International Journal of Basic & Clinical Pharmacology
Nachane A et al. Int J Basic Clin Pharmacol. 2025 Mar;14(2):275-280
http://www.ijbcp.com
pISSN 2319-2003 | eISSN 2279-0780

DOI: https://dx.doi.org/10.18203/2319-2003.ijbcp20250489

Original Research Article

Evaluation of the mechanism of action of *Aegle marmelos* in a murine model of 3% dextran sulphate sodium induced acute colitis

Alok Nachane[1]*, Sandhya K Kamat[2], Manoj Radhakrishnan[2], Gita Nataraj[3], Sunil S. Kuyare[3]

[1]Department of Pharmacology, MGM medical college Vashi, Navi Mumbai, Maharashtra, India
[2]Department of Pharmacology and Therapeutics, Seth G.S. Medical College and K. E. M. Hospital, Mumbai, Maharashtra, India
[3]Department of Microbiology Seth G.S. Medical College and K. E. M. Hospital, Mumbai, Maharashtra, India

Received: 22 January 2025
Accepted: 12 February 2024

***Correspondence:**
Dr. Alok Nachane,
Email: nachane_alok@mgmmcvashi.edu.in

ABSTRACT

Background: An earlier study by us in a murine model of dextran sulphate sodium (DSS) induced acute colitis showed that aqueous extract of unripe fruit of *Aegle marmelos* (780 mg/kg/day) was comparable with Sulfasalazine. In this study we evaluated the same extract for anti-inflammatory, anti-oxidant, and prebiotic activity in the same model.
Methods: 48 adult swiss albino mice (>6 weeks age) of either sex (18-25 grams) were divided into four groups (n=12/) i.e., normal control (distilled water-10 ml/kg/day), Disease control (Distilled water-10 ml/kg/day), Positive Control (Sulfasalazine-100 mg/kg/day) and Test drug (*A. marmelos*-780 mg/kg/day). The drug/vehicle was administered orally for 14 days from day 1 through day 14. Acute colitis was induced by adding 3% DSS in drinking water from day 8 to 14 in all groups except normal control. The animals were euthanized on day 15, each group were divided into two batches (n=6). One batches were used to estimate colonic myeloperoxidase (MPO) and TNF-α. The other batch was used to cultivate lactobacilli and aerobic microbiota from colonic contents, three animals from this batch were also used to estimate colonic MPO and TNF-α.
Results: Mice administered *A. marmelos*, and sulfasalazine showed significantly higher colon lengths, colon weight/ length ratios, colonic TNF-α and MPO levels, and both were significantly better than disease control. Lactobacilli and aerobic bacteria counts were significantly higher in *A. marmelos* group compared to the disease control and were comparable to normal control. However, sulfasalazine showed no improvement in the colonic microbiota counts.
Conclusions: *A. marmelos* showed anti-inflammatory, anti-oxidant, and prebiotic activity.

Keywords: Myeloperoxidase, Tumor Necrosis Factor- α, Sulfasalazine, Prebiotic, Disease activity index

INTRODUCTION

Inflammatory bowel disease (IBD), a chronic inflammatory disorder of the gastrointestinal tract that tends to relapse, affecting individuals throughout their lives.[1] IBD is categorized into two forms, ulcerative colitis (UC) and Crohn's disease (CD), and results from a combination of various factors like genetic susceptibility, dysregulated immunological response to intestinal microbiota, and environmental allergens.[2] In 2010, the estimated IBD population in India was 1.4 million. This number was the second highest in the world, with the highest prevalence in the USA (1.64 million). Although the disease prevalence in India is lesser than in the West, the disease burden is rising due to India's large population.[3] The current therapeutic approach is mostly symptomatic and not curative. The mainstay of treatment is anti-inflammatory agents such as sulfasalazine and corticosteroids, which have multiple side effects.[4] Thus,

patients of UC have a reduced quality of life from continuing disease activity and significant complications with a risk of developing colon cancer later in life, necessitating a search for newer agents. The literature of 'Ayurveda', the Indian traditional system of medicine, mentions numerous medicinal plants which are said to possess properties to strengthen the gastrointestinal system and have been recommended for use in ailments such as diarrhoea and gastritis.[5,6]

