Anti-Rheumatoid activity of Sesamin Complex in Freund's Complete Adjuvant induced arthritis in Wistar albino rats

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ABSTRACT

Objectives: To evaluate the anti-rheumatoid activity of Sesamin Complex in Freund’s Complete Adjuvant (FCA) induced arthritis in Wistar albino rats. Methodology: A total of 36 female Wistar albino rats weighing 150-200 g were selected and allocated to 6 groups with 6 rats in each. Group 1 served as normal control (NC). The animals in group 2 to 6 were injected with single dose of 0.1 ml of FCA intradermally into the left hind paw on day 0. All the animals developed arthritis by day 7. From day 7 to 28, the animals were treated with the following. Group 1 & 2 normal saline 10 ml/kg, Group 3, Diclofenac 25 mg/kg, Group 4, Methotrexate 50 μg/kg/week, Group 5, Sesamin complex (SC) 15 mg/kg and Group 6, SC 30 mg/kg per oral once daily. Body weight, temperature, spontaneous activity and paw volume (PV) were measured once in 7 days for all the groups. Erythrocyte sedimentation rate (ESR), C-reactive protein (CRP), Hemoglobin (Hb%), White blood cell (WBC) count, Red blood cell (RBC) count, Interleukin-6 (IL-6) and Tumor necrosis factor alpha (TNF-α) were estimated on day 7 and 28. Radiological and histopathological examinations of the ankle joint were done on day 28. The results were analyzed using One-way Analysis of Variance (ANOVA) followed by post hoc (Tukey’s) test. Results: All the animals injected with FCA developed arthritis. Animals treated with SC, diclofenac and methotrexate showed a significant decrease in PV, body temperature, WBC count, ESR, CRP, IL-6 and TNF-α and a significant increase in body weight, spontaneous activity, Hb%, RBC count when compared with RA control (P < 0.01). Histopathological and radiological examination showed improvement in joint space and reduction in joint swelling in SC treated as well as the groups treated with diclofenac and methotrexate. Both SC 15 mg/kg and 30 mg/kg produced significant anti-inflammatory effect. SC 30 mg/kg produced an effect equivalent to diclofenac and methotrexate. Conclusion: Sesamin complex had anti-rheumatoid activity. The effect of 30 mg/kg of Sesamin complex was comparable to diclofenac and methotrexate.

Keywords: Sesamin, Sesamolin, Rheumatoid, Freund’s Complete Adjuvant
of the biomarkers of auto-immune disorders (5). The management of RA includes drugs like Disease modifying anti-rheumatoid drugs (DMARDs), NSAIDs like Ibuprofen, Diclofenac, Corticosteroids, Biologicals such as TNF-α and IL antagonists and JANUS kinase inhibitors (JKIs) have been used to relieve pain and reduce immunological reaction mediated inflammation and joint damage. Conventional DMARDs have several limitations like slow onset of action, induction of partial remission and 5 years relapse rates. NSAIDs usage also has adverse effects like gastric ulcer, renal & vascular complications. However these drugs neither offer a complete cure nor are free from adverse effects. In recent years, there is an upsurge in the clinical usage of indigenous drugs in chronic arthritic disorders because of their efficacy & lack of serious adverse effects. Hence finding out new drugs is essential. In the Indian medical systems, many medicinal plants (e.g., Calotropis procera, Hemidesmus indicus, Tridax procumbens, Sesamum indicum) and their products are being used which are known to cause beneficial effects in RA. Among them, Sesamum indicum (SI) is almost used in day to day practice in every household. SI seed oil is used in cooking for many years in south India. It is known to have several medicinal properties like antioxidant, analgesic and anticonvulsant activities. Sesamum indicum (SI) is an age-old spice, one among the plants first used for its seeds. It is being used for many years and an oil seed of significance throughout the world (6). The pharmacological activities of SI seed extract were evaluated by previous researchers and reported that it has antioxidant, analgesic, anti-obesity, nephro and hepatoprotective activities (7, 8, 9, 10). The anti-rheumatoid activity has been recently reported (11). All these activities have been attributed to the phytoconstituents, sesamin, sesamolin and sesamol present in the seed extract. Sesamin and sesamolin are the two important lignans found in sesame seeds. Both these lignans together is available as sesamin complex. **Sesamin complex:** Generally, Sesamin Oil contains 0.6 to 0.7% of Sesamin and 0.3% to 0.2% of Sesamolin. These two active compounds will be extracted into solvent phase for more than 12 hours, concentrated and then subjected for crystallization below 5°C temperature for a period of 24 hours. The crystalline powder will be dried analysed for its purity. Normally, after completion of the process Sesamin and Sesamolin together would be 90%. This compound is commercially known as Sesamin Complex 90%, in which Sesamin would be 80% and Sesamolin would be 10% depending on the raw material/oil. The present study was undertaken to evaluate the effect of this complex in FCA induced animal model of rheumatoid arthritis.

