

# Anti-Inflammatory Activity of Ethnic Vegetables Osonshak (*Spilanthes calva*) and Chikipung (*Rumex vesicarius*) in Animal Model

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## Abstract

The aqueous leaf extract of ethnic vegetables Osonshak (*Spilanthes calva*), Chikipung (*Rumex vesicarius*) and Kochi ampata (*Mangifera indica*) widely consumed by the ethnic people of Chittagong Hill Tracts was investigated for their anti-inflammatory activity. Carrageenan induced rat paw edema model was employed to test the anti-inflammatory property. The plant extract (500 mg/kg body weight) was given orally to the rats. Diclofenac sodium (50 mg/kg) was used as the positive control. Osonshak and Chikipung showed significant anti-inflammatory activity.

#### **Keywords**

Osonshak, Chikipung, Leaf Extract, Antiinflammation, Rat Model

## **1. Introduction**

Ethnic edible plants are nutritionally rich [1]. These plants are growing wildly and consumed widely by the ethnic people. They contain phytochemicals including alkaloids, flavonoids, carotenoids, tannins and phenolic compounds and minerals and vitamins [1], and have been reported effective, traditionally in the treatment of many illnesses including oxidative stress, diabetes, CVS, degeneration, immune disorder, inflammation etc. [2] [3] [4]. In search of potential therapeutic agents, it is of outmost importance of screening medicinal plants for their biological activities and phytochemicals.

*Spilanthes calva* DC is an annual, spreading plant with bicolored, red or gold flower buds. It is an important medicinal herb. Its roots and flower heads are employed to relieve toothache, pyorrhea, infections of throat and gums. The

roots, flower heads and whole aerial part of it yield a compound known as spilanthol, which is a bioactive compound and has biological and pharmacological effects including analgesic, neuroprotective, antioxidant, immune enhancer, anti-cancer and anti-inflammatory [2]. *Rumex vesicarius* Linn is a branched succulent edible herb and contains important bioactive flavonoids, anthraquinones [5]. It is used to cure several diseases with CVDs, respiratory, hepatic disorder [3] [5]. *Mangifera indica* Linn is one of the most popular of all tropical fruits. It contains several phytochemicals mangiferin, catechins, quercetin, kaempferol, rhamnetin, anthocyanins, gallic and ellagic acids, propyl and methyl gallate, benzoic acid, and protocatechuic acid and glucosyl xanthone [6] [7] [8]. Mangiferin is a special polyphenol in the mango, which has demonstrated potential to combat degenerative diseases like heart diseases and cancer [8].

The present work aimed to investigate the anti-inflammatory activity of the ethnic vegetable Osonshak (*Spilanthes calva*), Chikipung (*Rumex vesicarius*) and Kochi Aampata (*Mangifera indica*) in carrageenan induced paw edema in rat model.

## 2. Materials and Method

#### **2.1. Ethnic Plants**

The plant samples were selected based on their medicinal importance in literature and information on their traditional uses in different ailments by the local people living in the Chittagong Hill Tracts (CHTs) through Focus Group Discusson. The vegetables were collected from local weekly markets at Bandarban, Rangamati and Khagrachari districts with the help of personnel of the Department of Agricultural Extension (DAE) there, and were then authenticated and certified by a taxonomist of the Department of Botany, University of Dhaka. Attempted was taken to collect for every sample from each of the markets or at least triplicates for every item, which was then mixed to make three analytes or composite test samples [9].

#### 2.2. Preparation of Plant Extract

Aqueous extraction was made to obtain water-soluble substances from plant materials. Vegetable samples were washed with tap water, rinsed with distilled water, whipped out extra water using tissue paper, then air-dried, and finally cut into small pieces. About 500 g of processed vegetable was blended in a blender and smashed in mortar with a pastel using a small amount of distilled water. The extract was filtered first through a muslin cloth and then re-filtered by filter paper (Whatman no 1), the total extract yield was approximately 350 ml. It was aliquoted into screw cap vials and stored in a refrigerator for analysis. Content of extract was measured by specific gravity as 1.15 g/ml.

#### 2.3. Animal Experimentation

Wistar Albino rats (80 g - 120 g) of either sex were procured from the animal

house of Jahangir Nagar University, Savar, Bangladesh. The rats were housed in metallic cages in a temperature-controlled room  $(26^{\circ}C - 28^{\circ}C)$  with light and dark cycle in well ventilated and sanitized animal experiment room at the INFS for 7 days. The animals were fed on standard laboratory animal diet formulated by ICDDR, B and water *ad libitum*. In order to keep the hydration rate constant, food and water were stopped 12 hours before the experiments. Experiments on animals were performed strictly following the guidelines provided by the Institutional Animal Ethics Committee.

#### 2.4. Carrageenan-Induced Rat Paw Edema

The anti-inflammatory activity of the extracts was evaluated by a carrageenan-induced edema model in Wistar adult rats [10]. The initial right hind paw volume of the rats was measured using a Plethysmometer. The Wistar rats were randomly divided into 5 groups of 5 rats each.