Aegle marmelos is one such plant extensively mentioned for treating gastrointestinal diseases. Its root is used in a formulation called "Dashmoolarishta," which is used as an anti-inflammatory and analgesic by Ayurvedic physicians.[5] Unripe fruits of *A. marmelos* are used as an astringent in treating diarrhoea, dysentery, and stomach ache. A literature search carried out by us revealed that various extracts of the fruit of *A. marmelos* had anti-inflammatory and anti-oxidant properties.[7,8] An earlier study carried out by us with the aqueous extract of the unripe fruit of *A. marmelos* (780 mg/kg/day) in an experimental model of DSS induced acute colitis in mice also showed *A. marmelos* to be comparable with the positive control sulfasalazine.[9] In addition, owing to the presence of various oligosaccharides in the fruit extract,[10] it was interesting to study if *A. marmelos* had any prebiotic activity. With this background, we planned a study to evaluate the aqueous extract of the unripe fruit of *A. marmelos* for its anti-inflammatory, anti-oxidant, and prebiotic activity using the same murine model of 3% DSS induced acute colitis as this model closely mimics UC. We felt that this study would shed light on whether *A. marmelos* was acting via anti-inflammatory (assessed by TNF-α levels), anti-oxidant (assessed by MPO levels), and/or prebiotic effects (assessed by lactobacilli and aerobic colony counts). This information would in turn open other avenues for its use in combination with other drugs and its possible uses in other disease conditions with similar etiology.

METHODS

Animals

The study was conducted after obtaining permission from the institutional animal ethics committee of Seth G.S. medical college and KEM hospital, Mumbai, which is registered with the committee for the control and supervision of experiments on animals (CCSEA) previously known as CPCSEA and was conducted as per the CCSEA guidelines. Forty-eight adult Swiss albino mice (>6 weeks of age) of either sex weighing 18-25 grams were procured from the Centre for Animal Studies of our institute. They were housed under standard conditions, and food and water were provided ad libitum.

Study drugs and chemicals

The disease-inducing agent, i.e., DSS (36,000-50,000 MW), was procured from MP biomedicals India Ltd. The positive control sulfasalazine was acquired from Sigma-Aldrich, Mumbai. The aqueous extract of the unripe fruits of *Aegle marmelos* (extractive value 11% w/w) was procured from Shri Dhootapapeshwar ayurvedic research foundation, Mumbai. The mouse TNF-α (Cat no-E0117Mo) and MPO (Cat no- E0436Mo) ELISA kits were acquired from bioassay technology laboratory, China.

Study groups

The study animals were divided into four groups of 12 animals each as follows: group 1-normal control, which received sterile distilled water-10 ml/kg/day; group 2-disease control which also received sterile distilled water-10 ml/kg/day, group 3 (Positive control): sulfasalazine-100 mg/kg/day, and group 4 (Test drug): A. *marmelos*-780 mg/kg/day. The dose of A. marmelos was derived based on the most effective dose found in the earlier study.[9]

Study procedure

Disease induction was carried out in all groups except normal control by giving 3% DSS in their drinking water from day 8 to 14. (9) The experimental animals were administered the study drugs or vehicle orally for 14 days from day 1 to day 14.

The body weight, stool consistency, and presence of blood in stools were assessed at baseline (day 0) and on days 10, 12, and 14 to estimate the disease activity index (DAI).[11]

All animals were euthanized by cervical dislocation on day 15 and dissected by making a midline incision on the ventral side of the animals. The colon was dissected by giving a cut at the colorectal margin and detaching it from the mesentery. The colo-caecal margin was cut, and the whole colon was removed carefully. Following this, the length and weight of each colon were measured using a Vernier caliper and a digital weighing scale, and the mean colon weight/length ratio was recorded.