**MATERIALS AND METHODS:**

**Drugs and chemicals:** Sesamin complex was purchased from VPL chemicals, Bangalore. Freund complete adjuvant reagent was purchased from Genei Laboratories Private Limited, Bangalore. Diclofenac was obtained from Cipla Limited, Mumbai. Methotrexate was obtained from Sun Pharmaceutical Industries Limited, Gujarat.

**Kits:** ELISA kits for the estimation of rat plasma TNF-α was purchased from Korain Biotech Co., Ltd and for IL-6 was purchased from Shanghai Coon Koon Biotech Co., Ltd.
Evaluation of anti-rheumatoid activity

Animals: Female Wistar albino rats were procured from the central animal house at Chettinad Hospital and Research Institute. The study was initiated after receiving the approval of Institutional animal ethics committee (IAEC/Alr.No:21/Dt.12.12.2017). The animals were handled as per the guidelines prescribed by the CPCSEA. They were maintained at temperature 23-25°C, humidity 50-60% in alternate light – dark cycle with water and food.

Experimental design: Thirty six female Wistar albino rats with body weight between 150-200 grams were selected and allocated to 6 different groups with 6 rats in each group. The day of induction of arthritis was taken as day ‘0’. On day ‘0’, before induction of arthritis, body weight, body temperature, spontaneous activity and paw volume were measured for all the animals. The study group details are as follows.

<table>
<thead>
<tr>
<th>Groups</th>
<th>Treatment</th>
</tr>
</thead>
<tbody>
<tr>
<td>Group 1</td>
<td>Normal control (NC) – Normal saline 10 ml/kg</td>
</tr>
<tr>
<td>Group 2</td>
<td>RA control (RAC) – Normal saline 10 ml/kg</td>
</tr>
<tr>
<td>Group 3</td>
<td>RA + Diclofenac (D) 25 mg/kg</td>
</tr>
<tr>
<td>Group 4</td>
<td>RA + Methotrexate (M) 50 μg/kg/week</td>
</tr>
<tr>
<td>Group 5</td>
<td>RA + Sesamin complex (SC) 15 mg/kg</td>
</tr>
<tr>
<td>Group 6</td>
<td>RA + Sesamin complex (SC) 30 mg/kg</td>
</tr>
</tbody>
</table>

The interventions were carried out once daily per oral from day 7–28.

All the test and standard drugs were given orally once a day in the morning Methotrexate alone was given once a week in the morning

Dose selection: The doses of 15 and 30 mg/kg were chosen based on a previous study in which a better response is reported for 30 mg/kg for reduction of oxidative stress and mortality in status epilepticus (14).

Induction of RA (12) All the rats in the experimental groups were injected with 0.1 ml of FCA intradermally into the left hind paw on day ‘0’. The rats were examined once in 3 days for assessment of arthritis from day 1 to 28. The signs of arthritis such as redness, swelling of the paw and restricted mobility of the animals developed fully on the 7th day. Blood samples were collected on day 7 by retro orbital puncture for estimation of biochemical parameters. Treatment of arthritis was started on day 7 and completed on day 28. Paw volume, body temperature, body weight and spontaneous activity were measured once in 7 days. On 28th day X-ray of the ankle joint was taken for all the rats and then sacrificed by using halothane in high dose. Blood samples were collected for post-treatment biochemical assays and joints were dissected for histological studies.

Assessment of physical parameters

Animal body weight: Body weight of animals was measured using a digital weighing machine on day 0, 7, 14, 21 and 28 and the mean was calculated (15).

Hind paw volume: Paw volume in all the groups was measured by plethysmometer on days 0, 7, 14, 21 and 28. The mean change in paw volume and its percentage reduction were calculated on days 14, 21 and 28 using the following formula.

\[
\text{Percentage inhibition of paw edema} = \frac{V_c - V_t \times 100}{V_c}
\]

Where \( V_c \) is the average paw volume of RA control and \( V_t \) is the average paw volume of treated group.

Temperature: Rectal temperature was measured using digital rectal thermometer (16).

