

Evaluation of anti-rheumatoid effect of hydroalcoholic extract of Sesamum indicum seeds in Freund's complete adjuvant induced arthritis in Wistar Albino rats

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Abstract

Background: Rheumatoid arthritis (RA) is one of the leading causes of joint problems in the world. It will affect the both genders. The present study aimed to evaluate the anti-rheumatoid effect of hydroalcoholic extract of Sesamum indicum seeds in Freund's complete adjuvant induced arthritis in Wistar Albino rats

Materials and Methods: Total of 30 rats were divided into 5 groups each of 6 rats. G-I (Normal Saline 0.9%), G-II Freund's adjuvant (0.1 ml/0.5%), G-III Dexamethasone (0.5mg/kg), G-IV Sesamum indicum (400mg/kg) and G-V Sesamum indicum (800mg/kg). Drugs were given to respective groups for 28 days. Blood samples were collected under anesthesia. Serum was separated and used for the estimation of rheumatoid factor, C-reactive protein and erythrocyte sedimentation rate on 1st, 7th, 14th and 28th day.

Results: Control groups showed significant difference compared to other groups. g-II showed significant

difference compared g-III, IV and V. High dose of test drug showed similar results like standard drug. Low dose of test drug showed less effect than high dose.

Conclusion: Hydroalcoholic extract of Sesamum indicum seeds significantly reduced the serum rheumatoid factors.

Keywords: Hydroalcoholic extract, C-reactive protein, Dexamethasone, Rheumatoid arthritis, Rheumatoid factor, Sesamum indicum

Introduction

Arthritis is an autoimmune disease characterized by the inflammation in joints and other parts of the body¹⁻³. It mainly affect the joints as disease progress can affect the organs, blood vessels, skin and other parts of the body. Auto antibody will produce in the body which affects the cells by releasing lytic enzymes and inflammatory mediators. These reactions mainly seen in the joints and produce the pain, inflammation and restrict the movements^{4,5}. But based on the pathology arthritis are mainly classified into two types. One is

inflammatory and second one is non-inflammatory arthritis. Rheumatoid arthritis is comes under inflammatory and osteoarthritis is called non-inflammatory arthritis⁶. In the rheumatoid arthritis symptoms will be restricted to the joint. It will affect the small, medium and large joints in the body. In the present medical practice various drugs are used to treat the arthritis. The major classes of drugs are non-steroidal anti-inflammatory drugs, steroids, immunosuppressants and disease modifying anti-rheumatoid drugs^{7,8}. To reduce progression of disease may require life time use of drugs. Long term use of these drugs will produce the severe adverse effects and some time it may contraindicated to use. These limitations can overcome with use of medical plants. According to Ayurveda Sesamum indicum seeds are used to treat various disorders. It used as mono or with combination of other drugs to treat the inflammatory disorders, reduce the pain, suppress the immune system, ulcer, asthma and central nervous disorders⁹⁻¹². From the literature the present study aimed to evaluate the anti-rheumatoid effect of hydroalcoholic extract of Sesamum indicum seeds in Freund's complete adjuvant induced arthritis in Wistar Albino rats

Materials and Methods

Animals

Wistar Albino rats weighing 150-200 g were obtained from Central Animal House. They were fed on standard rat pellet diet and water was provided in feeding bottle. All the animals were maintained under standard laboratory conditions temp 24 C and humidity 60-70%.

Collection of Sesamum indicum seeds and preparation of extract

Sesamum indicum seeds were purchased from local area. They were dried under sunlight. Dried seeds were made into power form. The power was soaked for 7

days in hydroalcoholic liquid (50:50). After 7 days the liquid was filtered and evaporated on water bath. Finally thick dark brown extract was collected and stored for further study.

Groups

Group-I: Normal Saline (0.9% orally/10ml/kg)

Group-II: Freund's complete adjuvant (0.1 ml/0.5%)

Group-III: Dexamethasone (0.5mg/kg)

Group-IV: Sesamum indicum (400 mg/kg)

Group-V: Sesamum indicum (800 mg/kg)

Procedure

Rats were induced arthritis by injecting 0.1 ml of 0.5% of Freund's complete adjuvant into the tibio tarsal joint of the left high paw. All the groups were given inducing agent except control group. Drugs were treated for 28 days. Blood samples were collected from all the rats on 1st, 7th, 14th and 28th day for the estimation of erythrocyte sedimentation rate, C-reactive protein and rheumatoid factor¹³.