In each group, colons from six mice were used to culture colonic contents. In addition, three of the six colons used for the culture of contents, and the colons of the remaining six mice (nine colons in total) were used to estimate levels of TNF-α and MPO in colonic tissue.

Estimation of disease activity index

DAI was calculated for each animal by summing up the scores for weight loss, stool consistency, and blood in stools on the respective day of measurement, as given in Table 1.[12]

The colonic contents were isolated by administering a longitudinal cut along the colon to release them. The contents were collected in a sterile container, and 50 mg of colonic contents were weighed and transferred under aseptic conditions to 5 ml buffered peptone water and sodium thioglycolate each. This was then used to culture

aerobic bacteria and lactobacilli in differing dilutions. Lactobacillus was cultured on MRS agar, while aerobic bacteria were cultured on 5% sheep blood agar.

Table 1: Disease activity index.

Score	Weight loss	Stool consistency	Blood in stool
0	None	Normal	Negative
1	1-5%	Loose stool	Negative
2	5-10%	Loose stool	Positive
3	11-15%	Loose stool	Positive
4	>15%	Diarrhoea	Gross rectal bleeding

Estimation of TNF-α and MPO

The colons (n=9) were given a longitudinal cut, and each colon was removed and placed in a petri plate on ice. The colonic contents and excess blood were removed by rinsing the colons in chilled Phosphate Buffer Solution (PBS) (pH 7.4), and the colon was homogenized in ice-cold PBS. The homogenate was centrifuged at 3000 RPM at 4ºC for 20 minutes, and the supernatant was collected in microcentrifuge tubes and stored at -80°C till further use. The levels of TNF-α and MPO were measured by using a sandwich ELISA kit for TNF-α and MPO by following the procedure mentioned in the leaflet supplied along with the kit mentioned above.

Statistical analysis

The results were expressed as mean±SD. The data were analyzed using GraphPad InStat version 3.3 (GraphPad, San Diego, CA). The intergroup analyses of parametric variables were performed using one-way ANOVA followed by post-hoc Tukey's test, whereas the Kruskal-Wallis test followed by the post-hoc Dunn's test was used for those variables which did not follow normal distribution. A $p<0.05$ was considered statistically significant.

RESULTS

Colon weight /length ratio

The mean colon weight/ length ratio of the *A. marmelos* group was not significantly different from the sulfasalazine group (Table 2).

Disease activity index

The DAI was significantly higher in the disease control group compared to normal control ($p<0.001$) on all three days. DAI was significantly lower in the sulfasalazine group on all three days compared to disease control ($p<0.001$ on Day 10, $p<0.05$ on day 12 and 14). Similarly, the DAI in the *A. marmelos* group was significantly lower than the disease control group on day 10 and 14 ($p<0.05$), but not on day 12. The mean DAI of *A. marmelos* group was not significantly different from the sulfasalazine group on all the 3 days (Table 2).

Colonic TNF-α and MPO levels

The colonic TNF-α and MPO levels were significantly higher ($p<0.001$) in the disease control group compared to normal control. The animals that were treated with sulfasalazine and *A. marmelos* and had significantly lower ($p<0.05$) levels of TNF-α and MPO as compared to disease control group. The mean levels of TNF-α and MPO observed with *A. marmelos* were not significantly different from that observed with sulfasalazine (Table 2).

Table 2: Estimation of different study variables.