Spontaneous activity: The spontaneous loco motor activity was assessed in digital actophotometer for duration of one minute (17).

Blood sample collection: 0.5 ml of blood was collected on day 7 and 28 from retro orbital sinus as per the method prescribed by Parasuraman S et.al (18).

Assessment of biochemical parameters: The blood samples were analyzed for the hematological parameters Hb, ESR, CRP, total RBC and total WBC count using an automated cell counter.

Determination of serum IL-6 and TNF-α levels: The serum was separated by centrifugation (3000 rpm, 10 mins) and stored at -20°C until used. Concentration of IL-6 and TNF-α was determined by ELISA. Optical density was assessed by ELISA reader at 450 nm.

Morphological changes of joints: Radiographic changes were assessed on the basis of radiographs taken with Siemens Heliphos-D X-ray machine.

Histopathological study of joints: All the animals were sacrificed by administering high dose of halothane. Hind limbs were detached and fixed in 10% buffered formalin. Decalcification was done using 5% formic acid and processed for paraffin embedding. The tissues were sectioned to 5 µm thickness and stained with haemotoxylin-eosin. Sections were examined for the presence of hyperplasia of the synovium, infiltration with inflammatory cells, pannus formation and the extent of destruction of the joint spaces under light microscope.

Statistical analysis: The results were expressed as mean ± SEM and the difference between the means was calculated using One-way ANOVA followed by post hoc (Tukey’s) test. P value less than 0.05 was considered statistically significant.

RESULTS

Effects on physical parameters Paw volume (PV)

Paw volume (PV) was increased in all the groups except the normal when measured on the 7th day. The mean PV in normal control group was 0.15±0.01 ml. There was a significant increase in PV in RA induced rats (1.16±0.02). After treatment PV was reduced in test and standard groups. The extent of PV reduction was found to be
significant when compared to RA control group on day 28 in all the treatment groups-Diclofenac (D), Methotrexate (M), Sesamin complex (SC) 15 and SC 30 (P < 0.01) (Figure 1). On day 28, the reduction in paw volume was almost equal in SC 30 and D (Figure 2).

**Body weight (BW):** Rats in the normal group had a gradual increase in body weight. Body weight was significantly decreased in RA control group. The rats treated with D, M and SC showed a gradual increase in body weight from 14th day to 28th day. The increase in body weight on 28th day was significant compared to RA control (D and SC 15: P < 0.05, SC 30: P < 0.01) (Figure 3).

**Body temperature (BT):** The mean normal body temperature was 36.70 ± 0.20 C. After arthritis was induced, the body temperature increased to 38.93 ± 0.100 C. Treatment with D, M and SC resulted in significant reduction in body temperature and the reduction was significant in D, M, SC 15 and SC 30 groups (P < 0.01) when compared with RA control on day 28 (Figure 4).

**Spontaneous activity (SA):** SA on day 0 was 173.50 ± 1.23. On 7th day, SA was reduced to 60.83 ± 2.50 in RA control group. On day 28, standard and test drugs improved the SA significantly (P < 0.01) when compared with RA control. D improved the SA from 58.50±3.27 on day 7 to 171.67±1.41 on day 28, whereas the group treated with SC showed the maximum SA of 172.50±1.02 on day 28 from the baseline, 59.00±3.21 on day 7 at the dose of 30 mg (P < 0.01). The improvement in SA shows reduction in inflammation and pain (Figure 5).

**Effects on RBC, Hb, WBC, ESR, CRP:** Induction of arthritis resulted in a reduction in Hb level and RBC count, and an increase in WBC count, ESR and CRP when compared to normal control. Treatment with Diclofenac, SC 15 and SC 30 had shown significant increase in Hb (P < 0.01) and RBC count (P < 0.05) on day 28 when compared with RA control and decrease in the total WBC count, ESR and CRP on day 28 when compared to RA control (P < 0.01). Treatment with M caused reduction in Hb% (10.83 ± 0.15) and RBC count (6.42 ± 0.17); but treatment with SC 30 mg/kg increased the Hb% (14.20 ± 0.32) and RBC count (7.58 ± 0.20.) Rats treated with M alone showed a decrease in Hb level and RBC count. This could be due to the anti-folate effect of methotrexate (Figure 6 -10).

