Antiinflammatory Activity of the Leaves of *Anacardium occidentale* Linn

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Ethanolic crude extract of *Anacardium occidentale* leaves and its five different crude fractions, petroleum ether, solvent ether, ethyl acetate, butanol and butanone were subjected to preliminary qualitative chemical investigations. The ethanolic extract and all other fractions were screened for antiinflammatory activity in albino rats (300 mg/kg). Ethanolic extract and butanone fraction exhibited significant antiinflammatory activity when compared with control and standard drug diclofenac sodium (100 mg/kg).

The search for antiinflammatory and analgesic agent in modern times was marked by the introduction of salicin for the treatment of inflammatory swellings due to rheumatic fever and rheumatoid arthritis[1]. Herbal drugs are being proved as effective as synthetic drugs with lesser side effects. Herbal medicines are in line with nature, with no hazardous reactions[2]. Of late the interest in the plant products rose all over the world due to the belief that many herbal medicines are known to be free from side effects, although this statement can he debatable. Furthermore, the fact that the discovery of a new synthetic drug is time consuming. expensive affair, and if we want to implement "Health for All by the year 2000", then perhaps the logical choice would be in exploring plant products as potential medicines to the maximum possible usage[3]. Due to the increasing frequency of intake of NSAIDs and their reported common side effects, there is need to focus on the scientific exploration of herbal drugs having fewer side effects. *Anacardium occidentale* (Anacardiceae) is a small spreading, evergreen tree, with a short thick crooked trunk, commonly known as cashew sometimes reaching a height of 12 m native to tropical America and naturalized in the warmer parts of India especially near the sea[4]. Various parts of this plant are used in the traditional systems of medicine. The fruit is acrid. sweet, hot, digestible, aphrodisiac. anthelmintic, tumors, ascites, fever, ulcers, leucoderma and skin diseases, dysentery, and piles. Decoction of the bark is used in diarrhea, diabetes, swellings and mouth ulcers. The bark and leaves are used in curing toothaches and sore gums[5,6].

Literature survey revealed that (-)-epicatechin, a biflavonoid isolated from *Anacardium occidentale* possessed significant antiinflammatory activity[7]. Beta-sitosterol, stigmasterol, sampesterol and cholesterol were isolated from the petroleum ether (60-80°) extract of the bark of *Anacardium occidentale*[8]. Myricetin, agathisflavone, robustaflavone, amentoflavone, quercetin, kaempferol, apigenin and two glycosides viz., quercetin-3-O-rhamnoside and quercetin-3-O-glucoside, are present in the methanol extract of the leaves[9].

Chloroform and methanol extracts of *Anacardium occidentale* bark and nut shell have been found to possess antimicrobial activity against gram-positive and gram-negative organisms[10]. Tannins obtained from the bark of *Anacardium occidentale* are found to possess

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antiinfiammatory activity[11]. So far very little is known about the biological activities of the leaf constituents, therefore in the present study, we report the preliminary chemical evaluation and antiinflammatory activity of the various extracts of the leaves of *Anacardium occidentale.*

The leaves were collected from a horticulture garden in Goa, India during December 1999 and were authenticated at the Department of Dravya Guna, K.L.E.S's B.M.K. Ayurvedic Medical College, Belgaum, India. A voucher specimen (A.C. -1) has been kept in our laboratory for future reference. In the present study, air dried leaves of *Anacardium occidentale,* around 1.5 kg were reduced to a fine powder and was subjected to hot continuous extraction with ethanol (95%) in 10 batches of 150 g each in a Soxhlet extractor. After the complete extraction, the solvent was distilled off and concentrated on a water bath to a dry residue.

The concentrated ethanol extract (about 250 g) was dispersed in 250 ml of distilled water and subjected to fractionation by using petroleum ether (40-60°), solvent ether, ethyl acetate, butanol and butanone in succession. Each fraction was washed with water, then dried over anhydrous Na_2SO_3 (sodium sulphite) and concentrated to a small volume and then evaporated to dryness. The dried ethanolic extracts and its fractions were stored in a desiccator and used for further experiment after suspending in gum acacia 2%[12]. The chemical constituents of the ethanolic extract and its fractions were identified by preliminary qualitative analysis and conformed by high-performance thin layer chromatography (HPTLC) for the presence of sterols, flavonoids and tannins.

Wister rats of either sex (180-200 g; M/S V. Mastiholi and co., Bangalore, India. Approved from CPCSEA Reg. No. 276.) were housed in standard metal cages. They were provided with food and water was freely available. The rats were allowed a one-week acclimatization period before the experimental sessions. The study was cleared from animal ethical committee of the institution.

The method of Winter *et al.*[13] was used to evaluate antiinfiammatory activity. The rats were divided into eight groups (each group containing 6 animals). Gum acacia 2% was used as vehicle, the suspensions of all the fractions were prepared in gum acacia and administered orally. The first group served as control and received vehicle only (5 ml/kg gum acacia 2%), second group of animals were administered standard drug diclofenac sodium through intraperitoneally (100 mg/kg). The animals of the third, fourth, fifth, sixth. seventh and eighth groups were treated with ethanol, petroleum ether, solvent ether, ethyl acetate, butanol and butanone fractions, respectively through oral route. A dose of 300 mg/kg was selected on basis of the acute toxicity studies.

