Antinociceptive, anti-inflammatory and antiarthritic activities of
*Bungarus fasciatus* venom in experimental animal models

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Pain and inflammation are intimately associated with rheumatoid arthritis, a growing bone-joint related problem of the modern society. Though several therapeutic managements are available for arthritis, their side effects not only limit their use, but also advocate the quest for natural therapies. In this study, we explored the antinociceptive, anti-inflammatory and antiarthritic activities of *Bungarus fasciatus* venom (BFV) in experimental animal models. Rheumatoid arthritis was induced by Freund’s complete adjuvant (FCA) in male Wistar albino rats. Lyophilized BFV was diluted in 0.9% NaCl. Antiarthritic activity showed that BFV significantly reduced the paw and ankle diameters; urinary hydroxyproline, glucosamine levels and serum ACP/ALP/TNF-α/IL-1β/IL-17/Cathepsin-K/MMP-1 levels. These parameters were significantly increased in FCA induced arthritic animals. Joint histopathology study indicated the partial restoration of joint structure. Treatment with BFV significantly reduced the mean latency time of tail flick response, acetic acid induced writhing response and formalin induced licking response in male albino mice. BFV treatment also significantly reduced carrageenan induced paw edema and xylene induced ear edema in male albino mice. The results indicated that BFV possess antinociceptive, anti-inflammatory and antiarthritic properties and further studies are warranted to find the active constituents present in BFV.

**Keywords**: Banded krait, Joint inflammation, Rheumatoid arthritis, Snake venom

Rheumatoid arthritis is an autoimmune, chronic inflammatory, debilitating disorder that involves joint and systemic inflammation with chronic pain. Joint inflammation damages bone, cartilage and tendons, while systemic inflammation affects the organs like lung, heart, etc. Inflammation is body’s own defense mechanism against injuries and infections. Any kind of physical, chemical or microbial, tissue injury triggers the release of neuromediators that turn on nociceptors, resulting in pain and inflammation. Inflammation is usually characterized by leucocyte migration, edema and granuloma formation at the site of injury and is also associated with pain, swelling, heat and redness. It was observed in experimental models of rheumatoid arthritis that peripheral sensitization was turned on following activation of nociceptive system which ultimately leads to the activation of central sensitization¹.

Available treatments for nociception and arthritis include nonsteroidal anti-inflammatory drugs (NSAIDS), glucocorticoids, disease-modifying antirheumatic drugs (DMARDs) and biological response modifiers. NSAIDS and glucocorticoids can reduce pain, inflammation and stiffness in the joints while DMARDS are also frequently used in clinical practice and TNF-α antagonists, IL-1 antagonist, anti-CD20 antibody are also used². Although these treatments offer some beneficial effects against pain and arthritis, they are also associated with certain side effects like gastrointestinal tract complications, gastric ulcer, haemorrhagic erosions, etc.³,⁴ Such limitations of different drugs and modern treatments trigger the search for alternative medicines using natural products. Indian medicinal text Ayurveda sixth chapter [Vishachikitsa (poison therapy)] too mentions about the use of snake venom in treating several inflammatory diseases like arthritis, thrombosis and cancer⁵. In Unani medicinal system also, use of cobra venom as treatment of pain, inflammation and arthritis has been highlighted⁶. Toxinological research has already reported the medicinal use of venom and toxins in different pathological conditions. An analgesic substance more potent than morphine was found from the venom of snakes belonging to Elapidae family⁷.

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**Materials and Methods**

**Chemicals**

All chemicals and reagents of analytical grade used in the experiment were as follows: acetic acid (Merck, India), carrageenan (Sigma, USA), copper sulphate (SRL, India), ethanol (SRL, India), Freund’s complete adjuvant (Sigma, USA), formaldehyde (SRL, India), glycine (SRL, India), hydrogen peroxide (Merck, India), hydrochloric acid (Merck, India), Osteomol (Merck, Germany), para dimethyl aminobenzaldehyde (SRL, India), p-nitrophenyl phosphate (SRL, India), sodium hydroxide (SRL, India), sodium chloride (SRL, India), sodium citrate (SRL, India), xylene (SRL, India), calcium kit (Merck, India), creatinine kit (Enoline, Merck, India), TNF-α, IL-1β, MMP-1 and cathepsin-k ELISA kit (R & D systems, USA) and IL-17 ELISA kit (Novus Biologicals, USA).

