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Anti-inflammatory and Anti-arthritic Potentials of Maesa Montana Roots

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Abstract

Maesa Montana is a flowering plant of the Myrsinaceae family, which is locally known as ramjani. The current study aimed to evaluate the *in-vitro* anti-arthritic and anti-inflammatory activities of the root extract of this plant. The anti-inflammatory performances were measured by the hindrance of egg white denaturation, and anti-arthritic activity was investigated by Bovine serum protein. In the anti-inflammatory activity test, the methanolic root extract of this plant showed 69.29 ± 1.19 % of inhibition at a concentration of 1000 µg/mL and standard drug exhibited 90.11 ± 1.45 % of inhibition at the same concentration. Furthermore, in the anti-arthritic activity test, the extract demonstrated 68.18 ± 1.34 % of inhibition at a concentration of 1000 µg/mL whereas the standard diclofenac drug showed 90.65 ± 1.19 % of inhibition at the same concentration. These results revealed that the root extract of this plant possesses significant anti-arthritic and anti-inflammatory activities based on the inhibition of BSA and protein denaturation.

Keywords: Maesa Montana, Anti-arthritic, Anti-inflammatory, Protein denaturation

HIGHLIGHTS

Introduction

The tradition of using plants to treat human diseases is as old as the evolution of human civilization itself. From the beginning of human civilization, medicinal plants have been playing an important role in the human welfare [1]. A medicinal plant can possibly be defined as the commonly used plants for treating and preventing different types of ailments and disorders. Plants, employing favorable pharmacological impacts on physical body are usually titled as "Medicinal plants." It can now be reached to the conclusion that plants, which normally integrate and possess some secondary metabolites, similar to tannins, alkaloids, glycosides and comprise minerals as well as vitamins, have therapeutic properties [2].

The popularity of the traditional medicine has been increasing in the developing countries as well as in the developed countries due to the fact that modern medicine failed to provide successful treatment for different chronic diseases, where there is an advent of the resistant variants of bacteria and parasites. The adverse side effects of synthetic medicines and their rising costs have also led to an increased interest on alternative treatments among the people [3].

Inflammation is a natural response of the body from an infection or damage of the tissue, which is manifested by hot, swollen pain and redness of the area adjacent inflammation. The causative agents of inflammation include toxic compounds or infection due to microbial agents. The defense mechanism of the body is to try to inactivate or kill the invading pathogen to eliminate the irritating substrates and repair

the damaged tissues [4]. Non-steroidal anti-inflammatory drugs are commonly used to treat inflammations, but they have different unfavorable effects, including gastric irritation that may lead to gastric ulcers. As the plant kingdom serves as a source of discovering new compounds, some of them may have significant anti-inflammatory effects without major side effects [5].

The term arthritis means any disorder that especially affects the joints. Joint pain, swelling, stiffness, inability to move freely are some of the most common arthritis symptoms [6,7]. The symptomatic relief of this problem is generally achieved by using non-steroidal anti-inflammatory drugs or anti-rheumatic agents. Plant extracts were traditionally used for reducing arthritic pain [8].

Maesa Montana is a big shrub, having a height of 2 - 3 m. The leaves are elliptic, rectangle or spear shaped and in rare cases can be roughly ellipsoid. Flowers are generally of white color, bell-shaped, ellipsoid petals, longer than a tube, with an entire border or toothed, rounded tip. The blooming period of this plant is from February to April [9,10].

Materials and methods

Plant material collection

The fresh roots of the plant *M. Montana* were collected in the month of January 2019 from the division of Chittagong, Bangladesh. After collecting, a professor of botany department of Chittagong University named Dr. Sheikh Bakhtiar Uddin identified the roots.

Plant extraction procedure

Plant materials were cut into small pieces for grinding. The small pieces of roots were ground into a coarse powder using a grinder. About 500 gm of the powder was soaked in a cleaned amber glass reagent bottle (5 L) using 1.6 L of methanol. The bottle was airtight and kept for 7 days at the optimum temperature with shaking at a regular interval and then filtered firstly through a cotton plug and then through a filter paper (pore size 110 mm). The solvent was evaporated with a water bath to yield a solid mass. The bulk of the extract collected was 23 gm. The prepared extract was kept in the refrigerator to prevent microbial contamination.

Anti-Inflammatory activity test

The anti-inflammatory test was performed following the technique described by Sakat *et al.* [11]; Leelaprakash and Dass [12]. The egg was made to use for the source of albumin and was reconstituted as a 5 % v/v aqueous solution with iso-saline. To prepare the sample solution, 1 mg root extract was mixed with 10 mL methanol. For preparing the standard solution, 10 mg diclofenac sodium was taken and mixed with 4 mL methanol.

