Section: Pharmacology

Original Research Article

EVALUATION OF ANTI-INFLAMMATORY ACTIVITY OF MORINGA OLEIFERA LAM. LEAVES EXTRACT TOPICAL GEL FORMULATION IN WISTAR RATS

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Received : 06/07/2024
Received in revised form : 01/09/2024
Accepted : 15/09/2024

Keywords:
Anti-inflammatory activity, Moringa oleifera Lam, Topical Gel, Carrageenan.

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DOI: 10.47009/jamp.2024.6.5.52

Source of Support: Nil,
Conflict of Interest: None declared

Int J Acad Med Pharm
2024; 6 (5); 282-284

Abstract

The topical preparation deliver drug directly to the target site and have less side effect(s) than other route of administration. Topical gel of ethanolic extract of Moringa oleifera Lam. leaves were tested to see the anti-inflammatory activity, in carrageenan induced paw edema in Wistar rats by using plethysmometer. The 2% gel leaves extract of Moringa oleifera Lam. showed significant reduction in paw volume compared to control. The observations indicate that ethanolic extract of Moringa oleifera Lam. leaves has potential topical anti- inflammatory activity.

INTRODUCTION

Moringa oleifera Lam, a tree of intermediate length, falls under Moringaceae. The family consists of a genus and 4-10 species. M. oleifera and M. conconcane, two common species, are distinguishable from the attributes of leaves.[1-3] Numerous medicinal properties have been credited to different parts of the plant including anti-inflammatory.[4]

Inflammation, a beneficial adaptive response to offender(s) and consequences of them, however, when dysregulated is a cause for numerous pathological state. The process is mediated in its ultimacy by variety of inflammatory mediators including prostaglandins.[1]

Multiple drugs with anti-inflammatory property are available but with many side effects.[2] Therefore there is need of an active drug but of diminished adverse potential.

The anti- inflammatory effect of various parts including leaves have been shown by number of experiments,[5] however, no anti-inflammatory effect of the topical preparation of leaves has been evaluated, till date, to the best of authors knowledge. Hence, this study was undertaken to evaluate the anti-inflammatory activity of topical application of Moringa oleifera Lam. leaves extract in Wistar rats.

MATERIALS AND METHODS

Plant material: The leaves of Moringa oleifera Lam. were collected from Manipal, Udupi, INDIA and it was authenticated by Dr. K. Gopalakrishna Bhat, Professor of Botany (Rtd.), Taxonomy Research, Centre, Department of Botany, Poornaprajna College Udupi – 576 101, Karnataka.

Chemicals: Carbopol 940, Diclofenac gel, Propylene glycol 400, Ethanol, Methyl paraben, Propyl paraben, Ethylenediaminetetraacetic acid (EDTA), Triethanolamine, Carragennan were purchased from the market

Preparation of plant extract: Leaves were shade dried, and coarsely powdered with cutter mill. The powder was subjected for extraction using Soxhlet apparatus and ethanol as solvent. The extracts were concentrated on a water bath at a temperature below 50° C and dried in a desiccator, stored in refrigerator until used.

Animals: The adult, female, Wistar rats weighing between 150-200 g were obtained from Central animal house and maintained under constant conditions (temperature 25±2°C, humidity 40-60%, 12 h light and 12 h dark cycle). During maintenance they received a standard diet and water ad libitum. The experiment was approved by the institutional animal ethics committee (IAEC/KMC/29/2015).

Preparation of topical gel: The extract gels were prepared using dried alcoholic extract of Moringa oleifera Lam leaves. The water required for these formulation was divided into two parts. In one part,

ISSN (O): 2687-5365; ISSN (P): 2753-6556

the exact amount of extract was dissolved to which calculated amount of propylene glycol 400 & ethanol was added while in other, carbopol-940 was dissolved to which methyl paraben, propyl paraben, EDTA were added. Both of these solutions were mixed in a beaker to which triethanolamine was added to obtain a gel consistency and pH. Blank gel was prepared in a similar fashion.[6]

Methodology: The Wistar albino rats were divided into four groups of six rat in each. The group-1 animals had received topical gel without containing ethanolic leaves extract (blank gel), while group-3 and 4 animals had received ethanolic leaves extract gel at dose of 2% and 4% respectively. The group-2 had received standard diclofenac 0.5% topically to left leg one hour before the injection of carrageenan.[6-8]

Assessment of anti-inflammatory activity: The anti-inflammatory activity of topical Moringa oleifera Lam. against acute inflammation was tested using carrageenan induced rat paw edema model. Pedal inflammation was produced according to the method described by winter et al. The edema was induced by injecting 0.1ml of carrageenan (1% w/v) in normal saline into the sub-plantar region of left hind paw.[6,7]

The paw was marked with ink at the level of lateral malleolus. The paw volume was measured by Plethysmometer at 0, 30, 60, 120, 180, and 240 minutes after carrageenan injection.