**Collection and preparation of snake venom**

Lyophilized *Bungarus fasciatus* venom (BFV) was purchased from Calcutta snake park, Kolkata, India. Collected BFV was dissolved in 0.9% NaCl to a concentration of 12 mg/mL (w/v) and stored at 4°C till further use.

**Animals**

Swiss albino male mice (20±2 g) and Wistar albino male rats (120±10 g) (n=6) were used for the experiments and procured from approved animal breeders of Calcutta University. They were kept 12 h day/night in polypropylene cages at 25±2°C, with 65% relative humidity and provided with pellet diet and water ad libitum. Before conducting experiments, animal ethical clearance was availed from the Department of Physiology, University of Calcutta (IAEC-III/Proposal/ AG-02/2012 dated 07.06.2012).

**Antiarthritic activity study**

**Development of experimental rheumatoid arthritis (RA)**

An emulsion of Freund’s complete adjuvant (FCA) in olive oil (1:1 v/v) was prepared at a concentration of 0.25 mg of heat killed *Mycobacterium tuberculosis*/mL. Experimental RA was induced by injecting 0.05 mL of that emulsion into the subplantar region of right hind paw of the rats. In the left hind paw same amount of saline was injected.

**Treatment schedule**

Animals were divided into 4 groups (n=6). Group I, Sham control; Group II, RA control; Group III, Standard drug, Indomethacin treated (0.25 mg kg⁻¹, p.o. ×5 days, alternatively); and Group IV, BFV treated (30 µg 100 g⁻¹, i.p. ×14 days).

**Physical parameters**

To determine degree of swelling, paw and ankle diameters were measured using electronic digital calipers (Mitutoyo, Japan) in an interval of 5 days from day 0 after FCA induction. Photographs of right hind paw were also taken with digital camera.

**Biochemical analysis of urinary and serum parameters**

Urinary glucosamine and urinary hydroxyl-proline levels were measured spectrophotometrically (Analab UV-180). Serum ACP and ALP levels were measured by biochemical method.

**Measurement of serum pro-inflammatory molecular markers**

Serum pro-inflammatory cytokines TNF-α, IL-1β, IL-17, cathepsin-k and MMP-1 levels were measured by ELISA kit (R & D systems, USA and Novus Biologicals, USA) using ELISA reader (BioTek, ELx800).

**Histopathological studies**

Paw and ankle joints were collected from rats and fixed in 10% buffered formalin for 24 h. Decalcification of the joints was carried out in Osteomol for 4-5 days. The joints were further processed by dehydrating in graded alcohol (50-100%), clearing in xylol, embedding in Paraffin (56-58°C) and blocks were prepared. Sections of 5 µm were cut using a rotary microtome (Weswox Optic, India), stained with haematoxylin-eosin and were observed under bright field microscope. Photographs were taken using Motic software (Motic Images Plus2.0 software).
Antinociceptive activity study of BFV
Tail flick response in mice model

Latency of tail flick response was analyzed in mice model\(^1\). Mice were divided into 3 groups (n=6). Gr. I, sham control; Gr. II, BFV treated (1.25 mg kg\(^{-1}\), i.p.); and Gr. III, standard drug, aspirin treated (150 mg kg\(^{-1}\), i.p.). After marking 3-4 cm area of the tail, it was dipped in the water bath, thermostatically maintained at 51±0.5ºC. The withdrawal time of the tail was recorded as latency in nociceptive response. To avoid the injury of tail tissue, the maximum cut off time of immersion of tail was kept up to 180 s. BFV (1.25 mg kg\(^{-1}\), i.p.) and standard drug, aspirin (150 mg kg\(^{-1}\), i.p.) were given 1 h before the experiment.

Acetic acid induced writhing response in mice model

Acetic acid induced writhing response was analyzed in mice model\(^1\). Mice were divided into three groups (n=6). Gr. I, acetic acid control; Gr. II, BFV treated (1.25 mg kg\(^{-1}\), i.p.); and Gr. III, standard drug, aspirin treated (150 mg kg\(^{-1}\), i.p.). About 0.6% acetic acid solution (10 mL kg\(^{-1}\)) was injected intraperitoneally in all animals, and the number of writhes for the next 15 min were recorded in both control and treatment group of animals. For the treated groups, BFV (1.25 mg kg\(^{-1}\), i.p.) and standard drug, aspirin (150 mg kg\(^{-1}\), i.p.) were given 1 h prior injection of acetic acid.

