* may be due to the presence of flavonoids, saponins and steroids as revealed in the preliminary phytochemical screening study.

Conclusion and Recomandations

Among these four plant parts, barks and seeds have showed the highest anti-inflammatory activity with similar effectiveness which can be considered in future recommendations of adulteration of barks and seeds of *A. odoratissima* for *A. lebbeck* in traditional medicine.

References

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