RESEARCH ARTICLE

In-Vitro Antioxidant and anti-Inflammatory activity of *Mussaendaphilippica and Mussaendaincana*

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ABSTRACT

The present research work has been undertaken with both *Mussaendaphilippica and Mussaendaincana* for antioxidant screening and to investigate the anti-inflammatory activity. Anti-inflammatory activity was done by carrageenan induced Hind paw edema method using a plethysmometer. Diclofenac used as a standard drug and control receive saline. Antioxidant activity was evaluated by DPPH assay method. For both of these activity test groups received methanolic extract (100, 200 and 400 mg/kg). Phytochemical screening of extracts of *Mussaendaphilippica and Mussaendaincana*revealed the presence of carbohydrates, Saponins, phytosterols, fix oil and fat,tannins, flavanoids and glycoside. In acute oral toxicity study, it was observed that there was no mortality at any doses up to 2 gm/kg. Both methanolic extract shows anti-inflammatory and antioxidant activity. The present studies support traditional use of *Mussaendaphilippica and Mussaendaincana*for its anti-inflammatory activity.

Keywords: Anti-inflammatory activity, *Mussaendaphilippica, Mussaendaincana,* Carrageenan

INTRODUCTION

Inflammation is a major condition associated with various diseases. Various molecules have been isolated from the plants which have been proven very effective in such condition. Drugs which are in use presently for the management of pain and inflammatory conditions are either narcotics e.g. opioids or non-narcotics e.g. salicylates and corticosteroids e.g. hydrocortisone. All of these drugs present well known side and toxic effects. It is well documented that these nonsteroidalanti-inflammatory drugs (NSAIDs) produce intestinal tract ulcers (with potential internal bleeding) in 10-30 percent of long-term users, and erosions of the stomach lining and intestinal tract in 30-50 percent of cases.[1] [1] As a result of these side effects, NSAID use is associated with 10,000 - 20,000 deaths per year in the U.S.[2] [2] even the new COX-2 inhibitor drugs have only been reported to reduce intestinal tract damage by 50 percent, and their toxicity to the liver and kidneys is still under review.[3] [3]

Mussaendaphilippica and *Mussaendaincana*are unexplored medicinal plant and traditional practitioners using the crude extract of this plant for many disorders. To support the traditional claim the present study was designed to evaluate the antioxidant and anti-inflammatory activity of methanolic extract of *Mussaendaphilippica*and *Mussaendaincana*against experimental animal model.

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Materials and methods

Plant material

The stems of *Mussaendaphilippica* and *Mussaendaincana*stems were collected in 2012 from Thalakona and Tirumala, Chitoor district. Andhra Pradesh. Botanical identification of the plants was done by Dr. K. MadhavaChetty, Department of Botany, S.V. University, Tirupati.

Animals

Wistar albino rats of either sex weighing 150-200 g werekept at departmental animal house of Annamacharya College of Pharmacy, Rajampet, at a temperature (25°C±2) and 12 h light/dark cyclerespectively for one week before and during the experimentsand fed with standard diet and water *ad libitum*. Animal studies were conducted according to the Institute Animal Ethics Committee regulations approved by the Committee for the Purpose of Control and Supervision of Experiments on Animals.

Preparation of extract

The dried stem samples were cut into small pieces and reduced to coarse powder by mechanical grinder, extracted with methanol as solvent in soxhlet extractor for 72 h. The extract was filtered and concentrated under reduced pressure using rotavapor. The methanolic extract of *Mussaendaphilippica* (MEMP) and *Mussaendaincana* (MEMI) wasfreeze-dried and stored in deep freezer for further use.

Preliminary phytochemical screening

The methanolic extract obtained was tested for the presence of various chemical constituents such as saponins, flavonoids, glycosides, alkaloids, tannins and reducing sugar.[4]

Determination of antioxidant activity by DPPH method

A 0.004% DPPH solution in methanol was prepared and 4 ml of this solution was added to 1 ml of sample extract solution in methanol at different concentrations (50-, 100, 200, 400 and 600 µg/ml). Left it for 30 minutes at room temperature for the reduction of the DPPH free radical and the absorbance was measured at 517 nm. The procedure was repeated for Vitamin C (Ascorbic acid) which was used as standard. The antioxidant activity of the extracts was expressed as IC_{50}, which is the inhibitory concentration required to scavenge 50% of DPPH free radicals.[5]

Acute toxicity study

Acute oral toxicity study was performed as per OECD – 423 guidelines (acute toxic class method). Albinowistar rats of either sex were selected randomly and treated with methanolicextract of *Mussaendaphilippica* and *Mussaendaincana* up to the dose of 2000 mg/kg body weight p.o., the animals were observed for any toxic signs for initial period of 2 h and further 24 h for mortality study. The animal does not show any signs of toxicity and mortality up to the dose of 2000 mg/kg.

Carrageenan-induced rat paw edema[6]

The rats were divided into eight groups, each group consisting of six animals. Edema was induced by subplantarinjection of 0.1% freshly prepared carrageenan suspention into the right hind paw of each rat. The paw volume was measured at 0,1, 2, 3 and 4 h after the injection of carrageenen, using a digital plathysmometer. Group I rats were received vehicle (distilled water 10 ml/kg BW p.o.), group II rats received 10 mg/kgdiclofenac sodium as positive drugcontrol and themethanol extract of *Mussaendaphilippica*and *Mussaendaincana*at 100, 200 and 400 mg kg doses was administered orally to Group III, IV, V, VI, VII and VIII groups of rats.Drug pretreatment was given 1 h before the injection of carrageenen.

Statistical analysis

The data are expressed a s mean ± S.E.M. of six observations. The results were obtained statistically analyzed by Oneway analysis of variance by using graph pad prism software version 5. Value $P < 0.05$ was taken as the criterion of significance.

