Analgesic Activity of Methanolic Extract of the Leaf of *Erythrina variegata*

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Erythrina variegata (Synonym: Erythrina indica Lamk.; Bengali name- Mandar) is a medium sized deciduous small tree belonging to the family Papilionaceae. The plant grows all over the Bangladesh.¹ The barks are used traditionally as astringent, febrifuge and in leprosy and fever. Leaves are anthelmintic, laxative and diuretic. Paste of leaves is applied externally to cure inflammations and to relieve pain in the joints. Juice is also used to relieve earache and toothache.¹ Previous phytochemical investigation showed that the stem bark contains three new isoflavones and a new isoflavanone.² Seed contains a fixed oil, fatty acids and lectins.¹ Though the plant is traditionally used in many parts of Bangladesh, no scientific report is available to validate the folkloric use. As a part of our continuing studies on the medicinal plants of Bangladesh,³⁻⁵ the study was undertaken to evaluate the analgesic activity using acetic acid induced writhing and radiant heat tail-flick test in mice models.

The leaf of the plant *E. variegata* was collected from Dhaka, Bangladesh in August 2004, and was identified by the experts at the Department of Botany, University of Dhaka. Collected plant parts, after cutting into small pieces, were dried and pulverized into a coarse powder, and stored into an air-tight container.

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The pulverized coarse powder (250 g) was extracted with methanol (750 ml \times 3) by cold extraction process. The extract obtained was filtered off and evaporated to dryness by rotary evaporator to get the methanolic extract (4.44 g) of *E. variegata* (EVM). Finally the extract was defatted by refrigeration at 4° C temperature.

Swiss albino mice (20-25 g) of either sex were obtained from the animal house of International Center for Diarrhoeal Disease and Research, Bangladesh (ICDDR, B). The animals were given standard feed developed by ICDDR,B and water *ad libitum* and kept in the laboratory environment (12 h dark/12 h light cycle) for seven days for acclimatization. Animals were kept under fasting for overnight and weighed before the experiment.

For the preparation of test materials 125 and 62.5 mg of the extracts were separately triturated by the addition of small amount of Tween-80. After proper mixing, saline water was slowly added and the final volume of the suspension of each extract was adjusted to 2.5 ml. For the preparation of standard, 12.5 mg of aminopyrine was taken and suspension of 2.5 ml was made with Tween-80 and saline water. Each 10 gm of experimental animal received 0.1 ml of the crude extract and the standard drug.

The peripheral analgesic activity of methanolic extract of leaf of *E. variegata* (EVM) was determined by the acetic acid induced writhing inhibition method.⁶ The inhibition of writhing in mice by the plant extract was compared against inhibition of writhing by a standard analgesic agent, aminopyrine given orally at a dose of 50 mg/kg body weight. The

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number of writhing was calculated for 10 min 5 minutes after the acetic acid injection. The percentage of pain protection was calculated.

The analgesic activity was determined by radiant heat tail-flick model in mice.⁷ Morphine was used as the standard analgesic agent. Tail-flick latency was assessed by the analgesiometer (Inco, India). Tailflick latency was measured 60 minutes after the drug administration.

The results were analyzed for statistical significance using one-way ANOVA followed by Dunnet's test. A P value <0.05 was considered significant.



Fig 1. Comparative study of the analgesic activity by acetic acid induced writhing method.

The abdominal constriction response induced by acetic acid is a sensitive procedure to establish peripherally acting analgesics. The response is thought to involve local peritoneal cells and is mediated by the prostaglandin pathways.⁸ The significant antinociceptive activity of root of EVM might be due to the presence of analgesic principles acting with the prostaglandin pathways. However, true analgesic activity can only be ensured by the combination of at least two methods as the acetic acid induced abdominal constriction test can provide false positive results.9 To investigate whether methanolic root extract of E. variegata has true analgesic potential, radiant heat tail-flick method was also used. As the crude extract appeared to be active in both animal models of nociception, it may possess peripherally and centrally acting compounds for its antinociceptive action.

The results of the animal experiments are shown in figures 1 and 2. In acetic acid induced writhing model the methanolic extract of the leaf of *E. variegata* at a dose of 500 mg/kg showed significant antinociceptive activity with 49.03% inhibition of writhing response. The results were statistically significant (P < 0.01) in comparison to the control. The antinociceptive activity of the extract at a dose of 250 mg/ kg body weight was not significant. In radiant heat tail-flick model, the extract also showed significant increase in the tail flick latency at a dose of 500 mg/kg body weight with 36.02% elongation of tail flick time (Figure 2).



Fig 2. Comparative study of the analgesic activity by radiant heat tail flick method.

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