Statistical analysis

The data was expressed in mean and standard deviation. Statistical Package for Social Sciences (Version 16.0) used for analysis. One ANOVA (Post hoc) followed by Dunnett t test applied to find the statistical significant between the groups. P value less than 0.05 ($p < 0.05$) considered statistically significant at 95% confidence interval.

Results

Group-I showed significant difference compared to other groups in all time periods ($p < 0.05$). Group-II showed significant increased in ESR, CRP and RAF compared to all other groups ($p < 0.05$). Group-III showed significant reduction in the ESR, CRP and RAF on 1st, 7th, 14th and 28th days compared to I, II and IV groups ($p < 0.05$). Group-III not showed significant difference compared to group-V ($p > 0.05$) in all time

periods. Low dose of seed extract showed less effect compared to high dose which is statistically significant. High dose of seed extract and standard drug showed similar effects in ESR, CRP and RAF in four time periods (Table-1, 2, 3 and Graph-1, 2, 3)

Discussion

In this study Freund's complete adjuvant used for the experimentally induced arthritis in Wistar Albino rats. Long acting steroid dexamethasone is used as a standard drug. Two doses of plant seed extract used for analysis. Dexamethasone is a known steroid and is used in the treatment of various immune and inflammatory conditions. Xi Chen et.al., study showed significant reduction in arthritis symptoms compared to control group¹⁴. Girija P et.al., in this study administration of dexamethasone reduced the levels of inflammatory mediators levels compared to other groups. The present study also showed significant reduction in ESR, CRP and RAF levels compared to other groups. Present study results showed similar effect like above mentioned studies¹⁵. *Sesamum indicum* an oldest plant which is first recorded in the Pedaliaceae family. The plant seeds showed various medical uses. It mainly used for anti-inflammatory, immunosuppressant, migraine, analgesic anti-oxidant, anti-cancer and hypolipidemic. It also showed hepatoprotective and kidney protective by anti-inflammatory and anti-oxidant effect. The present study used hydroalcoholic extract of *Sesamum indicum* seeds to screen the anti-rheumatoid effect in experimentally induced arthritis. The study showed significant effect compared to other groups. Hemshekhar M et.al., study showed the anti-inflammatory activity of *Sesamum indicum* in FAC induced arthritis. The study results concluded that administration of plant extract prevented the arthritis changes compared to other groups. In the study they

isolated sesamin from seed and used^{16,17}. In the present study also showed similar effect. High dose produced more effect compare to low dose. Hydroalcoholic extract also may contain same phytochemicals which is responsible for the results. High dose long term will produce better effect compared to low dose of hydroalcoholic extract of *Sesamum indicum*.

Conclusion

The study results concluded that hydroalcoholic extract of *Sesamum indicum* seeds showed significant effect experimentally induced arthritis. Further studies required to find the active principal and its mechanism of action in the prevention of disease progression.

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Legends Tables and Figures

Table 1: Comparison of erythrocyte sedimentation rate between the groups at same time period

Groups	Day-1 ESR (mm/hour) (MEAN±SD)	Day-7 ESR (mm/hour) (MEAN±SD)	Day-14 ESR (mm/hour) (MEAN±SD)	Day-28 ESR (mm/hour) (MEAN±SD)
Group-I	8.67±1.56	8.56±1.93	8.72±1.89	8.93±1.45
Group-II	29.87±1.78*	37.67±2.14*	41.34±2.97*	37.45±2.45*
Group-III	14.45±1.93* [#]	14.24±1.78* [#]	17.89±1.34* [#]	15.36±1.03* [#]
Group-IV	20.45±1.23* ^{#,\$}	23.89±0.93* ^{#,\$}	29.45±1.76* ^{#,\$}	26.56±1.22* ^{#,\$}
Group-V	15.17±1.94* ^{#,l}	15.34±1.04* ^{#,l}	18.34±2.67* ^{#,l}	116.04±2.45* ^{#,l}

(*p<0.05 significant compared group-I with other groups, [#]p<0.05 significant compared group-II with other groups, ^{\$}p<0.05 significant compared group-III with other groups, ^lp<0.05 significant compared group-V with other groups)

Graph 1: Comparison of erythrocyte sedimentation rate between the groups at different time periods