Exp, groups	Colon weight/length ratio (mg/cm) (n=12)^{AT}	Disease activity index (n=12) ^{KD}			Colonic TNF- α levels (ng/gm) (n=9) ^{KD}	Colonic MPO levels (ng/gm) (n=9) ^{KD}	Microbial count (x 10^8 cfu / gm) (n=6)^{AT}	
		Day 10	Day 1 2	Day 14			Lactobacilli	Aerobic bacteria
NC	22.42±0.70	0	0	0	38.26±5.60	4.79±1.37	8.42±1.09	5.37±1.08
DC	33.65± 0.86**	3.42± 0.67*	6.75± 1.22*	9.83± 1.26*	61.67± 5.26**	22.12± 17.28**	1.87± 0.64**	1.67± 0.64 **
SS	26.68± 0.68$^{\#\#**}$	0.50± 0.90$^{\#\#}$	2.58± 1.16$^{\#\#}$	5.17± 1.58$^{*\#\#}$	45.46± 5.33$^{\#\#**}$	5.99± 1.62$^{**\#}$	2.83± 1.06**	2.08± 1.80**
AM	27.85± 0.95$^{\#\#**}$	1.33± 0.09$^{\#}$	4.0± 2.044^{*NS2}	5.92± 1.08$^{*\#}$	51.54± 4.57$^{\#**}$	6.10± 2.39$^{\#**}$	5.67± 0.74$^{\# NS1}$	4.58± 0.73$^{\#\$NS1}$

NC-Normal control, DC-disease control, SS-Sulfasalazine, AM-*Aegle marmelos*, *p<0.05,**p<0.001 vs normal control; $^{\#}$p<0.05, $^{\#\#}$p<0.001 vs disease control; $^{\$}$p<0.05 vs SLZ; NS1: Non-significant vs normal control; NS2:Non-significant vs Disease control using ANOVA followed by post-hoc Tukey-test (AT), or using Kruskal-Wallis followed by post-hoc Dunn's test (KD).

Lactobacilli and aerobic bacterial count

The lactobacilli count as assessed by colony forming units (CFU) was significantly lower ($p<0.001$) in the disease control group compared to normal control. The lactobacilli count in the sulfasalazine group was not significantly different from the disease control group. The lactobacilli count in animals treated with *A. marmelos* was significantly higher ($p<0.05$) than disease control and comparable to normal control. No statistically significant difference was found between the *A. marmelos* and sulfasalazine (Table 2).

As compared to normal control the CFU of aerobic bacteria were significantly ($p<0.001$) lower in the disease control group. The CFU of aerobic bacteria in the sulfasalazine group were comparable to the disease control group. As observed with the lactobacilli CFU, animals treated with *A. marmelos* showed a significant increase in the number of CFU of aerobic bacilli ($p<0.001$) as compared to disease control group and the counts were comparable to the normal control group. The aerobic bacteria levels were significantly higher in *A. marmelos* group compared to sulfasalazine group (Table 2).

DISCUSSION

The current therapeutic goals of inflammatory bowel disorders are induction and maintenance of remission and prevention of complications. These goals are achieved through lifestyle modification, treatment with drugs such as sulfasalazine, corticosteroids, antibiotics, immunomodulators, biologics, and surgery whenever required. The drugs used in treating IBD have multiple side effects and can adversely affect the patient's quality of life.[4] Also, biologics are prohibitively expensive, especially in a country like India, where medical expenses are often out of pocket. Ayurvedic literature mentions many drugs for the treatment of gastrointestinal disorders. These drugs have been used for centuries, and no serious side effects have been reported with any of these drugs. One such agent, *A. marmelos*, is used by Ayurveda physicians to treat diarrhoea, dysentery, and abdominal pain. *Ayurvedic* texts mention it as an astringent with detoxifying and regulatory effects on the bowel.[5]

Various researchers have demonstrated the anti-inflammatory potential of different parts of the plant *A. marmelos.* in animal models of acute inflammation.[7,13-15] However, the effect of aqueous extract of the unripe fruit of *A. marmelos* in IBD was evaluated in very few studies.[9,16,17] Of these, only the study carried out by us has evaluated the effect of *A. marmelos* in an experimental model of DSS induced acute colitis and compared its effects with sulfasalazine.[9] In this study, we observed that *A. marmelos* significantly ameliorated DSS induced acute colitis in mice at 780 mg/kg/day.[9] Hence, we chose the same dose of *Aegle marmelos* in the current study. We used sulfasalazine as the positive control as recent guidelines recommend it as the first line of treatment in cases of acute colitis. Moreover, sulfasalazine acts by suppressing inflammation and reduces the levels of TNF-α.[18-20] We selected murine models for our studies as the acute colitis model using DSS is a well-standardized model in mice and has been used by us in the past to assess the effect of various drugs.[9,16] Also, the gut microbiome of the Swiss albino mice and human gut microbiome are functionally comparable, with 95.2% of its Kyoto Encyclopaedia of genes and genomes (KEGG) orthologous groups in common.[21]