**Effect on level of IL-6 and TNF-α:** In RA control, the mean levels of IL-6 and TNF-α were 80.98 ± 0.81 pg/ ml and 71.23 ± 1.99 pg/ml, respectively on day 28. The mean levels of IL-6 and TNF- α were significantly reduced in all the treatment groups, when compared to RA control (P < 0.01). On day 28, following treatment with SC 30, the level of IL-6 was reduced to 55.02±0.47 and the level of TNF- α was reduced to 50.28 ± 0.96 pg/ml. In D group, the mean IL-6 and TNF- α levels were 56.30 ± 0.93 pg/ml and 50.65 ± 0.58 pg/ml respectively on day 28 (Figure 11 and 12).

**SC 30 Vs Diclofenac:** The difference in all the above-mentioned parameters was analysed statistically using unpaired t-test between SC 30 and the standard drug Diclofenac. It was observed that there was no statistically significant difference in any of these parameters between SC 30 and diclofenac. Hence, SC 30 is almost equally efficacious as diclofenac in FCA induced arthritis in rats. The comparison of SC 30 and diclofenac is shown in Table-1.
Radiological changes: Radiographic examination of RA induced joint showed absence of joint space and increased soft tissue swelling whereas the joint space was increased and soft tissue swelling was reduced in rats treated with standard and test drugs indicating the reduction in inflammation. But there was no difference between standard and test groups (Figure-13).

Histopathological changes: In normal control rats, the hind paw joints had intact articular cartilage with normal joint space and there was no inflammation. Damaged articular cartilage, narrowed joint space along with inflammatory changes were observed in RA control rats. The joint inflammation was reduced and joint space improved in rats treated with 15 and 30 mg/kg of SC, D and M. The improvement in joint changes was better with SC than D and M (Figure-14).

Figure 1: Effect on Paw volume

Values expressed as Mean ± SEM, n=6, ** P < 0.01 with RA control, # P < 0.01 with diclofenac, a P < 0.01 with Methotrexate
Figure 2: Percentage inhibition of paw volume
Values expressed as Mean±SEM, n=6, * P < 0.05, ** P < 0.01 with RA control
Figure 4: Effect on body temperature

Values expressed as Mean±SEM, n=6, ** P < 0.01 with RA control
Figure 5: Effect on spontaneous activity

Values expressed as Mean±SEM, n=6, ** P < 0.01 with RA control, # P < 0.01 with diclofenac, a P < 0.01 with Methotrexate
Figure 6: Effect on RBC count

Values expressed as Mean±SEM, n=6, * P < 0.05, a P < 0.01 with Methotrexate

Figure 7: Effect on hemoglobin

Values expressed as Mean±SEM, n=6, ** P < 0.01 with RA control, a P < 0.01 with Methotrexate
Figure 8: Effect on ESR

Values expressed as Mean±SEM, n=6, ** P < 0.01 with RA control

Figure 9: Effect on total WBC count

Values expressed as Mean±SEM, n=6, ** P < 0.01 with RA control
**Figure 10: Effect on C-reactive protein**

Values expressed as Mean±SEM, n=6, ** P < 0.01 with RA control.

**Figure 11: Effect on interleukin-6 levels**

Values expressed as Mean±SEM, n=6, ** P < 0.01 with RA control.
Figure 12: Effect on TNF-α levels

Table 1: SC 30 Vs Diclofenac

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Mean difference between day 7 and day 28</th>
<th>P value (Unpaired t test)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Diclofenac</td>
<td>SC 30</td>
</tr>
<tr>
<td>Reduction in paw volume</td>
<td>0.83</td>
<td>0.8</td>
</tr>
<tr>
<td>Reduction in body temperature</td>
<td>1.6</td>
<td>1.73</td>
</tr>
<tr>
<td>Increase in RBC count</td>
<td>0.53</td>
<td>0.53</td>
</tr>
<tr>
<td>Increase in Hb%</td>
<td>1.93</td>
<td>2.28</td>
</tr>
<tr>
<td>Reduction in ESR</td>
<td>5.67</td>
<td>5.67</td>
</tr>
<tr>
<td>Reduction in CRP</td>
<td>6.28</td>
<td>6.27</td>
</tr>
<tr>
<td>Reduction in IL-6</td>
<td>12.88</td>
<td>14.12</td>
</tr>
<tr>
<td>Reduction in TNF-α</td>
<td>9.63</td>
<td>10.88</td>
</tr>
</tbody>
</table>
Table 2: Percentage difference in all the parameters