For the preparation of test solution, egg albumin (0.2 mL), phosphate buffered having a pH of 6.4 (2.8 mL) and varying concentrations of the test extract (2 mL) was taken in different test tubes. The standard solution was prepared in a similar manner using diclofenac sodium instead of the extract. Same volume of distilled water was used as a control. Then, the all the test tubes were incubated at around 37 °C in an incubator for a period of 15 min. Then, it was heated for 5 min at a temperature of 70 °C. After cooling, the absorbance of the test tubes were measured at 660 nm against the blank [13]. The percentage inhibition of protein denaturation was calculated by using the following formula:

% inhibition = $[Vc - Vt] Vc \times 100 \%$

where, Vt = Absorbance of test sample, Vc = Absorbance of control. Fifty Percent of albumin denaturation of the extract was determined from the dose-response curve.

Anti-arthritic activity

Protein denaturation inhibition by using BSA was determined following the procedure narrated by Pavithra *et al.* [9]; Kumari *et al.* [14].

5 % w/v BSA solution was made by dissolving 5 gm BSA in 100 mL distilled water. In the case of preparing the phosphate buffer, 0.45 gm NaH₂PO₄ and 0.887 gm Na₂HPO₄ were taken in a beaker. Then up to 100 mL distilled water was added to prepare the solution. The pH 6.3 was adjusted by using 1 N HCl. Here, 10 mg of the extract was liquefied in 10 mL of distilled water to prepare a stock solution with 1 mg/mL concentration. Then, the test solutions were prepared by diluting the stock solutions (1000, 500, 250, 125, 62.5 and 31.75 ug/mL). The standard solution was prepared in a similar manner using the diclofenac sodium. The test control solution was prepared by dissolving 0.2 mL of BSA and 2 mL of distilled water in 2.8 mL of phosphate buffer. The test solution was prepared using 2 mL of extract or standard solution of varying concentration, 2.8 mL of phosphate buffer and 0.2 mL BSA solution, then the product control was prepared by dissolving 2 mL of extract or standard solution of varying concentration, 2.8 mL of distilled water. All the test tubes mixtures were kept at 37 °C for around 30 min and then again incubated at 57 °C for 5 min. The absorbance was taken at 660 nm. The following equation can determine the anti-arthritic activity of the plant extract:

 $= 100 - \frac{\textit{Absorbaceoftestsolution} - \textit{Absorbanceofproductcontrol}}{\textit{Absorbanceofcontrol}} \times 100 \%.$

Results and discussion

In the current study, the methanolic extract of *M. Montana* showed mean inhibition of protein denaturation of around 42.72, 48.82, 56.35, 66.52, 79.76, 90.18 % for the doses of 78.115, 156.25, 312.5, 625, 1250 and 2500 μ g/mL, respectively whereas for the standard diclofenac sodium, it was found around 38.94, 46.11, 53.64, 58.88, 62.56 and 69.29 % for the same doses respectively (**Table 1** and **Figure 1**). Here, the IC₅₀ value of the standard was 64.35 and for the sample, it was 351.90. The anti-inflammatory assay was performed by using egg albumin. From this experiment, it was noticed that the root extract had a moderate anti-inflammatory effect.

Concentration	Sample	Standard
	% inhibition	% inhibition
78.115	38.35±1.09	42.72±1.67
156.25	46.35±1.11	48.82 ± 1.02
312.50	53.64±0.79	56.35±1.36
625	58.88±0.48	66.52±1.06
1250	62.56±1.03	79.76 ± 0.58
2500	69.29±1.19	90.11±1.45

 Table 1 Anti-inflammatory results of the methanolic extract of M. Montana and standard diclofenac powder.



Figure 1 Percent of protein denaturation inhibition of *M. Montana* root extract and diclofenac sodium (standard).

The extract showed significant anti-arthritic activities with 20.77, 29.78, 37.87, 51.51, 56.06 and 68.18 % of inhibition according to the concentration of 31.75, 62.75, 125, 250, 500 and 1000 μ g/mL; where the standard drug showed 38.33, 50.44, 58.09, 70.87, 80.56 and 90.65 % of inhibition with their belonging concentrations (**Table 2** and **Figure 2**). The anti-arthritis properties of these extracts indicated the possibility of the presence of alkaloids, flavonoids and steroid compounds in the methanolic extract [15]. Though it had a moderate activity, it could be a possible source of anti-arthritic drugs from further research studies.

Concentration (µg/mL)	Sample	Standard
	% inhibition	% inhibition
31.75	20.77±1.19	38.33±1.22
62.75	29.78 ± 0.67	$50.44{\pm}1.88$
125	37.87±1.08	58.09±1.67
250	51.51±1.20	70.87±1.12
500	56.06 ± 0.98	80.56 ± 0.56
1000	68.18±1.34	90.65±1.19

Table 2 Anti-arthritic result of methanolic extract of M. Montana.

http://wjst.wu.ac.th



Figure 2 Percent of BSA denaturation inhibition of M. Montana root extract and diclofenac sodium (standard).

Conclusions

It can be concluded that this well-acquainted study appraised moderate in vitro anti-arthritic and anti-inflammatory features of the methanolic extract of M. Montana at different doses. However, all pharmacological profiles have not been investigated for this plant. Further investigations using advanced techniques and animal models could be attained through careful and systemic schemes.

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