As observed in our earlier study, the group which received *A. marmelos* showed comparable findings to sulfasalazine with respect to colon weight/length ratio and DAI, and both were significantly better than the disease control group.

It is well known that TNF- α is involved in initiating and continuing inflammatory processes.[22] It has been observed that its levels are chronically elevated both locally and systemically among IBD patients.[23] In this study, the colonic levels of TNF-α were decreased in the *A. marmelos* group and were comparable to sulfasalazine. Kasinathan et al have also reported a decrease in the expression of TNF-α among the animals treated by *A. marmelos* in DSS induced colitis; however, unlike in our study, there was no positive control in their study.[17] *In vitro* studies have also reported lowering of TNF-α expression levels in response to marmelosin derived from ethyl acetate extract of the fruit of *A. marmelos*.[8] Rathee et al observed that 70% ethanolic extract of *A. marmelos* leaves lowered TNF-α levels in a model of paracetamol-induced hepatic injury in wistar rats.[24]

An important chain of events in IBD is oxidative stress. MPO released by leukocytes plays a crucial role in inflammation and oxidative stress at the cellular level.[25] In this study, the colonic levels of MPO were decreased in the group administered *A. marmelos* to a level comparable to sulfasalazine. Ghatule et al and Gautam et al have also reported a reduction in MPO levels in their study as compared to the disease control group; however, as mentioned earlier, they did not compare their results to sulfasalazine.[26,27]

Treatment with both probiotics and prebiotics has been tried in IBD. Various studies have been performed to examine the effect of probiotics, such as *Lactobacillus*, *Bifidobacterium, and Enterococcus*, in treating IBD. Unfortunately, studies with probiotics have shown inconsistent results. Some studies have found that probiotics can restore the function of the disturbed mucosal barrier, correct intestinal microbiota imbalance, competitively inhibit the growth of potential pathogens, improve systemic and local immunity, and enhance intestinal barrier function. Others have shown they promote gastrointestinal peristalsis, and alter the frequency of stool, leading to diarrhoea, and no improvement in disease activity.[28] A study evaluating the effect of prebiotics in IBD demonstrated that fructooligosaccharides (FOS) and lactulose lead to worsening or no significant improvement among CD patients, respectively. However, it has shown that FOS and other prebiotics, such as germinated barley foodstuff and ispaghula husk, may benefit both active and inactive UC cases.[29] To our knowledge, no study has assessed prebiotic activity effect of *A. marmelos* singly or in combination with probiotics *in vivo*. In our study, lactobacilli and aerobic bacteria CFU counts were higher in the *A. marmelos group* than in disease control group. Also, these counts were better than positive control sulfasalazine and comparable to the normal control group signifying restoration of gut microbiome to near-normal levels.

A limitation of this study was that of the various cytokines involved in UC we studied only the levels of TNF-α which were decreased by *A. marmelos*. Further studies in which the entire cytokine panel is studied are necessary to understand better how *A. marmelos* exerts its action. Also, our findings need to be validated in other animal models of IBD. In addition, clinical trials to assess if *A. marmelos* can induce clinical remissions in UC patients can provide useful information that will aid in developing a safe and effective treatment for UC in humans.

CONCLUSION

UC is still not adequately managed necessitating study of new treatments. In the current study, the DAI, was comparable in animals treated with *A. marmelos* and sulfasalazine. *A. marmelos* also reduced the colonic TNF-α and MPO levels significantly as compared to the disease control group and comparable to levels seen in normal control group thus demonstrating anti-inflammatory and antioxidant property. Also, it was superior to sulfasalazine in reinstating the normal gut microbiome which plays a vital role in the disease pathophysiology. Thus, *A. marmelos* can prove to be a valuable addition to the current treatment of ulcerative colitis.