Formalin induced licking response in mice model

Formalin induced licking response was analyzed in mice model\(^1\). Mice were divided into 3 groups (n=6). Gr. I, formalin control; Gr. II, BFV treated (1.25 mg kg\(^{-1}\), i.p.); and Gr. III, standard drug, aspirin treated (150 mg kg\(^{-1}\), i.p.). 0.1% formalin (2.5% formalin solution; 10 mL kg\(^{-1}\)) was injected intraperitoneally in all animals, and the number of licking for the next 5 min were recorded in both control and treatment group of animals. For the treated groups, BFV (1.25 mg kg\(^{-1}\), i.p.) and standard drug, aspirin (150 mg kg\(^{-1}\), i.p.) were given 1 h prior injection of formalin.

Anti-inflammatory activity study of BFV
Carrageenan induced paw edema in mice model

Carrageenan induced paw edema was analyzed in mice model\(^1\). Mice were divided into three groups (n=6). Gr. I, carrageenan control; Gr. II, BFV treated (1.25 mg kg\(^{-1}\), i.p.); and Gr. III, standard drug, aspirin treated (150 mg kg\(^{-1}\), i.p.). Initially, volume of the right hind paw of mice was measured using digital calipers. Then, 0.05 mL of 1% carrageenan was injected subcutaneously into the subplantar region of right hind paw. To determine the volume of paw edema, the volume of right hind paw was measured at 1, 2, 3, 4 and 5 h after carrageenan injection. Treated groups were given standard drug, aspirin (150 mg kg\(^{-1}\), i.p.) and BFV (1.25 mg kg\(^{-1}\), i.p.) 1 h prior to carrageenan injection.

Xylene induced ear edema in mice model

Xylene induced ear edema was analyzed in mice model\(^1\). Mice were divided into 3 groups (n=6). Gr. I, Xylene control; Gr. II, BFV treated (1.25 mg kg\(^{-1}\), i.p.); and Gr. III, standard drug, indomethacin (1 mg kg\(^{-1}\), orally). Ear edema was induced by applying xylene (0.03 mL) to both the anterior and posterior surfaces of right ear of mice. Untreated left ear served as control. Treated groups were given indomethacin (1 mg kg\(^{-1}\), orally) and BFV (1.25 mg kg\(^{-1}\), i.p.) were administered one hour before xylene application. After one hour of giving xylene, animals were killed by cervical dislocation and circular sections (7 mm) were cut from both right and left ears of each mouse using a cork borer and weighed to record the ear edema level.

Statistical analysis

Data expressed in terms of mean ± SEM (n=6). One way ANOVA was used for determination of significant levels. \( *P <0.05 \) was considered as statistically significant.

Results

Antiarthritic activity of BFV

Effect of BFV on physical parameters

BFV (30 µg 100 g\(^{-1}\)) treatment significantly reduced paw and ankle diameters; by 23.28 and 9.08%, respectively whereas indomethacin treated group showed 27.56 and 8.27% restoration of paw and ankle diameters, respectively as compared to RA control group (Gr. II) rats. On the other hand, paw and ankle diameters were significantly increased in RA control rats (Gr. II) as compared to sham control (Gr I) rats (Fig. 1).

![Fig. 1—Effect of BFV and standard drug (Indomethacin) on (A) paw; and (B) ankle diameter of FCA induced rats. [Data expressed as mean ± SEM (n=6). Gr. I, Sham control; Gr. II, Arthritis control; Gr. III, Standard drug (Indomethacin) treated; and Gr. IV, BFV (30 µg 100 g\(^{-1}\)) treated]](image)
Table 1—Effect of BFV and indomethacin on urinary hydroxyproline and glucosamine levels, serum ACP/ALP levels and serum pro-inflammatory cytokine MMP-1 levels in FCA induced rats

