## REPORT

# ANTI-INFLAMMATORY AND ANALGESIC ACTIVITIES OF ETHANOLIC EXTRACT OF *SPHAERANTHUS INDICUS* LINN

**ADAM ALI, SYED BAQIR SHYUM NAQVI, SHAHNAZ GAUHAR* AND REHANA SAEED**

*Department of Pharmaceutics, Faculty of Pharmacy, University of Karachi, Karachi, Pakistan*

**ABSTRACT**

The aim of the present study was to evaluate the intensity of the anti-inflammatory and analgesic activities of *Sphaeranthus indicus* on albino mice and rat of either sex. The flowers of *S. indicus* are an important herb used in folk eastern medicines. In this study, the ethanolic extract of *S. indicus* in doses of 300 and 500 mg/kg was used. Anti-inflammatory activity was evaluated by measuring the mean decrease in hind paw volume after the sub planter injection of carrageenan. The analgesic activity was tested against acetic acid induced writhing response using albino rats. Result of the study shows that at the end of one hour the inhibition of paw edema was 42.66 and 50.5% respectively and the percentage of protection from writhing was 62.79 and 68.21 respectively. *S. indicus* possesses several important pharmaceutical and pharmacological properties. The current study describes that flower of *S. indicus* has significant anti-inflammatory and analgesic properties. Conclusion of the study is that this herbal medicine can be used as an alternative therapy for the treatment of minor to moderate types of inflammation and as a painkiller

**Keywords**: Anti-inflammatory, analgesic, *Sphaeranthus indicus.*

## INTRODUCTION

Medicinal herbs play an important role in health care throughout the world especially in non-industrial continents such as Africa, South America and parts of Asia. However, even in many industrialized countries, numbers of traditional herbs are still in use by a majority of people for minor to moderate everyday ailments for self-medication (Christopher Hobbs, 1998).

*Sphaeranthus indicus* is indigenous to Indo-Pak subcontinent and distributed through out the Pakistan. Due to immense medicinal properties, it is very commonly used in Tibbi and Ayurvedic system of medicines (Kirtikar and Basu, 1984).

Chadha (1976) made a detailed study of *S. indicus* Linn. All parts of plant find medicinal uses. The juice of the plant is styptic and said to be useful in liver and gastric disorders. The paste of herb made with oil is applied in itch. The herb has a bitter sharp flavor with bitter taste. It increases the appetite, enriches the blood, cools the brain and gives luster to the eye. It is useful in jaundice and scalding of urine as a diuretic. The plant with cumin is stomachic and with honey, it is prescribed for cough. It was further reported by Chadha (1976). The whole plant can be used as fish poison.

Kirtikar and Basu (1918) reported that flowers are highly esteemed as alterative, depurative, cooling and tonic. They are also used as blood purifiers in skin diseases. The flowers are employed internally as well as externally in chronic skin, ulcerations, irritation, scabies, ringworms and other eruptive skin ailments due to blood disorders (Kirtikar and Basu, 1918).

Kirtikar and Basu (1918) reported that the grounded bark mixed with whey is a valuable remedy for piles. Hindus used to make a kind of confection of the young plant by rubbing it up with clarified butter, flour and sugar. A portion of this taken daily is said to be a good tonic and prevents the hair turning white or falling off.

Dhar *et al* (1968) reported that 50% ethanolic extracts of *S. indicus* Linn exhibited a wide range of biological activity. The antibacterial activity was determined against *B.subtilis, Staph.aureus, Sal.typhi, E.coli* and *Mycobacterium tuberculosis.* The plant extract also exhibited anticancer activity.

The aim of the present study was to investigate the anti-inflammatory and analgesic activity of ethanolic extract of flowers of *Sphaeranthus indicus* because now a day the use of herbal medicine is becoming more popular in both developing and developed countries.

## MATERIALS AND METHODS

### *Material*

Acetylsalicylic acid (Aspirin, 300mg tablets), carrageenan, acetic acid, saline, distilled water, plethysomometer (7150 Ugo Basile), open field apparatus, head tip box, traction test rod, 1 ml syringes,

**Corresponding author:* e-mail: shahnaz_gauhar@yahoo.com

needles, feeding tube, vials, eclectronic balance, stop watch, gloves and other laboratory glass wares.

***Plant material and extract preparation***

The flowers of the plant *Sphaeranthus indicus* were purchased from (Hakeem & Company, Herb Medicine Supplier, Allah Bakaya Street, Jodia bazaar, Karachi-74000, Pakistan and were identified by Prof. Dr. Ghazala H. Rizwani, Dean, Faculty of Pharmacy, University of Karachi. The powdered flowers were soaked in 50% ethanol for 10 days with occasional shaking and were concentrated using a rotary vacuum evaporator under reducing pressure and controlled temperature at 40° C. A semi-solid mass of dark brown color obtained, which was then lyophilized and finally brown powder of *S. indicus* were obtained.