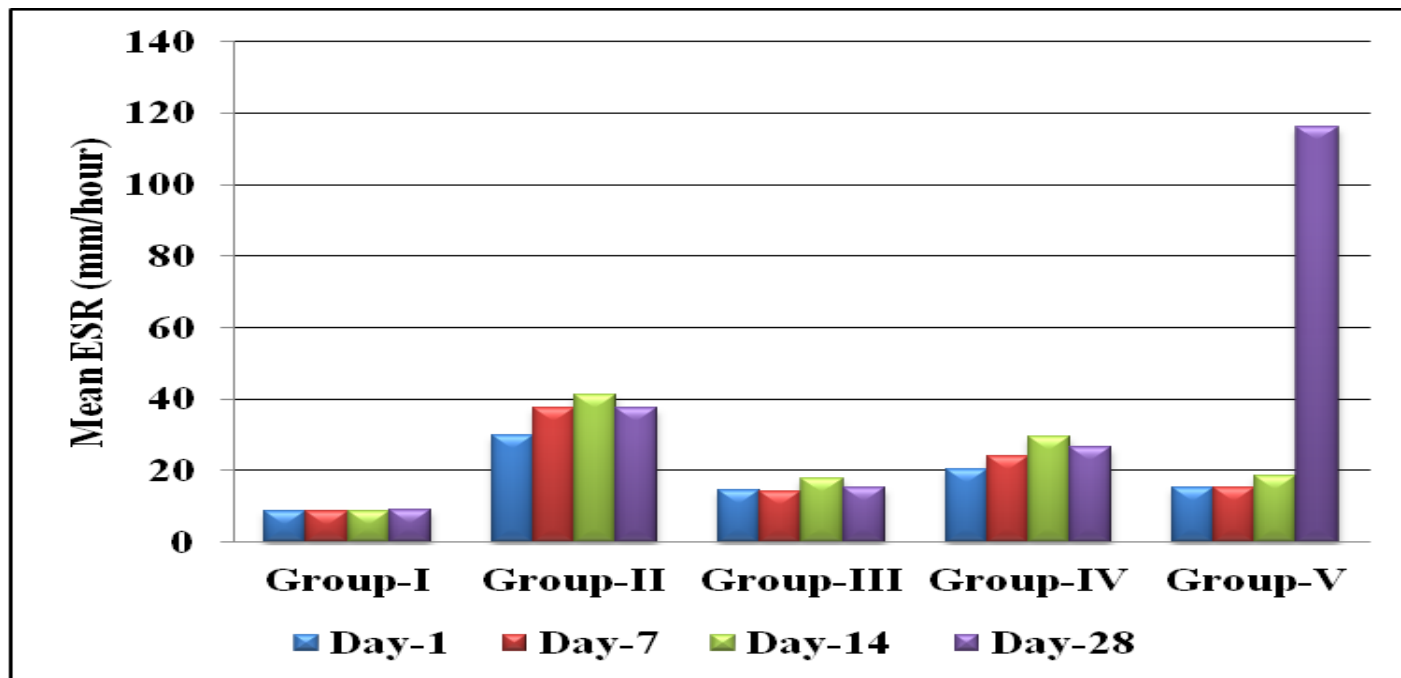


Table 2: Comparison of C-reactive protein levels between the groups at same time period

Groups	Day-1 CRP (mg/dl) (MEAN±SD)	Day-7 CRP (mg/dl) (MEAN±SD)	Day-14 CRP (mg/dl) (MEAN±SD)	Day-28 CRP (mg/dl) (MEAN±SD)
Group-I	5.73±0.84	5.34±0.56	5.23±0.95	5.34±0.45
Group-II	17.93±0.23*	20.45±0.35*	22.45±0.78*	24.31±0.93*
Group-III	8.43±0.56* [#]	11.56±0.12* [#]	12.34±0.18* [#]	10.34±0.45* [#]
Group-IV	12.56±0.34* ^{#,\$}	14.10±0.34* ^{#,\$}	16.23±0.45* ^{#,\$}	16.29±0.84* ^{#,\$}
Group-V	8.89±0.29* ^{#,l}	11.83±0.45* ^{#,l}	12.84±0.84* ^{#,l}	10.67±0.94* ^{#,l}

(*p<0.05 significant compared group-I with other groups, [#]p<0.05 significant compared group-II with other groups, ^{\$}p<0.05 significant compared group-III with other groups, ^lp<0.05 significant compared group-V with other groups)

Graph 2: Comparison of C-reactive protein levels between the groups at different time periods