The percentage inhibition of edema was calculated using the formula,

Percentage inhibition of edema = (1- Vt/Vc) ×100

Where, Vt= mean volume of paw edema in drug treated group

Vc= mean volume of paw edema in control group

Statistical analysis: The data was analysed using one-way ANOVA followed by post hoc test. P value less than 0.05 was considered significant.

RESULTS & DISCUSSION

The standard drug showed significant reduction in paw volume at 120, 180, 240 minutes. Paw volume of the rats subjected to 2% gel leaves extract of Moringa oleifera Lam. was found to be significantly reduced when compared with control at 4th hour of treatment, P= 0.04 (Table-2), the percentage reduction in paw volume at 4th hour was 21% and was comparable to the standard (0.5% Diclofenac, showed 26% reduction).

Inflammation is the protective response that involves immune cells, blood vessels & molecular mediators. The paw edema in rats, induced by carrageenan locally is inhibited by anti-inflammatory agents given by different routes having their own advantages. Carrageenan induces biphasic inflammation-early phase inflammation (due to histamine, serotonin, bradykinin) and late phase inflammation (due to prostaglandins).The late phase is inhibited by NSAIDs (Diclofenac) but not early phase7,10,12. The topical gel of leaves extract of Moringa oleifera Lam. having 2% strength showed reduction in paw volume at 240 minute reflecting the probable mechanisms of cyclooxygenase enzyme inhibition and thereby causing a production of prostaglandins. The earlier literature reported ethanolic extract of leaves extract of Moringa oleifera Lam. given orally had significant reduction of paw volume at 3 hr13.

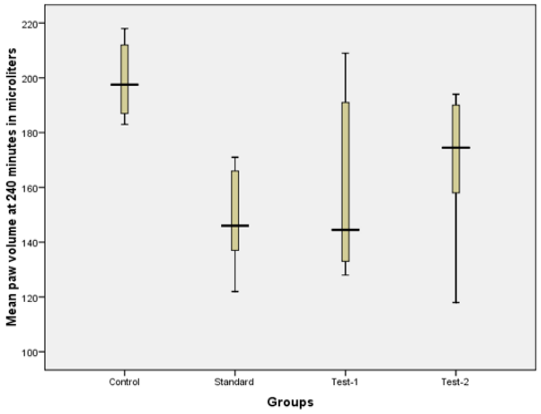

Figure 1: Difference of paw volume between the groups at 240 minutes.

Table 1: Composition of gel formulation.

Formulation	Carbopol-940(%)	Extract (%)	Propylene glycol (%)	Ethanol (%)	Methy paraben	Propyl paraben	EDTA	water
2% gel (M.O.L.E.G.)	0.5	1	2	1.5	0.1	0.01	0.015	Upto 50 ml
4% (M.O.L.E.G.)	0.5	2	2	1.5	o.1	0.01	0.015	Upto 50 ml

Table 2: Paw volume at different time intervals (Mean ± S.D.)

Groups	0 minute	30 minute	60 minute	120 minute	180 minute	240 minute
Control	146.83±10.24	166.83±6.36	194.50±13.69	236.17±29.37	211.50±24.75	199.17±13.76
Standard	123.33±12.70	155.67±21.11	199.50±16.13	195.67±26.68*	179.00±16.86*	148.00±18.25*
Test -1	157.83±14.27	182.00±10.02	199.33±14.12	209.00±17.93	186.17±19.17	158.33±34.18*
Test -2	140.50±30.23	153.00±28.40	180.83±27.89	209.00±15.20	199.67±8.82	168.17±28.66

CONCLUSION

These observations indicate that herbal gel formulation of ethanolic extract of Moringa oleifera Lam. leaves has potential topical anti- inflammatory activity on rats in carrageenan induced inflammation model. Further study is required for isolation and identification of compounds present in Moringa

ISSN (O): 2687-5365; ISSN (P): 2753-6556

oleifera Lam. responsible for topical anti-inflammatory effect and its associated adverse effects in comparison to other routes of drugs administration.

Acknowledgments: We would like to thanks Shivkumar, PhD Scholar, for the statistical analysis.

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ISSN (O): 2687-5365; ISSN (P): 2753-6556