The group 1, 2 and 3 rats were feed with the extract of Osonhak (*Spilanthes calva*), Chikipung (*Rumex vesicarius*) and Kochi ampata (*Mangifera indica*) orally. Group 4 received diclofenac sodium (10 mg/kg orally), and the group 5 (control group) received normal saline. After 30 minutes, 100  $\mu$ l of 1% (w/v) carrageenan ((Sigma Chemical Company, St. Louis, MO, USA) in normal saline was injected subcutaneously to each of the rats in all groups into the sub plantar region of the right hind paw. The edema volume in the right hind paw of every rat in the groups was measured with the Plethysmometer at 0, 1, 2, 3 and 4 h after the carrageenan injection.

The volume of the paw edema was evaluated for anti-inflammatory activity and it was expressed as % inhibition or decreasing of the edema volume.

Inhibition or decreasing 
$$(\%) = \frac{Vc - Vt}{Vc} \times 100$$

where Vc = Average paw volume of the control group, Vt = Average paw volume of the treatment groups.

#### 2.5. Statistics

The data obtained were expressed as mean  $\pm$  SEM. Statistical differences between groups were calculated by use of one-way analysis of variance (one-way ANOVA) following by Dunnett's test. P values less than 0.05 (P < 0.05) were used as the significance level.

#### 3. Results and Discussion

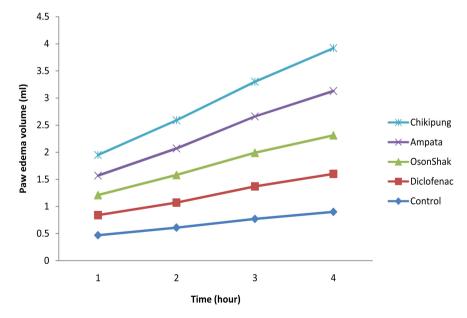
**Table 1** describes the anti-inflammatory effect of ethnic vegetables on the carrageenan-induced rat paw edema, which is a suitable model to assess acute inflammation. The development of edema has been described as a biphasic event in which various mediators operate in sequence to produce the inflammatory response. The initial phase of edema (0 - 1 h) is attributed to the release of histamine, 5-hydroxytryptamine (5-HT) and bradykinin [11] and the second acce-

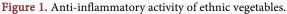
lerating phase of swelling (1 - 6 h) is correlated with elevated production of prostaglandins and more recently it has been attributed the release of cyclo-oxygenase in the paw. In the present experiment, Osonshak extract showed significant inhibition of paw edema in both the phases of edema, inhibition was found to be  $0.71 \pm 0.03$ , which was equivalent to 20.43% inhibition at 4 hours. The inhibitory mechanism of inflammation is not clear. It may be due inhibition of release and/or action of inflammatory mediators those induced edema. Cyc-looxygenase synthesizes prostaglandins that lead to acute inflammation [12]. Oson shak might have inhibited the synthesis of cyclo-oxygenase. The study finding is supported by others for the similar plant [13] [14]. The present findings suggest that these ethnic plants specially Osonshak suppresses carragee-nan-induced paw edema, thus, it can be a promising alternative to nonsteroidal antiinflammatory drugs (NSAID) (Figure 1).

 Table 1. Anti-inflammatory effect of ethnic vegetables on carrageenan-induced paw edema in rats.

| Ethnic vegetable                     | Animal<br>group | Oral dose<br>mg/kg | Paw edema volume (ml) |                      |                      |                      | % inhibition<br>at 4 hour |
|--------------------------------------|-----------------|--------------------|-----------------------|----------------------|----------------------|----------------------|---------------------------|
|                                      |                 |                    | 1 hour                | 2 hour               | 3 hour               | 4 hour               |                           |
| OsonShak<br>Spilanthes calva         | 1               | 500                | 0.37 ± 0.01**         | 0.51 ± 0.03*         | 0.62 ± 0.02*         | 0.71 ± 0.03*         | 20.43                     |
| Chikipung<br><i>Rumex vesicarius</i> | 2               | 500                | 0.38 ± 0.02**         | $0.52 \pm 0.03$      | 0.64 ± 0.03*         | $0.79\pm0.04$        | 10.2                      |
| Kochi ampata<br>Mangifera indica     | 3               | 500                | 0.36 ± 0.01**         | 0.49 ± 0.01**        | $0.67 \pm 0.03$      | $0.82 \pm 0.03$      | 8.88                      |
| Diclofenac sodium                    | 4               | 10                 | $0.37 \pm 0.01^{**}$  | $0.46 \pm 0.02^{**}$ | $0.60 \pm 0.01^{**}$ | $0.70 \pm 0.01^{**}$ | 23.66                     |
| Control                              | 5               | Normal saline      | $0.47\pm0.02$         | $0.61 \pm 0.01$      | $0.77\pm0.02$        | $0.90\pm0.03$        | -                         |

Edema volume: Mean ± SEM. \*p < 0.05, \*\*p < 0.01 as compared to control group.





### 4. Conclusion

The aqueous extracts of the ethnic Osonshak indicated a significant anti-inflammatory activity, which could be used in the treatment of inflammation. These observations enhance potential interest for improving the efficacy of the ethnic vegetables as neutraceuticals and pharmacological products. The consumption of these plants may play a role in preventing human diseases such as cancer, cardiovascular diseases and aging. However, further research work is required to segregate the active constituents from the extract exhibiting significant anti-inflammatory and the research regarding the mechanism responsible for this activity is also required which will guarantee its clinical worth.

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## **Conflicts of Interest**

The authors declare no conflicts of interest regarding the publication of this article.

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