<table>
<thead>
<tr>
<th>S.No</th>
<th>Parameters</th>
<th>Control</th>
<th>RA control</th>
<th>Diclofenac</th>
<th>Methotrexate</th>
<th>SC 15</th>
<th>SC 30</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Paw volume</td>
<td>6.67</td>
<td>-25.39</td>
<td>-73.97</td>
<td>-58.52</td>
<td>-68.18</td>
<td>-71.64</td>
</tr>
<tr>
<td>2</td>
<td>Body weight</td>
<td>10.76</td>
<td>-8.57</td>
<td>7.21</td>
<td>4.72</td>
<td>5.13</td>
<td>9.01</td>
</tr>
<tr>
<td>3</td>
<td>Body temperature</td>
<td>0.23</td>
<td>2.11</td>
<td>-4.23</td>
<td>-4.53</td>
<td>-4.06</td>
<td>-4.57</td>
</tr>
<tr>
<td>4</td>
<td>Spontaneous activity</td>
<td>0.67</td>
<td>112.6</td>
<td>193.45</td>
<td>172.39</td>
<td>149.74</td>
<td>192.37</td>
</tr>
<tr>
<td>5</td>
<td>Total RBC</td>
<td>-1.3</td>
<td>-7.11</td>
<td>7.39</td>
<td>-6.62</td>
<td>4.46</td>
<td>10.52</td>
</tr>
<tr>
<td>6</td>
<td>Hb</td>
<td>1</td>
<td>-12.6</td>
<td>16.34</td>
<td>-11.68</td>
<td>11.32</td>
<td>19.16</td>
</tr>
<tr>
<td>7</td>
<td>ESR</td>
<td>3.45</td>
<td>9.77</td>
<td>-50.75</td>
<td>-49.33</td>
<td>-46.38</td>
<td>-51.52</td>
</tr>
<tr>
<td>8</td>
<td>Total WBC</td>
<td>0.4</td>
<td>17.65</td>
<td>-33.88</td>
<td>-46.14</td>
<td>-33.29</td>
<td>-36.42</td>
</tr>
<tr>
<td>9</td>
<td>CRP</td>
<td>-0.4</td>
<td>20.84</td>
<td>-63.54</td>
<td>-62.13</td>
<td>-54.55</td>
<td>-61.64</td>
</tr>
<tr>
<td>11</td>
<td>TNF-α</td>
<td>0.73</td>
<td>19.02</td>
<td>-15.98</td>
<td>-18.58</td>
<td>-14.5</td>
<td>-17.79</td>
</tr>
</tbody>
</table>

Percentage change was calculated using the formula = [(Day 28 value - Day 7 value) ÷ Day 7 value] x 100 Symbol (-) indicates reduction
Figure 13: Radiological changes in joint
Figure 14: Histopathological changes of joints

B – Bone, C – Cartilage, SP – Synovial space
DISCUSSION

Rheumatoid arthritis is a chronic disorder characterized by inflammatory synovitis affecting mainly peripheral joints leading to progressive damage of cartilages and bones. Pain, stiffness, swelling, deformity and eventually loss of function of the joints are common manifestations of RA. The actual cause of RA is unknown, but the inflammatory mediators responsible for the pathogenesis have been extensively studied and they include pro-inflammatory cytokines, IL-1, IL-6, IL-17, IL-18, TNF-α & Interferons (13) and the inflammatory mediators, mainly prostaglandins (19). In addition to these known mediators acetyl choline (20) and substance P (21) have been also identified to play a role in inflammation. The Sesamum indicum seed is used in day to day practice almost in every household. Sesamum indicum seed oil is used in cooking for many years in south India. It is known to have several medicinal properties like antioxidant, analgesic and anticonvulsant activity. The oil of SI seed is the most commonly used household remedy for pain and inflammation in India. If SI products are found to be effective in controlling the inflammation in RA, they would offer the most cost-effective remedy without causing unacceptable adverse effects. The anti-rheumatoid effect of SI seed extract has been reported by previous authors in their study on FCA induced RA arthritis in wistar rats (11). The crude extract contains phytochemicals such as the lignans, sesamin and sesamolin, polyphenol such as sesamol, as well as saponins, flavonoids, tannins, glycosides and terpenes, oleic acid, α-tocopherol, γ-tocopherol, palmitic acid, stearic acid and linoleic acid and α–linolenic acid. Hence the effect of the extract could be due to all or any of the phytochemicals present in the extract. Among these chemicals sesamin and sesamolin is available as sesamin complex (SC). This complex was taken up for the current study.