A mark was made on both the hind paws just below the tibio-tarsal junction so that every time the paw could be dipped in the mercury column of plethysmograph upto the mark to ensure constant paw volume. After 30 mm of above treatment an inflammatory oedema was induced in the left hind paw by injecting 0.05 ml of carrageenan 1% w/v in saline, in the planter tissue of all the animals. The paw volume was measured at 1 h and followed by every hour till the 4 h after administration of carrageenan to each group. The difference between the initial and subsequent reading gave the actual oedema volume.

Per cent inhibition of inflammation was calculated using the formula, % inhibition = 100 (1 -vt/vc), where 'vc' represents oedema volume in control and 'vt' oedema volume in-group treated with test compound. The data were analysed using student's 't' test and the level of significance was set at $P<0.001$.

The average percentage yield of ethanolic (95%) extract of leaves of *Anacardium occidentale* was found to be 27.2% w/w and the corresponding values for petroleum ether fraction 4%, solvent ether fraction 5.2%, ethyl acetate fraction 7.8%, butanol fraction 12%, butanone fraction 25.6% w/w, respectively. Out of all the samples tested for antiinflammatory activity, the butanone fraction showed significant ($P<0.001$) oedema suppressant activity to that of diclofenac sodium in 3 h and 4 h followed by the ethanolic extract. The results are shown in Table 1.

On preliminary phytochemical screening of the leaves of *Anacardium occidentale,* the presence of sterols, tannins and flavonoids is indicated and on pharmacological screening of the crude extracts of *Anacardium occidentale* ethanol extract and its butanone fraction exhibited significant antiinflammatory activity.

Pain, swelling and fever has been treated with *Anacardium occidentale* leaves in the traditional system of medicine[5,6]. Tannins obtained from the bark of *Anacardium occidentale* are found to possess antiinflammatory activity[8]. Thus our results, support the antiinflammatory activity of *Anacardium occidentale* as claimed in the Ayurvedic Literature. Thus it can also be concluded that antiinflammatory activity may be due to combined effect of sterols, tannins and flavonoids.

TABLE 1: EFFECT OF VARIOUS EXTRACTS OF *ANACARDIUM OCCIDENTALE* ON CARRAGEENAN INDUCED RAT PAW OEDEMA.

Group	Dose	Mean increase in paw volume (ml) ± SEM				% of oedema inhibition at 4 h
		1 h	2 h	3 h	4 h	
Control	5 ml/kg**	0.45±0.06	0.49±0.09	0.57±0.0	0.58±0.00	-
Standard	100 mg/kg	0.19±0.01	0.23±0.00	0.29±0.5	0.31±0.01*	46.56
Pet, ether (40°-60°)	300 mg/kg	0.31±0.04	0.38±0.06	0.43±0.0	0.44±0.09	24.14
Solvent ether	300 mg/kg	0.36±0.01	0.39±0.06	0.42±0.0	0.45±0.05	22.42
Ethyl acetate	300 mg/kg	0.33±0.04	0.34±0.07	0.38±0.3	0.40±0.08	31.04
Butanol	300 mg/kg	0.38±0.05	0.42±0.05	0.46±0.6	0.51±0.01	12.07
Butanone	300 mg/kg	0.21±0.0	0.23±0.00	0.28±0.5	0.33±0.05*	43.11
Ethanol	300 mg/kg	0.20±0.05	0.24±0.03	0.31±0.5	0.37+0.06*	36.21

Significance relative to respective control values: *P<0.001; N=6 (N indicates no. of animals used in each group) **Gum acacia 2% as vehicle.

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REFERENCES

1. Harish V., **Pharma Times,** 2001, 33, 14.
2. Rama Rao. A.V. and Gurjar, M.K., **Pharma Times,** 1990, 22, 19.
3. Goodman and Gilman's., In; The pharmacological basis of therapeutics. 9th Edn., 1996, 617.
4. The Wealth of India- Raw Materials, Vol. 1, Publications and Information's Directorate, CSIR, New Delhi. 1985, 236.
5. Kirtikar, K.R. and Basu, B.D., In; Indian Medicinal Plants, Vol. I, Bishen Singh, Mahendra Pal Singh, Dehra Dun, 1980, 657.
6. Kurian, J.C., In; Plants that Heal, Oriental Watchman Publishing House, Pune, 1995, 34.
7. Swarnalakshmi, T. Gomathi, K. Sulochana, N. Amala Basker, E. and Parmar. N.S., **Indian J. Pharm. Sci.,** 1981, 43. 205.
8. Dinda. B., Chatterjee, J. and Banerjee. J., **J. Indian Chem. Soc.,** 1987, 64, 647.
9. Arya, R., Babu, V., Ilyas, M. and Nasim, K.T., **J. Indian Chem., Soc.,** 1989, 66, 67.
10. Sathawane. P.N., Patel, D.L., Kasture, V.S., Kasture, S.B. and Pal, S.C., **Indian Drugs.,** 1987, 34, 459.
11. Mota, M.L.R., Thomas, G. and Barbosa Filho, J.M., **J. Ethanopharmacol,** 1985, 13, 289.
12. Hukkeri, V.I., Patil, M.B., Jalalpure, S.S. and Ashraf, A., **Indian J. Pharm. Sci.,** 2001, 63, 429.
13. Winter, C.A., Risley, E.A. and Nuss. G.W., **Proc. Soc. Exp. Biol. Med.,** 1962, 111, 544.