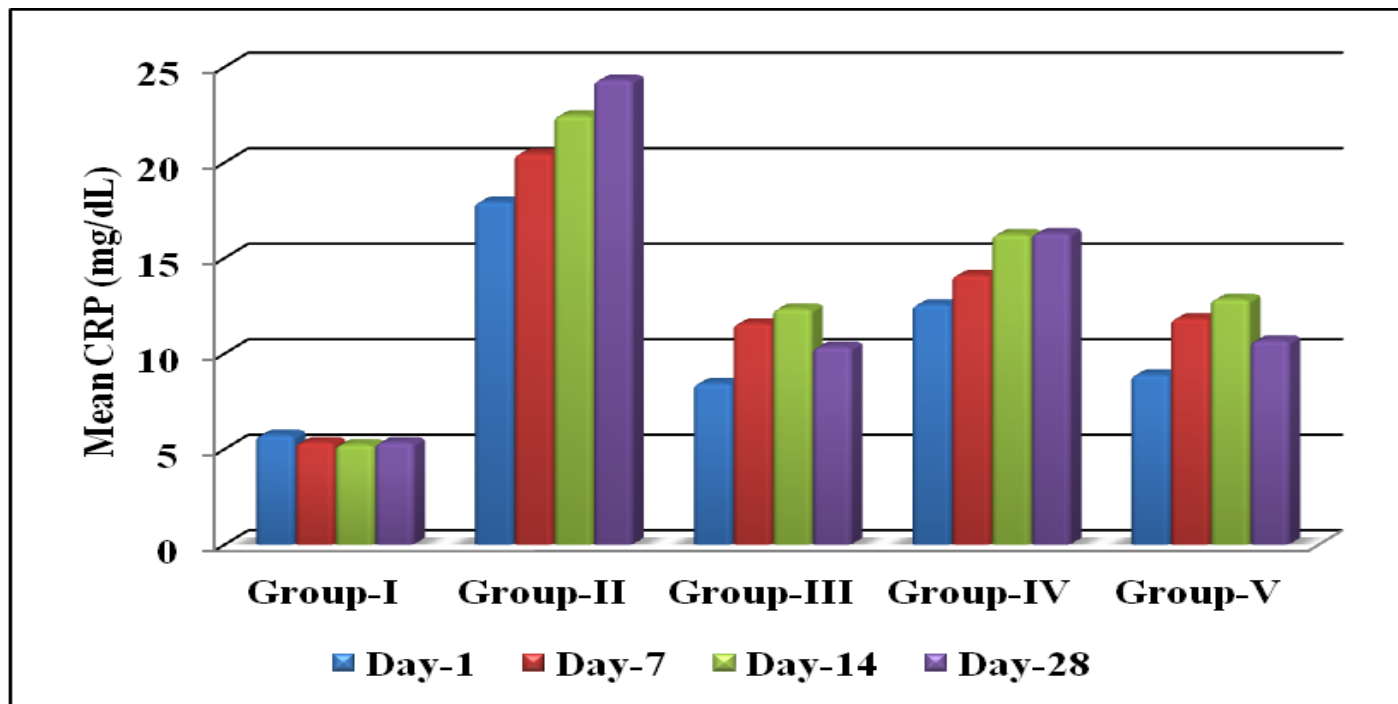


Table 3: Comparison of rheumatoid factor between the groups at same time period

Groups	Day-1 RAF (IU/dl) (MEAN±SD)	Day-7 RAF (IU/dl) (MEAN±SD)	Day-14 RAF (IU/dl) (MEAN±SD)	Day-28 RAF (IU/dl) (MEAN±SD)
Group-I	17.89±12.89	17.34±12.67	17.23±11.98	17.10±12.45
Group-II	163.89±12.78*	195.45±13.46*	200.45±12.45*	204.78±11.78*
Group-III	135.33±10.45* [#]	149.89±15.45* [#]	98.56±14.90* [#]	76.89±15.45* [#]
Group-IV	145.95±14.34* ^{#,§}	176.95±12.56* ^{#,§}	114.95±11.78* ^{#,§}	98.24±13.56* ^{#,§}
Group-V	139.45±11.45* ^{#,1}	154.89±11.45* ^{#,1}	100.29±14.89* ^{#,1}	79.45±11.89* ^{#,1}

(*p<0.05 significant compared group-I with other groups, [#]p<0.05 significant compared group-II with other groups, [§]p<0.05 significant compared group-III with other groups, ¹p<0.05 significant compared group-V with other groups)

Graph 3: Comparison of rheumatoid factor between the groups at different time periods