<table>
<thead>
<tr>
<th>Groups</th>
<th>Hydroxyproline (µg/ml/18hr/rat)</th>
<th>Glucosamine (µg/ml/18hr/rat)</th>
<th>ACP (µmole of PNPP/min)</th>
<th>ALP (µmole of PNPP/min)</th>
<th>MMP-1 (pg/ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td>I</td>
<td>52.32±2.53</td>
<td>101.02±2.13</td>
<td>12.29±1.13</td>
<td>60.26±2.63</td>
<td>266.27±6.39</td>
</tr>
<tr>
<td>II</td>
<td>152.25±2.87*</td>
<td>504.98±5.96*</td>
<td>38.98±2.75*</td>
<td>194.78±4.41*</td>
<td>667.27±10.23*</td>
</tr>
<tr>
<td>III</td>
<td>62.86±2.51**</td>
<td>242.68±4.62**</td>
<td>17.51±1.51**</td>
<td>134.07±2.75**</td>
<td>419.73±9.44**</td>
</tr>
<tr>
<td>IV</td>
<td>81.49±3.13**</td>
<td>265.55±4.51**</td>
<td>20.12±2.05**</td>
<td>141.96±3.13**</td>
<td>390.51±7.33**</td>
</tr>
</tbody>
</table>

[Data expressed as mean ± SEM (n=6). One-way ANOVA was done for statistical analysis and *P <0.05 was considered as significant, (Gr. I vs. Gr. II) and **P <0.05 was considered as significant, (Gr. II vs. Gr. III/Gr. IV) where Gr. I, Sham control; Gr. II, Arthritis control; Gr. III, Standard drug (Indomethacin) treated; and Gr. IV, BFV (30 µg 100 g\(^{-1}\)) treated]

**Effect of BFV on urinary parameters**

BFV (30 µg 100 g\(^{-1}\)) treated group showed significant reduction in urinary hydroxyproline and glucosamine levels by 46.48 and 47.41%, respectively; whereas indomethacin treated group showed reduction of the above parameters by 58.71 and 51.94%, respectively as compared to the respective RA control (Gr. II) rats. On the other hand, urinary hydroxyproline and glucosamine levels were significantly increased in RA control group as compared to sham control (Gr. I) rats (Table I).

**Effect of BFV on serum parameters**

BFV (30 µg 100 g\(^{-1}\)) treated group showed a significant reduction in serum ACP and ALP levels by 48.38 and 27.12%, respectively; whereas standard drug indomethacin showed a significant reduction of serum ACP and ALP levels by 55.08 and 31.17% as compared to RA control group rats.

BFV (30 µg 100 g\(^{-1}\)) treatment showed a significant reduction in serum interleukins TNF-α, IL-1β, IL-17, MMP-1 and cathepsin-K levels by 49.84, 59.92, 64.57, 41.48 and 54.95%, respectively when compared to RA control group (Gr. II). Whereas, indomethacin treated group showed reduction of the above parameters by 47.04, 54.63, 48.54, 37.09 and 43.55, respectively as compared to RA control group. On the other hand, serum ACP, ALP, TNF-α, IL-1β, IL-17, MMP-1 and cathepsin-K levels were found significantly increased in RA control group as compared to sham control (Gr. I) rats (Table I, Fig. 2).

**Effect of BFV on joint histology**

BFV (30 µg 100 g\(^{-1}\)) treatment showed significant increase in synovial space and partial healing of synovial membrane. Whereas, decreased synovial space and increased tearing down of synovial membrane with pannus formation was seen in RA control (Gr. II) rats. (Fig. 3)
Table 2—Effect of BFV and standard drug on Carrageenan induced paw edema, xylene induced ear edema, tail flick response, acetic acid induced writhing response and formalin induced licking response in mice

<table>
<thead>
<tr>
<th>Groups</th>
<th>Paw edema (cm)</th>
<th>Ear edema (mg)</th>
<th>Latency of nociceptive response (sec)</th>
<th>Number of writhings</th>
<th>Number of lickings</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Early phase (0-5 min)</td>
<td>Late phase (15-30 min)</td>
</tr>
<tr>
<td>I</td>
<td>1.61±0.26</td>
<td>8.80±0.75</td>
<td>5.50±0.85</td>
<td>32.67±1.01</td>
<td>22.14±1.24</td>
</tr>
<tr>
<td>II</td>
<td>0.69±0.09*</td>
<td>3.51±0.62*</td>
<td>20.33±1.48*</td>
<td>8.33±1.19*</td>
<td>5.09±0.87*</td>
</tr>
<tr>
<td>III</td>
<td>0.57±0.04*</td>
<td>2.61±0.36*</td>
<td>30.67±1.69*</td>
<td>5.50±0.59*</td>
<td>8.41±1.31*</td>
</tr>
</tbody>
</table>