RESULTS

Phytochemical screening

The methanolic extract of *Mussaendaphilippica* and *Mussaendaincana* was tested and showed presence ofalkaloids, carbohydrates, saponins, flavanoids, anthocyanins and tannins.

Antioxidant activity of MEMP and MEMI

Several concentrations ranging from 50 to 600 µg/ml of MEMP stems and MEMI were tested for their antioxidant activity by DPPH method.DPPH has been widely used to evaluate the effectiveness of various antioxidant substances. It was observed that the test compounds scavenged the free radicals in a concentration-dependent manner.

Table 1: *In-vitro* antioxidant activity of MEMP and MEMI

S. No	Concentration (µg/ml)	Percentage inhibition (%)		
		MEMP	MEMI	Ascorbic acid
1.	50	39.4	33.5	69.4
2.	100	55.4	49.9	73.5
3.	200	67.4	88.3	83
4.	400	83.08	88.9	94.2
5	600	86.7	85	95.2
EC_{50} value **(µg/ml)**		**96**	**102**	**36**

Anti-inflammatory activity of MEMP and MEMI

Methanolic extract of MEMP and MEMI significantly reduced the inflammation produced by carrageenan administration at 4th h. the results were present in table 2.

Table 2: Anti-inflammatory activity of MEMP and MEMI

Treatment	0 min	1 hr	2 hrs	3 hrs	4 hrs
GROUP I	1.18±0.028	1.28±0.054#	1.34±0.015#	1.38±0.016#	1.12±0.035#
GROUP II	1.13±0.044	0.86±0.028**	0.80±0.011***	0.75±0.011***	0.41±0.020***
GROUP III	1.18±0.07	1.12±0.072***	1.09±0.06**	1.05±0.084**	1.05±0.015***
GROUP IV	1.12±0.055	0.98±0.043***	0.85±0.046***	0.80±0.043**	0.80±0.073***
GROUP V	1.06±0.065	0.96±0.009***	0.85±0.007***	0.74±0.03***	0.72±0.022***
GROUP VI	1.16±0.07	1.11±0.66	1.07±0.069	1.02±0.084	1.06±0.015
GROUP VII	1.06±0.065	1.00±0.043	0.87±0.046	0.78±0.034	0.80±0.073
GROUP VIII	1.12±0.058	0.97±0.009	0.85±0.007	0.74±0.03	0.72±0.022

All values are shown as mean ±SEM. One Way ANOVA followed by Dunnet's Multiple Comparison Test. and n=6. *p<0.05, **p<0.01 and ***p<0.001 when compared with control group.

Group I: rats were received vehicle (distilled water 10 ml/kg BW p.o.),

Group II: rats received 10 mg/kgdiclofenac sodium

Group III-VIII: methanol extract of *Mussaendaphilippica*and *Mussaendaincana*at 100, 200 and 400 mg kg doses respectively

DISCUSSION

The most widely used primary test to screen new anti-inflammatory agents, measure the ability of a compound to reduce local edema induced in the rat paw by an injection of an irritant agents.[7] carrageenan induced edema has been commonly used as an experimental animal model for acute inflammation and is believed to be biphasic. The early phase (1-2 h) of the carrageenan model is mainly mediated by histamine, serotonin and increased synthesis of prostaglandins in the damaged tissue surroundings. The late phase is sustained by prostaglandin release and mediated by bradykinin, leukotrienes, polymorphonuclear cells and prostaglandins produced by tissue macrophages.[8,9]

In the present study, the methaolic extract of MEMP and MEMI significantly produced anti-inflammatory activity against carrageenan indued paw edema model. The effect was comparable to standard drug diclofenac sodium. The methanolic extract of MEMP and MEMI in all doses shows significant protection.

CONCLUSION

The methanolic extract of MEMP and MEMI significantly shows anti-inflammatory activity and antioxidant property.

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REFERENCE

1. Hayliyar J, Macpherson A, Bjarnason I. Gastro protection and nonsteroidal anti-inflammatory drugs. Drug Saf 1992, 7, 86-105.

2. Ament PW, Childers RS. Prophylaxis and treatment of NSAID induced gastropathy. Am FamPhys 1997, 4, 1323-1326
3. Silverstein FE, FaichG, Goldstein JL, Simon LS, Pincus T, Whelton A. Gastrointestinal toxicity with celecoxibvsnonsteroidal anti-inflammatory drugs for osteoarthritis and rheumatoid arthritis, JAMA. 2000, 284, 1247-1255.
4. Evans WC, Trease. Text Book of Pharmacognosy, 15th Ed; ELBS London, 2002.
5. Koti BC, Ashok P, Diuretic activity of extract of Mimusopselengi Linn. Bark, Int. J. Green. Pharm.2010, 4, 90-2.
6. Winters WD, Hance AJ, Cadd GG, Quam DD, Benthuysen JL. Study of analgesic and anti-inflammatory activity from plant extract of LactucaScariola and Artemisia Absinthium, J PharmacolExp Therapeutics. 1987, 244, 51-57.
7. Winter CA, Risley E, Nuss G. Carrageenan induced edema in hind aw of the rat as an assay for anti-inflammatory drugs, Proc. Soc. Exp. Biol. Med. 1962, 544-547.
8. Gupta M, Mazumder UK, Gomathi P, Thamilselvan V, Antiinflammatory evaluation of leaves of Plumeria acuminate, Vol.6 BMC Complementary and alternative medicine, 2006, 1472-6882
9. Koti BC, Ashok P, Diuretic activity of extract of Mimusopselengi Linn. Bark, Int. J. Green. Pharm.2010, 4, 90-2

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