SC was evaluated in the present study for its effect in FCA induced arthritis. It exhibited anti-inflammatory property evidenced by the reduction of cytokines IL-6 and TNF-α, CRP, ESR, total WBC count, animal body temperature and increase in Hb% and spontaneous activity. (Table 2 & Fig. 13, 14). When the effects of SC was compared with diclofenac and methotrexate its effects were comparable to both diclofenac and methotrexate. A dose dependent reduction in PV was observed with SC. SC at the dose of 15 mg/kg and 30 mg/kg showed marked reduction in PV which was equivalent to that of diclofenac and methotrexate. The reduction in PV with SC 30 mg/kg was highly significant when compared with Methotrexate. Though all these three drugs have reduced the inflammatory cytokines IL-6 and TNF-α, reduction seen with SC 30 mg/kg was equivalent to methotrexate and diclofenac. (p < 0.01, Fig 1 & 2)

One of the important features of RA is anaemia. Increase in IL-6 level is related to increased hepcidin level which an iron is regulating hormone formed in hepatocytes. The iron release from the macrophages in the spleen and uptake of iron in duodenum is inhibited by hepcidin which results anaemia (22, 23). Hb level was reduced in RA induced rats (10.8%) compared to normal control rats (13.47%). Treatment with SC has reduced the level of IL-6 which would consequently decrease the level of hepcidin. Hence iron release and uptake will improve. This has led to an increase in Hb% from 10.8% to 14.2% in SC treated animals (Fig.7) The increase in Hb% with SC was found to be significant (p< 0.01). However, the animals treated with methotrexate had low RBC count and Hb level (10.8%). Though methotrexate can also decrease IL-6 level, inhibition of folic acid
metabolism and bone marrow suppression are responsible for decreased hemopoiesis seen in the methotrexate group. Hence SC can be a safer alternative in this regard.

The increase in total white blood cell count in RA can be due to immune system stimulation against antigen invasion. The decrease in WBC count in SC treated groups shows their immunomodulatory effect. The ESR level which was significantly increased in arthritic control group has been remarkably decreased by SC as well as diclofenac and methotrexate. ESR is an indirect indicator of acute phase response (APR). Reduction in ESR is an evidence for reduced APR.

SC 30 mg/kg produced a better overall effect than SC 15. The reduction in paw volume, blood parameters, IL-6 and TNF-α when compared between SC 30 and diclofenac, they did not show any significant difference (Table-1).

The percentage change observed in all the above parameters has clearly indicated that SC 30 mg/kg produced an effect that was similar to diclofenac and methotrexate (Table-2).

The anti-arthritic changes observed in the joint tissue has also shown that SC has produced significant reversal of joint damage compared to diclofenac and methotrexate. The anti-arthritic effect is also confirmed by favorable radiological & histopathological changes. Histopathologically there was marked reduction in cellular infiltration, cartilage damage and improvement in synovial space (Fig. 14). There was reduction in soft tissue swelling and improvement in joint space seen radiologically (Fig.13).

When compared to the results of previous study conducted on FCA induced arthritis using SI seed ethanolic extract, the effect of SC was found to be superior in our study. The mean difference in paw volume by 800 mg/kg of the Sesamum indicum seed extract was 0.77 (11) and that by SC in our study was 0.80 which shows that SC produces similar effect as that of crude extract. But the crude extract contains multiple phytochemicals which can produce effects other than anti-rheumatoid effect.

Some of the earlier studies have shown different pharmacological actions of Sesamum indicum. The anti-inflammatory and antinociceptive effects had been evaluated for sesame oil and sesamin in both rats and mice. The conclusion drawn from this study was that sesamin could be used in the treatment of pain and inflammation (24).

As SC reduced paw volume, IL6, TNF-α, CRP, ESR, body temperature and improved hemoglobin, spontaneous activity and body weight comparable to diclofenac and better than methotrexate due its favorable effect on hemoglobin it can be further evaluated for its use in RA in humans.

**CONCLUSIONS:** Sesamin complex has anti-rheumatoid activity in FCA induced arthritis in rats at 15mg and 30 mg/kg and the effect produced by 30 mg/kg is equivalent to diclofenac and methotrexate. Sesamin complex was found to be safer than diclofenac and methotrexate in terms of hematological, biochemical and physical parameters. Sesamin complex can be further investigated for its use as a safer alternative to diclofenac and methotrexate in the treatment of rheumatoid arthritis in humans.

**ACKNOWLEDGEMENT:**
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**CONFLICT OF INTEREST:** The authors declare that there is no conflict of interest.

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