Short Communication

Anti-inflammatory activity of *Ruta graveolens* Linn on carrageenan induced paw edema in wistar male rats

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Aqueous, ethanolic and methanolic extracts of *Ruta graveolens* were investigated for anti-inflammatory activity in carrageenan induced paw edema in wistar male rats, and compared to a positive control drug, Voveran. These extracts were given (ip) in a concentration of 20 and 50 mg/kg b.w. before carrageenan injection. Methanolic extracts of *R. graveolens* with a concentration of 20 mg/kg b.w. and ethanolic extract with a concentration of 50 mg/kg b.w. showed maximum (90.9%) inhibition on carrageenan induced rat paw edema. The effect was significantly (P< 0.05) higher than that of the standard drug Voveran (72.72%). Methanol extract with a concentration of 50 mg/kg b.w. produced 81.81% inhibition, which was also high as compared to the standard drug. Ethanolic extract with a dose of 20 mg/kg b.w and the two doses of aqueous extract produce less percentage of inhibition as compared to the standard drug voveran.

Key words: Ruta graveolens, anti-inflammation, carrageenan.

INTRODUCTION

Herbal medicines derived from plant extracts are being increasingly utilized to treat a wide variety of clinical diseases, though relatively little knowledge about their mode of action is available. There is a growing interest in the pharmacological evaluation of various plants used in Indian traditional systems of medicine. Thus, the present investigation was carried out to evaluate the antiinflammatory potential of *Ruta graveolens* in experimental animal models.

R. graveolens, commonly known as Rue, is a dicot in the family Rutaceae. It is an herbaceous perennial, originally native to the Mediterranean region. It is now cultivated in many parts of the world. It has blue-green foliage and yellow flowers. There are two main species used in traditional medicine: *Ruta chalepensis* and *R. graveolens* (lauk et al., 2004). The plant is used as a contraceptive (Browner, 1985), to relieve symptoms of hangover (Chávez et al., 2003), applied externally as a poultice against rheumatic pain (Chávez et al., 2003; Atta and Alkofahi, 1998; Linares, 1994). Rue's active ingredients may have antifungal property, which could be beneficial

to agriculture and medicine (Mancebo et al., 2002; Trovato et al., 2002; Ojala et al., 2000).

On the basis of these common uses of this plant in traditional folk medicine and its above reported activities in the literature, we have evaluated the anti-inflammatory effect of various extracts of *R. graveolens*

MATERIALS AND METHODS

Collection of plant materials

Arial part of *R. graveolens* were collected from Kannur district of Kerala state, India. The plant was identified and authenticated by Dr. Valsaladevi G, Department of Botany, and University of Kerala.

Aqueous extract

500 g arial parts of *R. graveolens* were collected, washed thoroughly and dried in shade. It was then crushed and taken in a round-bottomed flask. 500 ml distilled water was added to cover the material, refluxed in a water bath for 1 h at 90 - 95° C. The supernatant was removed and the extraction repeated once again. The supernatant obtained were combined and filtered through a Whatman No. 1 filter paper. The filtrate was concentrated at low temperature by lyophilization. The residue was designated as aqueous extract.

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Extracts	Doses (mg/kg, ip)	Change in paw edema mean (mm)	% Edema inhibition relative to control at 3 rd hour
Control (normal Saline, 0.9%)	0.3 ml	1.1 ± 0.05	
Aqueous extract			
	20	$0.9 \pm 0.03^{*}$	18.2
	50	0.7 ± 0.27*	36.3
Methanolic extract			
	20	0.1 ± 0.004*	90.9
	50	0.2 ± 0.007*	81.81
Ethanolic extract			
	20	$0.4 \pm 0.02^*$	63.6
	50	0.1± 0.003*	90.9
Voveran			
	20	0.3 ± 0.01*	72.72

Table 1. Anti-inflammatory activity of the aqueous, ethanolic and methanolic extract of *R. graveolens*.

Values are mean ± S.E.M.

* P< 0.05, significantly different from control.

Methanol extract

Dried arial parts of *R. graveolens* were reduced to a fine powder with a mechanical grinder. The powder plant material (200 g) was soaked in 3 l of 70% methanol and allowed to stand for 3 days for extraction. The extract was concentrated to dryness using a rotary evaporator attached to a vacuum pump and stored at a temperature of -4° C until use.

Ethanol extract

Dried ariel parts of *R. graveolens* were reduced to a fine powder with a mechanical grinder. The powder plant material (200 g) was soaked in 3 l of 80% ethanol and stand for 3 days. The extract was concentrated to dryness and stored at a temperature of -4° C until use.

