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EVALUATION OF ANTI-INFLAMMATORY ACTIVITY OF FRUITS OF *TRAPA NATANS* LINN

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ABSTRACT

Fruit of *Trapa natans* L. var. *bispinosa* commonly known as Shingoda and is reported as anti-inflammatory in traditional literature. So, the present research work has been undertaken with both pericarp and seed extract of fruits of *Trapa natans* for phytochemical screening and to investigate the anti-inflammatory activity. Anti-inflammatory activity was done by carrageenan induced Hind paw edema method using a plethysmometer. Aspirin used as a standard drug and control receive saline. For both of these activity test groups received pericarp extract (200 mg/kg and 300 mg/kg) and seed extract (200 mg/kg and 300 mg/kg) of *T.natans*. Phytochemical screening of seed extracts of fruits of *T.natans* revealed the presence of carbohydrates, Saponins, phytosterols, fix oil and fat while in pericarp extract of fruits of *T.natans* revealed the presence of tannins, flavanoids and glycoside & their presence were substantiated by thin layer chromatographic studies. In acute oral toxicity study, it was observed that there was no mortality at any doses up to 2 gm/kg. Both aqueous extract of pericarp and seed shows anti-inflammatory activity but pericarp shows more potent action than seed. The present studies support traditional use of fruits of *T.natans* for its anti-inflammatory activity.

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Key Words

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INTRODUCTION

Inflammation is a major condition associated with various diseases. Rheumatoid arthritis is one of the challenging disorders associated with inflammatory condition. 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 nonsteroidal anti-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] As a result of these side effects, NSAID use is associated with 10,000 - 20,000 deaths per year in the U.S. ^[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]

Fruit of *Trapa natans* L. var. *bispinosa* commonly known as Shingoda and is reported as anti-inflammatory in traditional literature. Systemic studies with respect to standardization of this drug are not reported in literature; therefore an attempt has been made to develop their standardization profile. Attempts are being made globally to get scientific evidences for these traditionally reported anti-inflammatory herbal drugs to sort out the problems.

So, the present research work has been undertaken with the aqueous fruit extract of *Trapa natans* L. var. *bispinosa* stand for phytochemical screening and to investigate anti-inflammatory activity.

MATERIALS AND METHODS

MATERIAL

All the chemicals used were of analytical grade available. All the solvent used in the experiments were of analytical grade. Carrageenan was obtained from Sarda Gums & Chemicals, Mumbai. The fruits of *Trapa natans* were collected from Mehsana district of Gujarat, India. Their identity was confirmed at the Department of

Botany by Dr. M.M. Prajapati, HNSB Science college, Himatnagar.

METHODS

Morphological evaluation of fruits of *Trapa natans*.

The morphology description of a crude drug include size, shape, nature of outer surfaces, types of fracture, and organoleptic characters like colour, odour, taste etc. ^[4]

Pharmacological investigation

Animals

Wister albino rats (180-250 g) and mice (20-25 g) of either sex have used for the Diuretic and anti-inflammatory activity & acute oral toxicity. They were gifted by Zydus Cadila Pharmaceuticals, Ahmedabad. The animals were grouped and housed in polyacrylic cages, with not more than two animals per cage. The animals were kept at 27 ±2°C, relative humidity 44–56% and light and dark cycles of 10 and 14 h respectively for 1 week before and during the experiments. Animals were provided with standard diet. ^[5] Experiment was performed according to current guidelines for the care of the laboratory animals and the ethical guidelines for the investigation of experimental pain in conscious animals. The Protocol of the experiment was approved by the Institutional Animal Ethical Committee as per the guidance of the Committee for the Purpose of Control and Supervision of Experiments on Animals (CPCSEA), Ministry of Social Justice and Empowerment, Government of India (Proposal no. 2011/03, APMC, Himatnagar).

Acute toxicity study

The acute oral toxicity study was carried out as per the guidelines set by organization for economic co-operation and development (OECD) revised draft guidelines 423 B ("Up and Down" method) received from committee for the purpose of control and supervision of experiments on animals (CPCSEA), Ministry of Social Justice and Empowerment, Government of India (See annexure A-6). The LD₅₀ cut-off dose for AqE of Fruits and 1/10th of the LD₅₀ dose is taken as a therapeutic dose. ^[6,7]

In Vivo Anti-inflammatory activity

Hind paw edema method

In present study anti-inflammatory activity is determined in albino rats of either sex according to the method Winters et al. [8] Using six animals in each group. The animals were injected carrageenan (1% w/v suspension in 0.9% saline) [9] in the right hind foot under the plantar aponeurosis.