ACKNOWLEDGEMENTS

Authors would like to thank Dhootapapeshwar Ayurvedic research foundation and research society of Seth G. S. medical college and KEM hospital also, to department of health research, government of India.

Funding: Funding sources Research society of Seth GS Medical College and KEM hospital and the Seth G S medical college multidisciplinary research unit
Conflict of interest: None declared
Ethical approval: The study was approved by the Institutional Ethics Committee

REFERENCES

1. Singh P, Ananthakrishnan A, Ahuja V. Pivot to Asia: inflammatory bowel disease burden. Intest Res. 2017;15(1):138.
2. Ng WK, Wong SH, Ng SC. Changing epidemiological trends of inflammatory bowel disease in Asia. Intest Res. 2016;14(2):111.
3. Kedia S, Ahuja V. Epidemiology of Inflammatory Bowel Disease in India: The Great Shift East. Inflamm Intest Dis. 2017;2(2):102-15.
4. Carter MJ, Lobo AJ, Travis SP. Guidelines for the management of inflammatory bowel disease in adults. Gut. 2004;53(5):v1-6.
5. Gogaṭe VM. Ayurvedic pharmacology and therapeutic uses of medicinal plants (Dravyagunavignyan). Bharatiya Vidya Bhavan; 2000;841.
6. Sharma PPV. Dravyaguna-Vijnana. 4th ed. Varanasi: Chaukhambha Sanskrit Sansthan. Varanasi. 1978;455-7.
7. Kamat SK, Singh KNM. Evaluation of the effect of *Aegle marmelos* and *Punica granatum* in a murine model of dextran sulfate sodium-induced acute colitis. National J Physiol Pharmacy Pharmacol. 2019;9(4):1-8.
8. Charoensiddhi S, Anprung P. Characterization of bael fruit (*Aegle marmelos* [L.] correa) hydrolysate as affected by enzyme treatment. J Food Biochem. 2010;34(6):1249-67.
9. Park YH, Kim N, Shim YK, Choi YJ, Nam RH, Choi YJ, et al. Adequate dextran sodium sulfate-induced colitis model in mice and effective outcome measurement method. J Cancer Prevention. 2015;20(4):260-7.
10. Arul V, Miyazaki S, Dhananjayan R. Studies on the anti-inflammatory, antipyretic and analgesic properties of the leaves of *Aegle marmelos* Corr. J Ethnopharmacol. 2005;96(1-2):159-63.
11. Benni JM, Jayanthi MK, Suresha RN. Evaluation of the anti-inflammatory activity of *Aegle marmelos* (Bilwa) root. Indian J Pharmacol. 2011;43(4):393-7.
12. Hidalgo-Cantabrana C, Algieri F, Rodriguez-Nogales A. Effect of a ropy Exopolysaccharide-producing *Bifidobacterium animalis* subsp. Lactis strain orally administered on DSS-induced colitis mice model. Front Microbiol. 2016:7:868.
13. Rajaram A, Vanaja GR, Vyakaranam P, Rachamallu A, Reddy GV, Anilkumar K, et al. Anti-inflammatory profile of *Aegle marmelos* (L) Correa (Bilva) with special reference to young roots grown in different parts of India. J Ayur integrative Med. 2018;9(2):90-8.
14. Ibrahim M, Parveen B, Zahiruddin S, Gautam G, Parveen R, Khan MA, et al. Analysis of polyphenols in *Aegle marmelos* leaf and ameliorative efficacy against diabetic mice through restoration of antioxidant and anti-inflammatory status. J Food Biochemistr. 2021;46(4):e13852.
15. Kasinathan NK, Subramaniya BR, Pandian I, Sivasithamparam ND. *Aegle marmelos* fruit extract abates dextran sodium sulfate induced acute colitis in mice: Repression of pro-inflammatory cytokines during colonic inflammation. Biomed Prev Nutr. 2014;4(2):307-17.
16. Behera JP, Mohanty B, Ramani YR, Rath B, Pradhan S. Effect of aqueous extract of *Aegle marmelos* unripe fruit on inflammatory bowel disease. Indian J Pharmacol. 2012;44(5):614-8.
17. Gibson PR, Jewell DP. Sulphasalazine and derivatives, natural killer activity and ulcerative colitis. Clinical Science. 1985;69(2):177-84.
18. Kang BY, Chung SW, Im SY, Choe YK, Kim TS. Sulfasalazine prevents T-helper 1 immune response by suppressing interleukin-12 production in macrophages. Immunology. 1999;98(1):98-103.
19. Rodenburg RJ, Ganga A, Van Lent PL, Van De Putte LB, Van Venrooij WJ. The anti-inflammatory drug sulfasalazine inhibits tumor necrosis factor α expression in macrophages by inducing apoptosis.