***Animals***

Albino mice and rats of either sex obtained from the (reliable animals shop) local market of Karachi, Pakistan and were used to determine the anti-inflammatory and analgesic activities. Mice weighing 25-34gms and rats 200-300gms were used. Animals were kept in colony cages (Five animals in each group) with access to food and water. They were maintained in a climate and light controlled room ($30^oC\pm 1^oC$ 12/12hours light/dark cycle) at least 7 days before the experiments.

***Anti-inflammatory activity***

Anti-inflammatory activity was determined by measuring the mean decrease in hind paw volume after the sub planter injection of carrageenan in comparison with control group (Winter *et al*., 1962). The animals were injected with 0.1 ml 1% carrageenan in 0.9% saline in the right hind foot under the planter aponeurosis. Rats were divided into 4 groups (i-e Group A for control, Group B and Group C for 300 and 500gms/kg oral doses of crude extract respectively, and Group D for standard). Each group comprised of five (5) animals, weighing 200-300gms. Acetylcylic acid (Aspirin) 300 mg/kg oral dose was used as standard. The working solution was prepared by dissolving crude extract and the acetyl salicylic acid in distilled water separately and then administered orally 30-40 minutes before carrageenan injection.

The hind paw volume was quantitated in terms of millimeters using plethysmometer, immediately before carrageenan injection. The inflammation was determined after one hour of carrageenan injection up to seven hours. The percent inhibition of edema was calculated for each group with respect to its vehicle-treated control group. The anti-inflammatory activity was calculated by the following relationship (Palanichamy and Nagarajan 1990).

$$\frac{A-B}{A}\times 100$$

Where

A = mean increase in paw volume of control
B = mean increase in paw volume of drug-treated

***Analgesic activity***

The modified form of Koster *et al*, (1959) method was used to evaluate the analgesic activity of *S. indicus* using mice. According to this method writhes were induced by intra-peritoneal administration of acetic acid solution 10ml/kg. Thirty minutes prior to the administration of acetic acid, the animals were treated orally with the test substance. Number of writhes was counted for 30 minutes immediately after acetic acid administration. A reduction in number of writhing as compared to the control animals was considered as evidence for the presence of analgesia and expressed as percent inhibition of writhing.

Mice were divided into 4 groups, 5 animals each (i-e Group A for control, Group B and C for 300mg/kg and 500mg/kg oral doses of crude extract respectively, and Group D for standard). The 300mg/kg oral dose of acetylcylic acid (Aspirin) was used as reference compound .The *S. indicus* extract and the acetyl salicylic acid both were diluted in distilled water and then given to the animal by oral route. Where as the control animals were treated with the same volume of saline as the crude extract of *S. indicus*.

## STATISTICAL ANALYSIS

Values for observations were expressed as mean after drug administration ± SEM. The significance of difference between means was determined by Dunnitt's *t-test* and values of $p<0.05$ were considered significant and $p<0.01$ as highly significant. All stastical procedures were performed according to the method of Alcaraz and Jimenez, 1989.

## RESULTS

Herbal medicines derived from plant extracts are being increasingly utilized to treat a wide variety of clinical diseases, though relatively little knowledge about their mode of action is available. There is a growing interest in the pharmacological evaluation of various plants used in traditional systems of medicine (Ratheesh and Helen, 2007).

The present study was carried out to evaluate the anti-inflammatory and analgesic activity of *S. indicus o*n experimental animals.

***Anti-inflammatory activity***

Anti-inflammatory effect of crude extract of *S. indicus* and aspirin on carrageenan-induced rat paw edema was evaluated (table 1). Test extract produced significant inhibition of paw edema as compared to the control.

***Analgesic activity***

Analgesic activity of the crude extract of *S. indicus* and aspirin was evaluated on acetic acid writhing in mice (table 2). Results were found to be highly significant in comparison to the control.

## DISCUSSION

The inflammatory diseases are much more common and create a major health problem among people in which arthritis is the number one crippling disease all over the world. Today it is so wide spread that one in six people and one in three families are affected by it. It is estimated that arthritis affects approximately 80% of people in United States (Eidelson, 2004) and about 60% of population in Pakistan and India (Ahmed 2004).

There is growing evidence suggesting the therapeutic potential of plants. Herbal medicines are again becoming more popular throughout the world (Wohlmuth, 2002).

Inflammation is considered as a primary physiologic defence mechanism that help body to protect itself against infection, burn, toxic chemicals, allergens or other noxious stimuli an uncontrolled and persistent inflammation may act as an etiologic factor for many of these chronic illness (Kumar *et al.*, 2004).