[Data expressed as mean ± SEM (n=6). One-way ANOVA was done for statistical analysis and *P <0.05 was considered as significant, (Gr. I vs. Gr. II/Gr. III) where Gr. I, Control; Gr. II, BFV (25 µg 100 g⁻¹) treated; and Gr. III, Standard drug treated]

Analgesic activity of BFV

Effect of BFV on tail flick response

BFV (1.25 mg kg⁻¹) and aspirin (150 mg kg⁻¹) treated groups showed significant analgesic effects on pain and the percentage of analgesic activity exhibited by them were 72.95 and 82.07 %, respectively as compared to sham control group (Table 2).

Effect of BFV on acetic acid induced writhing response

BFV (1.25 mg kg⁻¹) and aspirin (150 mg kg⁻¹) treated groups showed reduction in writhing numbers and the percentage exhibited by them were 74.50 and 83.16%, respectively as compared to acetic acid control group (Table 2).

Effect of BFV on formalin induced licking response

BFV (1.25 mg kg⁻¹) and aspirin (150 mg kg⁻¹) treated groups showed reduction in licking numbers and the percentage exhibited by them were 77.01 and 62.01%, respectively in early phase (0-5 min) and 73.38 and 56.28%, respectively in late phase (15-30 min) as compared to formalin control group (Table 2).

Anti-inflammatory activity of BFV

Effect of BFV on Carrageenan induced paw edema in mice

BFV (1.25 mg kg⁻¹) treatment showed a 57.10% reduction of paw edema volume and aspirin (150 mg kg⁻¹) treated group showed 64.60% reduction of paw edema volume after 3 h of carrageenan injection as compared to carrageenan control group (Table 2).

Effect of BFV on Xylene induced ear edema in mice

BFV (1.25 mg kg⁻¹) treatment showed a 60.11% reduction of ear edema volume and indomethacin (1 mg kg⁻¹) treated group showed 70.34% reduction of ear edema volume as compared to xylene control group (Table 2).

Discussion

In the present study, the antiarthritic potential of *Bungarus fasciatus* venom (BFV) was evaluated in FCA-induced experimental arthritis model. As pain and inflammation are intimately associated with rheumatoid arthritis, effect of BFV on pain and inflammation was also evaluated in different model. Rheumatoid arthritis is associated with swelling of paw and ankle resulting from chronic inflammation. BFV treatment significantly reduced paw/ankle swelling of arthritic rats indicating that it might reduce inflammation associated with RA.

Hydroxyproline, a nonessential imino acid, is formed in the body in collagen as a post-translational modification of the amino acid proline by prolyl hydroxylase and it imparts stability to the collagen triple helix. Increased excretion of hydroxyproline in urine is associated with collagen degradation. On the other hand, collagen degradation was seen in arthritis. An increased excretion of hydroxyproline in urine was observed in arthritic control rats whereas BFV treatment significantly restored urinary hydroxyproline levels indicating that it could inhibit degradation of collagen in arthritis. Arthritis is also associated with cartilage degradation. In cases of cartilage degradation, an increased excretion of glucosamine is seen in urine. This is due to destruction of cartilage matrix glycoprotein by glycohydrolase activity. Glucosamine in urine serves as a laboratory marker for arthritis. An increased urinary glucosamine level was found in arthritic control rats whereas BFV treatment significantly restored urinary glucosamine levels showing that it could prevent cartilage damage by balance the making and breaking of cartilage.

In rheumatoid arthritis, due to disintegrity of lysosomal membrane, serum ACP and ALP levels increases markedly and results in the degradation of
extracellular matrix. Significant restoration of serum ACP and ALP levels in BFV treated group as compared to arthritic control group indicated that BFV could restore lysosomal membrane integrity. Cathepsin-K, a proteolytic enzyme, is capable of cleaving fibrillar collagen which ultimately leads to articular cartilage destruction\(^\text{28}\). Increased levels of cathepsin-K was found in serum of arthritic control rats but BFV treatment significantly restored cathepsin-K levels, again indicating that it could prevent cartilage damage.