Anti-inflammatory activity

Male Wistar rats (120 - 170 g) kept at the laboratory Animal home of the Faculty of Biochemistry, University of Kerala, India were used. The animals were maintained under standard environmental conditions and had free access to standard diet and water. Antiinflammatory activity was measured using carrageenan induced rat paw oedema assay (Winter et al., 1962; Adeyemi et al., 2002). Groups of 6 rats were given a dose of the extract (plant extracts were dissolved in sterile distilled water and administered intra peritoneally at different dose levels). After 1h, 0.1 ml, 1% carrageenan suspension in 0.9% NaCl solution was injected into the sub-plantar tissue of the right hind paw. The linear paw circumference was measured at hourly interval for 3 h (Bamgbose and Noamesi, 1981) Anti-inflammatory activity was measured as the percentage reduction in oedema level when drug was present, relative to control (Duffy et al., 2001) as shown in Table 1

Statistical analysis

All data were expressed as mean \pm SEM and one-way ANOVA was applied to determine the significance of the difference between the control groups and rat treated with the test compounds.

RESULTS AND DISCUSSION

Carrageenan-induced rat paw oedema is used widely as a working model of inflammation in the search for new anti-inflammatory drug. The anti inflammatory activity of the aqueous, methanolic and ethanolic extract of R. graveolens was evaluated by carrageenan-induced rat paw oedema method (Winter et al., 1962; Adeyemi et al., 2002) and the result is shown in Table 1. The extracts were tested at two different dose levels. The results showed that the methanolic extract with a dose of 20 mg/kg b.w and ethanolic extract with a concentration of 50 mg/kg b.w. showed 90.9% of inhibition on carrageenan induced rat paw edema at third hour. This result indicated that methanolic extract with a dose of 20 mg/kg b.w and ethanolic extract with a concentration of 50 mg/kg b.w. showed a maximum anti-inflammatory activity as compared to the reference drug Voveran, which showed only 72.72% inhibition. Methanolic extract with a dose 50 mg/kg b.wt produced 81.81% of inhibition and is also high as compared to the reference drug. Ethanolic extract with a dose of 50 mg/kg b.wt produced 63.6% of inhibition and is low as compared to the reference drug. Aqueous extract with two different doses 20 mg/kg b.w and 50 mg/kg showed only 18.2% and 36.3% inhibition respectively. It was lower as compared to the reference drug.

The development of odema in the paw of the rat after the injection of carrageenan is due to release of histamine, serotonin and prostaglandin like substances (Vinegar et al., 1969). Significantly high anti-inflammatory activity of methanolic extract (20 mg/kg b.wt) and ethanolic extract (50 mg/kg b.w) of *R. graveolens* may be due to inhibition of the mediators of inflammation such as histamine, serotonin and prostaglandin. The present result indicates the efficacy of methanolic extract (20 mg/kg b.wt) and ethanolic extract (50 mg/kg b.wt) of *R.* graveolens as an efficient therapeutic agent in acute antiinflammatory conditions.

REFERENCE

- Adeyemi OO, Okpo SO, Ogunti OO (2002). Analgesic and antiinflammatory effect of the aqueous extract of leaves of *Persea americana* Mill (Lauraceae). Fitoterapia. 73: 375-380.
- Atta AH, Alkofahi A (1998). Anti-nociceptive and anti-inflammatory effects of some Jordanian medicinal plant extracts. J. Ethnopharmacol. 60(2): 117-124.
- Bamgbose SOA, Noamesi BK (1981). Studies on cryptolepine II:Inhibition of carrageenan induced oedema by cryptolepine. Planta Med. 41: 392-396.
- Browner CH (1985). Plants used for reproductive health in Oaxaca, Mexico. Econ. Bot. 39(4): 482-504.
- Chávez M, Franco I, González M (2003). Tlatenco: Tradición Herbolaria y Remedios Caseros.Mexico City: Ce-Ácatl, A.C.pp. 80-81.
- Duffy JC, Dearden JC, Rostron C (2001). Design, Synthesis and biological testing of a novel series of anti- inflammatory drugs. J. Pharm. Pharmacol. 53: 1505-1514.
- Iauk L, Mangano K, Rapisarda A (2004). Protection against murine endotoxemia by treatment with Ruta chalepensis L., a plant with antiinflammatory properties. J. Ethnopharmacol. 90(2-3): 267-272.

- Mancebo F, Hilje L, Mora GA (2001). Biological activity of Ruta chalepensis (Rutaceae) and Sechium pittieri (Cucurbitaceae) extracts on Hypsipyla grandella (Lepidoptera: Pyralidae) larvae. Rev. Biol. Trop. 49(2): 501-508.
- Ojala T, Remes S, Haansuu P (2000). Antimicrobial activity of some coumarin containing herbal plants growing in Finland. J. Ethnopharmacol. 73(1-2): 299-305.
- Trovato A, Monforte MT, Forestieri AM, Pizzimenti F (2000). In vitro anti-mycotic activity of some medicinal plants containing flavonoids. Boll Chim Farm. 139(5): 225-227.
- Vinegar R, Schreiber W, Hugo R (1969). Biphasic development of carrageenan in rats. J. Pharmacol. Exp. Ther. 166: 96-103.
- Winter CA, Risley EA, Nuss GW (1962). Carrageenan-induced oedema in the hind paw of rat as an assay for anti-inflammatory activity. ProcSoc. Exp. Biol. Ther. 111: 544-547.