The search for non-toxic and ant-inflammatory substances of natural origin from medicinal plants has long been expected because most common and serious draw back of all analgesic and anti-inflammatory drugs is that they cause serious acidity or severe burning problem which limits there use. Where as the drugs of plant origin have no side effects or only marginal.

Keeping in view the use of *S. indicus* Linn in folk medicine and eastern system of medicine including availability of some commercial products in sub-continent (India & Pakistan) attempt has been made to study the anti-inflammatory and analgesic activity of extract of flowers of *S. indicus*.

The anti-inflammatory activity of the extract of flowers of *S. indicus* and aspirin was estimated on carrageenan-induced rat paw edema (table 1). Result indicated a decrease in hind paw volume of drug tested animals as compared to the control group. The animals were tested in two different doses as 300mg/kg and 500mg/kg. Significant dose dependent anti-inflammatory activity at both doses was observed.

When the extract tested animals were compared with standard aspirin 300mg/kg dose the result shows less anti-inflammatory activity. Further, it was noted that anti-inflammatory activity of crude extract of flowers of *S. indicus* and aspirin was maximum after one hour and then

**Table 1**: Anti-inflammatory effects of crude ethanolic extract of *S. indicus* and Aspirin on carrageenan-induced rat paw edema.

| Treatment | Dose mg/ kg orally | Before carra-geenan | Mean paw volume ± SEM (ml) | | | | | | | Inhibition in edema | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | +1h | +2h | +3h | +4h | +5h | +6h | +7h | +1h | +2h | +3h | +4h | +5h | +6h | +7h |
| Control | 0.5ml saline orally | 3.051 | 3.651± 0.061 | 3.676± 0.042 | 3.693± 0.041 | 3.712± 0.035 | 3.731± 0.046 | 3.759± 0.039 | 3.776± 0.047 | | | | | | | |
| Crude ethanolic extract of Sphaeranthus indicus | 300 mg/kg orally | 3.032 | 3.376± 0.021 | 3.382± * 0.029 | 3.390± * 0.031 | 3.409± * 0.033 | 3.432± * 0.035 | 3.460± * 0.027 | 3.469± * 0.029 | 42.66 | 44.0 | 44.23 | 42.96 | 41.17 | 39.54 | 39.72 |
| | 500 mg/kg orally | 3.022 | 3.319± 0.026 | 3.349± * 0.034 | 3.367± * 0.032 | 3.391± * 0.036 | 3.408± * 0.036 | 3.435± * 0.025 | 3.461± * 0.027 | 50.50 | 47.68 | 46.26 | 44.17 | 43.23 | 41.66 | 39.44 |
| Aspirin | 300 mg/kg orally | 3.011 | 3.276± 0.039 | 3.321± * 0.025 | 3.361± ** 0.024 | 3.369± * 0.019 | 3.387± ** 0.016 | 3.407± * 0.014 | 3.415± ** 0.014 | 55.83 | 50.40 | 45.48 | 45.83 | 44.70 | 44.06 | 44.27 |

**Table 2**: Effects of crude ethanolic extract of *Sphaeranthus indicus* and Aspirin on acetic acid induced writhing in mice

| Treatment | Dose mg/kg orally | Mean no. of writhes ± S.E.M. | Inhibition (%) |
|---|---|---|---|
| Control | 0.5ml saline orally | 129 ± 4.7097 | 00 |
| Crude ethanolic extract of *Sphaeranthus indicus* | 300mg/kg orally | 48 ± 2.2516* | 62.79% |
| | 500mg/kg orally | 41 ± 1.9523** | 68.21% |
| Aspirin | 300mg/kg orally | 37 ± 2.3126** | 71.31% |

Result based on (Mean ± S.E.M.) N = 5, Significance with respect to control (* = Significant result, ** = Highly significant result)

started to decrease after four hours of administration. The present results are in confirmation with the work of Farzana *et al* (2006), Achints *et al* (2005), Ranajit *et al* (2007) with the difference that the plant extracts used by them were different otherwise similar anti-inflammatory activity was noted in all the plant extracts.

The analgesic activity of flowers of *S. indicus* was determined by writhing test. In acetic acid induced writhing test aspirin 300mg/kg orally was used as reference compound (table 2). The result showed that in control animal mean number of writhes induced by intra-peritoneal ingestion of acetic acid was 129 writhes which was reduced to 48 and 41 in animals with 300mg/kg and 500mg/kg oral doses of the test extract respectively. The results of writhes test proved highly significant when compared with aspirin that produced 37 writhes. The percentage inhibition of writhes with two doses of crude extract of *S. indicus* was 62.79 by 300mg/kg and 68.21 by 500mg/kg where as with aspirin it was 71.31%.