Further, due to chronic infiltration of T-lymphocytes in rheumatoid arthritis, plasma cells and macrophages in the synovial membrane, levels of pro-inflammatory cytokines such as TNF-\(\alpha\), IL-1\(\beta\) and IL-17 levels are increased\(^\text{29}\). Disparity between pro- and anti-inflammatory cytokines induces autoimmunity and chronic inflammation. T-cells activate macrophages and the activated macrophages secrete TNF-\(\alpha\). TNF-\(\alpha\) regulates the synthesis of IL-1\(\beta\), which leads to the production of MMPs. IL-17 also stimulates the production of TNF-\(\alpha\) and IL-1\(\beta\)\(^\text{23}\). Another pro-inflammatory cytokine, CINC-1, recruits neutrophils in the affected joints and aggravates the inflammatory condition\(^\text{30}\). In the present study also, we have observed an increased level of pro-inflammatory mediators (TNF-\(\alpha\), IL-1\(\beta\), CINC-1, IL-17 and MMP-1) in arthritis control rats. BFV treatment significantly restored the level of those pro-inflammatory mediators, indicating that it could prevent inflammation and help to maintain a balance of cytokines in joints. Joint histological features in rheumatoid arthritis exhibit increased destruction of synovial membrane and decreased synovial space\(^\text{27}\). BFV treatment significantly restored the normal architecture of joints, as also evidenced by the changes in urinary and serum markers.

Chronic pain in the joints can be regarded as one of the major symptoms of rheumatoid arthritis\(^\text{37}\). In the tail flick model, in control group, pain was reflected by fast withdrawal of tail from hot water bath whereas delayed withdrawal of tail was seen in BFV treated group. Ligands of \(\mu\)-opioid receptors are known to produce analgesia by activating the \(\mu\)-opioid receptor system\(^\text{31}\). Therefore, BFV, by producing analgesia, might have exerted its effect through \(\mu\)-opioid receptors. Acetic acid produces nociceptive writhing by activating peritoneal macrophages and mast cells and inducing them to release inflammatory cytokines like TNF\(\alpha\), IL-1\(\beta\) and IL-8\(^\text{32}\). Analgesic effect of BFV was probably due to its inhibitory effect on those cytokines. To estimate the response caused by continuous pain that was generated by injured tissue, formalin-induced pain test was done. The formalin test has two phases known as the early and late phases. The early phase or neurogenic phase (0-5 min) is due to the release of substance P in the paw and the late phase or inflammatory phase (15-30 min) is due to the liberation of histamine, serotonin, bradykinin and prostaglandin\(^\text{33}\). Decrease in the number of lickings of paw in both early and late phases of BFV treated group as compared to the formalin induced control group was observed. Analgesic activity of BFV might be due to inhibition of the release of those inflammatory mediators.

In carrageenan induced paw edema model, carrageenan causes edema by inducing the release of kinins, histamine and 5-hydroxytryptamine\(^\text{34}\). Due to the action of these inflammatory agents, neutrophils migrate to the site of inflammation resulting in paw edema. Inhibitory effect of BFV on paw edema might be due to inhibiting the release or synthesis of these inflammatory mediators\(^\text{29}\). In xylene induced ear edema model, xylene induces ear edema by stimulating the release of histamine, serotonin and bradykinin which causes vasodilation and increases vascular permeability, resulting in easy passage of neutrophils from blood circulation to inflammation site\(^\text{35}\). BFV, by reducing the volume of ear edema, might have inhibited the release or interfere with the action of the above inflammatory mediators whose levels flared up in xylene control group resulting in increased volume of ear edema.

Many snake venom protein toxins like NKCT-1, NN-32 exhibit cytotoxic effect to inflamed cells and also antiarthritic\(^\text{36,37}\). \textit{Bungarus fasciatus} venom (BFV) also possesses cytotoxic activity\(^\text{12}\). As both cancer and arthritis is linked with inflammation, antiarthritic activity of BFV could be attributed to inactivation of some of the components of signaling pathways associated with inflammation. Further studies are necessary to establish BFV as an antiarthritic natural agent.

**Conclusion**

It may be concluded that \textit{Bungarus fasciatus} venom (BFV) possess antinoceptive, anti-inflammatory and antiarthritic activity in experimental animal models. Further work is in progress to purify the active compound from BFV and to determine the detail molecular mechanism of its antiarthritic property.
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Conflict of interest
We declare that there is no conflict of interest among the authors.

References
7 Bisset NG, One man’s poison, another man’s medicine? J Ethnopharmacol, 32 (1991) 71.