- The test groups of rats were given orally 200 and 300 mg/kg of aqueous pericarp extract of plants one hour before the carrageenan injection.
- Another test groups of rats were given orally 200 and 300 mg/kg of aqueous seed extract of plants one hour before the carrageenan injection.
- The controls were given the same volume of saline as in test group.
- Another group of rats were treated with 300 mg/kg of acetylsalicylic acid orally one hour before carrageenan injection.

The inflammation was quantitated in terms of ml i.e. replacement of water by edema using a plethysmometer immediately before carrageenan injection and then 1, 3 and 5 hours after carrageenan injection. The percent inhibition of edema as calculated for each group with respect to its vehicle-treated control group. The anti-inflammatory activity was calculated by using the relation used by

$$\frac{V_c - V_t}{V_c} \times 100$$

Used by Planichamy, 1990 [10] where V_c and V_t denote mean increase in paw volume of control and drug-treated animals respectively.

Statistical analysis

Values for anti-inflammatory activity are expressed as "mean increase in paw volume \pm SEM". The significance of difference between means is determined by student's t-test values of $p < 0.05$ are considered significant. [11]

RESULTS

Pharmacognostic investigation

Morphological evaluation of fruit of *Trapa natans*.

Fruits are Large (2.5 cm), variously-shaped nuts are swollen at the middle and have 2-4 sharp spines. fruits obovoid, bony, angular with a short conical beak in the centre at the apex and a spreading, flattened, very sharp spinous horn on either side, indehiscent, one seeded, seed white.

ORAL ACUTE TOXICITY STUDY

In acute oral toxicity study, It was observed that there was no mortality at any of the doses (up to 2 gm/kg) at the end of 14 days of observation. so it suggested that its LD₅₀ is more than 2 gm/kg.

IN VIVO ANTI-INFLAMMATORY ACTIVITY

The anti-inflammatory activity was compared between pericarp and seed parts of fruits of *Trapa natans* along with control (saline). From the result it can be observed that aqueous extract of pericarp has shown significant anti-inflammatory activity by decreasing paw volume at 3hr and 5hr compared with control while in the aqueous extract of seed shows significant anti-inflammatory activity by decreasing paw volume at 5hr compared with control. The Result obtained in anti-inflammatory assay of fruits of *Trapa natans* are shown in Table 1 & Table 2. Fig. 1 shows percentage inhibition at different time interval for different groups.

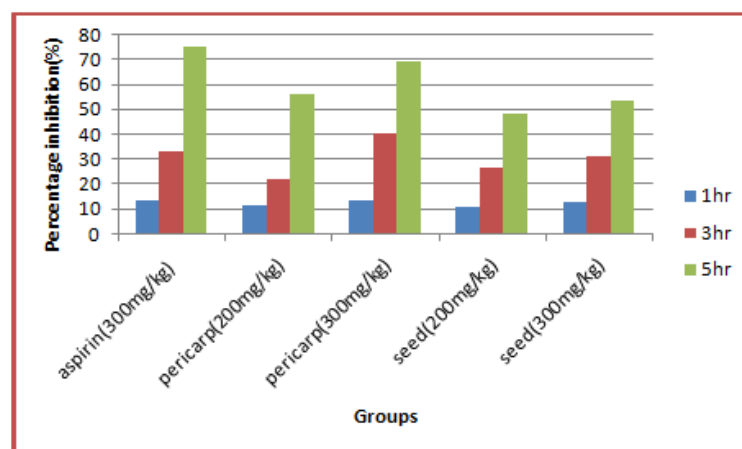


Fig. 1: Anti-inflammatory activity of *T.natans* shows Time vs percentage inhibition

Table 1: *In Vivo* Anti-inflammatory activity of aqueous extract of fruits of *Trapa natans* measuring Paw volume.

Treatment group	Mean Paw volume(ml) ± SEM			
	Before injection	1hr	3hr	5hr
Control (saline)	1.35 ± 0.03	2.44 ± 0.15	2.65 ± 0.09	3.07 ± 0.19
Aspirin (300 mg/kg)	1.42 ± 0.06	2.36 ± 0.05	2.29± 0.05**	1.85±0.03**
Aqueous pericarp extract (200 mg/kg)	1.35 ± 0.09	2.31 ± 0.12	2.36 ± 0.12*	2.10±0.09**
Aqueous pericarp extract (300 mg/kg)	1.48 ±0.07	2.42 ± 0.07	2.25± 0.04**	2.00±0.06**
Aqueous seed extract (200 mg/kg)	1.46 ± 0.05	2.43 ± 0.04	2.41 ± 0.04	2.34±0.02**
Aqueous seed extract (300 mg/kg)	1.50 ± 0.05	2.45 ± 0.05	2.39 ± 0.05	2.30±0.07**

All the values are expressed as mean ± SEM (n=6), *P<0.05, **P<0.01 compared with control (Dunnett's Test)

Table 2: *In Vivo* Anti-inflammatory activity of aqueous extract of fruits of *Trapa natans* measuring Percentage inhibition at various time intervals.