The present results are in confirmation with the work of Howlader *et al* (2005), Ranajit *et al* (2007) reported 52.43% inhibition of writhing response. Achints *et al* (2005) reported significant inhibition of paw edema by 41.09% and 30.15% respectively which was comparable to that of standard drug phenylbutazone (42.15%).

## CONCLUSION

The work result and literature survey focuses on *S. indicus* indicating the herb to be beneficial for the treatment of number of illness and disorders. The result indicates that *S. indicus* have a significant anti-inflammatory and analgesic efficiency in comparison to aspirin (NSAID) that can be hard on the stomach therefore special buffered and enteric coated forms have been developed.

Conclusion of the present study is that the *S. indicus* have potential to cure disease and would prove to be a delightful herbal plant for the treatment of number of infection, inflammation, fever and as well as in neurological activity.

## REFERENCES


Alcaraz MJ and Jimenez MJ (1989). Anti-inflammatory compound from Sideritis javalambrensis n-hexane extract. *J. Nat. Prod.*, **52**: 1088-1091.

Ahmed.M, Saeed F. and Anwer M. (2004). An overview "Arthritis" How to cure and care 1st Edition BCC & T Press, University of Karachi. Pakistan.

Achinto Saha, Kawshik Kumar Chowhury, Sitesh Chandra Bachar, Surash Chandra Roy and Joydeb Kumar Kundu. (2005). Anti-inflammatory, analgesic and diuretic activity of *Pohygorum Lanatum* Roxh. *Pak. J. Pharm. Sci.*, **18**: 13-17.

Christopher Hobbs (1998). From Native American Panacea to Modern Phytopharmaceutical Echinacea Pharmacy in History, obtained from Health World Online, California, US.

Chadha YR (ed.) (1976). The Wealth of India, Published by the Publication and Information Directorate, CSIR, New Dehli, Vol. X, pp. 591

Dhar ML, Dhar MM, Dhawan BN, Mehrotra BN and Rag C (1968). Screening of medicinal plants for biological activity, Part-1, *Ind. Jour. Exp. Bio.*, **I**: 232.

Eidelson SG (2004). Arthritis, Ostesarthritis, Rheumotoid arthritis, Anky losing spondylitis-spinal stenosis. Common spinal disorders: In save your aching back and neck. A patient's guide 2nd Edition. Spine Universe Founder Bocaraton FL, USA, pp.3-28.

Farzana Sadaf, Saima Hashmi, and Tariq Shrafatullah. (2006). Anti Inflammatory and antinociceptive activity in herbal drug-Aujaie. *Pak. J. Pharmcol.*, **23**: 1-5.

Howlader MAB, Bachar SC, Begum F and Rouf ASS (2005). Diuretic and analgesic effect of the methanolic extract of *Phaenix sylvestris* roots. *Pak. J. Pham. Sci.*, **19**: 330-332.

Kirtikar KR and Basu BD (1984). Indian Medicinal plants. 2nd Edition Vol. II, Dehli., pp.1347-1348.

Kirtikar KR and Basu BD (1918). Indian Medicinal Plants, published by Lalit Mohan Basu, 49, Leader Road Allahabad, India, Vol. I & II, pp.715

Koster R, Anderson M and De Bear EJ (1959). Acetic acid for analgesic screening. *Fed. Proceed.*, **18**: 412-416.

Kumar V, Abbas AK and Fausts N (2004). Robbins and Cortran (eds.) Pathological basis of disease. 7th Edition Elsevier Saunder's Philadelphia, Pennsylvania, pp.47-86.

Nadkarni KM (1976). Indian Materia Medica, published by Bombay Popular Prekashan, Vol.1, Bombay, p.1290.

Palanichamy S and Nagarajan S (1990). Analgesic activity of Cassia Alanta leaf extract and Kaempferol-3-0-sophoroside. *Fitoterapia*, **LXI**: 44-47.

Ratheesh M and Helen A (2007). Anti-inflammatory activity of *Ruta graveolens* Linn on carrageenan induced paw edema in wistar male rats. AJB; **6**: 1209-1211.

Sutradhar RK, Akm M. Rahman, Ahmed M., Chandra SB., Saha A. and Roy TG. (2007) Anti-inflammatory and analgesic alkaloid from *Sida cordifolia* Linn. *Pak. J. Pham. Sci.*, **20**: 185-188.

Winter CA, Risley EA and Nuss GW (1962). Carrageenan induced edema in hind paw of the rat as an assay for anti-inflammatory drugs. *Proc. Soc. Exp. Biol. Med*., **111**: 544-547.

Wohlmuth H, Oliver C and Nathan PJ (2002). A review of the status of western herbal medicine in Australia. *J. Herbal Pharmacother*., **2**: 33-46.