Arthrit Rheumat. 2000;43(9):1941-50.
20. Xiao L, Feng Q, Liang S, Sonne SB, Xia Z, Qiu X, et al. A catalog of the mouse gut metagenome. Nat Biotechnol. 2015;33(10):1103-8.
21. Naito Y, Takagi T, Handa O, Ishikawa T, Nakagawa S, Yamaguchi T, et al. Enhanced intestinal inflammation induced by dextran sulfate sodium in tumor necrosis factor-alpha deficient mice. J Gastroenterol Hepatol. 2003;18(5):560-9.
22. Jones-Hall YL, Nakatsu CH. The Intersection of TNF, IBD and the Microbiome. Gut Microbes. 2016;7(1):58-62.
23. Pynam H, Dharmesh SM. Antioxidant and anti-inflammatory properties of marmelosin from Bael (*Aegle marmelos* L.); Inhibition of TNF-α mediated inflammatory/tumor markers. Biomed Pharmacother. 2018;106:98-108.
24. Rathee D, Kamboj A, Sachdev RK, Sidhu S. Hepatoprotective effect of *Aegle marmelos* augmented with piperine co-administration in paracetamol model. Rev Bras Farmacogn. 2018;28:65-72.
25. Anatoliotakis N, Deftereos S, Bouras G, Giannopoulos G, Tsounis D, Angelidis C, et al. Myeloperoxidase: expressing inflammation and oxidative stress in cardiovascular disease. Curr Top Med Chem. 2013;13(2):115-38.
26. Ghatule RR, Gautam MK, Goel S, Singh A, Joshi VK, Goel RK. Protective effects of *Aegle marmelos* fruit pulp on 2,4,6-trinitrobenzene sulfonic acid-induced experimental colitis. Pharmacogn Mag. 2014;10(1):S147-52.
27. Gautam MK, Ghatule RR, Singh A, Purohit V, Gangwar M, Kumar M, et al. Healing effects of *Aegle marmelos* (L.) Correa fruit extract on experimental colitis. Indian J Exp Biol. 2013;51(2):157-64.
28. Shen Z-H, Zhu C-X, Quan Y-S, Yang Z-Y, Wu S, Luo W-W, et al. Relationship between intestinal microbiota and ulcerative colitis: Mechanisms and clinical application of probiotics and fecal microbiota transplantation. World J Gastroenterol. 2018;24(1):5-14.
29. Martyniak A, Medyńska-Przęczek A, Wędrychowicz A, Skoczeń S, Tomasik PJ. Prebiotics, probiotics, synbiotics, paraprobiotics and postbiotic compounds in IBD. Biomolecules. 2021;11(12):1903.

Cite this article as: Nachane A, Kamat SK, Radhakrishnan M, Nataraj G, Kuyare SS. Evaluation of the mechanism of action of *Aegle marmelos* in a murine model of 3% dextran sulphate sodium induced acute colitis. Int J Basic Clin Pharmacol 2025;14:275-80.