Treatment group	Percentage inhibition (%) at various times intervals		
	1hr	3hr	5hr
Aspirin (300 mg/kg)	14.06	33.33	75.19
Aqueous pericarp extract (200 mg/kg)	11.92	22.30	56.39
Aqueous pericarp extract (300 mg/kg)	13.76	40.76	69.76
Aqueous seed extract (200 mg/kg)	11.00	26.92	48.83
Aqueous seed extract (300 mg/kg)	12.84	31.53	53.48

DISCUSSION AND CONCLUSION

In the present investigation, Fruit of *Trapa natans* traditionally used as anti inflammatory was evaluated for its activity by *In Vivo* (Hind paw edema method) study. This study was carried out for both pericarp and seed parts of the fruit and compared these activities in both of this part.

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 injection of an irritant agent.^[12] Carrageenan induced oedema 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.^[13, 14]

The anti-inflammatory activity was expressed as "mean increase in paw volume \pm SEM" in terms of ml and percentage inhibition in paw volume by different doses of the extract. The result of the present study revealed that aqueous pericarp extract of *T.natans* present a potent anti-inflammatory activity by inhibitory effect on "mean increase in paw volume" induced by carrageenan injection in the sub-plantar region of rat's paw. The aqueous pericarp and seed extract produced dose-dependent and significant inhibition of carrageenan-induced paw edema. The aqueous pericarp extract of *T.natans* has shown significant (** $P < 0.01$ and * $P < 0.05$) anti-inflammatory activity by decreasing mean paw volume at 3hr at the dose of 300mg/kg and 200mg/kg while in seed extract at dose of 300mg/kg and 200mg/kg do not show any significant anti-inflammatory activity by decreasing mean paw volume compared with control. While at 5hr both the aqueous extract of pericarp and seed shown significant (** $P < 0.01$) anti-inflammatory activity by decreasing mean paw volume compared with control.

In conclusion, the present studies support traditional use of fruits of *T.natans* for its anti-inflammatory activity.

REFERENCES

1. Hayliyar J, Macpherson A, and 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. Fam. Phys.* 1997, 4, 1323-6.
3. Silverstein FE, Faich G, Goldstein JL, Simon LS, Pincus T, Whelton A, et al. Gastrointestinal toxicity with celecoxib vs nonsteroidal 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. Winters WD, Hance AJ, Cadd GG, Quam DD, Benthuyzen JL, Study of analgesic and anti-inflammatory activity from plant extract of *Lactuca Scariola* and *Artemisia Absinthium*, *J Pharmacol Exp Therapeutics.* 1987, 244, 51-57.
6. Gosh MN, *Fundamentals of experimental pharmacology.* 3rd Ed, Hilton and company, Kolkata, 2005, pp 190-197.
7. OECD, *Guideline for testing of chemicals*, No.420, Acute oral toxicity-fixed dose procedure, Paris, France: Organization for Economic Cooperation and Development. 2000.
8. Ocete MA, Risw S, Zarzuelo A, Study of analgesic and anti-inflammatory activity from plant extract of *Lactuca Scariola* and *Artemisia Absinthium*, *J Ethnopharmacol.* 1988, 25, 305-313.
9. Planichamy S, Study of analgesic and anti-inflammatory activity from plant extract of *Lactuca Scariola* and *Artemisia Absinthium*, *Fitoterapia.* 1990, 61, 73-78.
10. Alcaraz MJ, Jimenez MJ, Study of analgesic and anti-inflammatory activity from plant extract of *Lactuca Scariola* and *Artemisia Absinthium*, *J Nat Prod.* 1983,52,1088-1091.
11. Brito ARMS, Antonio MA, Oral antiinflammatory and antiulcerogenic activities of a hydroalcoholic extract and partitioned fractions of *Turnera*

- ulmifolia* (Turneraceae), J Ethnopharmacol. 1998, 61, 215-228.
12. Winter CA, Risley E, Nuss G, Carrageenan-induced edema in hind paw of the rat as an assay for anti-inflammatory drugs, *Proc. Soc. Exp. Biol. Med.* 1962, 544-547.
13. Gupta M, Mazumder UK, Gomathi P, Thamilselvan V, Antiinflammatory evaluation of leaves of *Plumeria acuminata*, Vol.6 BMC Complementary and alternative medicine, 2006, 1472-6882.
14. Koti BC, Ashok P, Diuretic activity of extract of *Mimusops elengi* Linn. Bark, *Int. J. Green. Pharm.* 2010, 4, 90-2.
