

EVALUATION OF ANALGESIC, ANTI-INFLAMMATORY AND ANTIPYRETIC POTENTIAL OF METHANOL EXTRACT OF *SWERTIA CORYMBOSA* (GRISEB.) WIGHT EX C.B. CLARKE

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ABSTRACT

Objective: To evaluate the Analgesic, anti-inflammatory and antipyretic effects of methanolic extract of *Swertia corymbosa*, a folklore medicinal plant.

Methods: Methanol extract of *Swertia corymbosa* (100 and 200mg, body weight) was screened for analgesic (Eddy's hot plate and acetic acid induced writhing), anti-inflammatory (Carrageenan induced paw edema) and yeast induced hyperthermic in rats.

Results: Treatment with methanol extract of *S. corymbosa* showed significant ($P < 0.05$ and $P < 0.01$) and dose dependant increase in paw licking time in Eddy's hot plate method. In writhing test, extract was significantly reduced the number of writhes. A dose dependant and significant inhibition of edema was observed in carrageenan induced paw edema. Methanol extract at a dose of 200 mg/kg significantly reduction in elevated body temperature.

Conclusions: In vivo studies of *S. corymbosa* showed prominent antinociceptive, anti-inflammatory and antipyretic activities with ample safety profile and thus provided pharmacological base for the traditional uses of the plant in various painful conditions and pyrexia. Additional detail studies are required to ascertain its clinical application.

Keywords: Analgesic, Anti-inflammatory activity, Carrageenan-induced paw edema, Methanol extract, *Swertia corymbosa*

INTRODUCTION

Inflammatory response is a complex process mediated by a variety of signaling molecules released by nerve endings, mast cells, platelets and leukocytes. Some of these molecules and their precursors (prostaglandins, nitric oxide, adenosine deaminase and myeloperoxidase) are used as markers of inflammation [1]. Study of inflammation with carrageenan as a phlogistic agent has revealed increased vascular permeability, formation of exudate and a large number of cellular infiltrates a polymorphonuclear leukocyte [2].

Tumor necrosis factor alpha (TNF- α), an important inflammatory mediator, is a multifunctional cytokine that can regulate many cellular and biological processes, such as immune function, cell differentiation, proliferation, apoptosis, and energy metabolism. Furthermore, TNF- α can regulate the production of other pro-inflammatory cytokines, such as interleukin-6 (IL-6) and interleukin-1 (IL-1), to mediate and/or amplify their effects in peripheral organs [3]. During the acute inflammatory process, overproduction of TNF- α is crucial to the induction of inflammatory genes and the recruitment and activation of host immune cells [4, 5].

Non-steroidal anti-inflammatory drugs (NSAIDs) are used throughout the world for the treatment and management of inflammation, pain and fever. The use of NSAIDs, however, has not been therapeutically successful in all conditions of inflammation [6]. Moreover, adverse effects associated with NSAIDs can lead to ulcers and hemorrhage. As an alternative, plant based medicines are getting an increased therapeutics market share due to their mild action and fewer adverse effects. According to the World Health Organization nearly 80% of the world population prefers plant based drugs [7].

Swertia, an important genus of the family Gentianaceae, comprises about 250 species distributed in Euro-Asia, Africa and Madagascar and represented by 40 species in India [8]. *Swertia* species are widely used in Ayurvedic, Unani and Siddha systems of medicines [9]. In Chinese traditional medicine, *Swertia* are used for the treatment of hepatic, choleric, and inflammatory diseases [10]. *Swertia corymbosa* (Griseb.) Wight ex C.B. Clarke commonly known as Shirattakuchi by Irulars tribe. This plant has a long history of being used by Irulars and Paliyan ethnic medical practitioners have been used for medicinally as diarrhea, fever, jaundice, diabetic,

inflammation, anxiety, promote sleep, antiepileptic, nervous disorders, antidote and as a stomach wash in cattle[10,11]. In order to understand the highly acclaimed properties of *S. corymbosa* and its usage in the traditional systems of medicine, we have attempted to evaluate its analgesic, anti-inflammatory and antipyretic potential through the use several animal models and this will provide insight information with reference to their medicinal value.

MATERIALS AND METHODS

Plant material

The fresh aerial parts of *S. corymbosa* were collected during the month of August, 2012 from the Vellingiri hills, Coimbatore, Tamil Nadu, India. The plant species was authenticated by Botanical Survey of India, southern circle, Coimbatore, and voucher specimen (BUBH6144) was deposited in the Department of Botany Herbarium, Bharathiar University, Coimbatore. Freshly collected plant materials were cleaned to remove adhering dust and then dried under shade. The dried samples were powdered in a Willy Mill to 60- mesh size and used for solvent extraction.

Preparation of the extract

Coarse powder from the shadow dried aerial parts was extracted in soxhlet extractor with methanol. The extracted solvent was then filtered and was concentrated by using rotary evaporator until the solvent was completely removed and a dark brownish viscous residue was obtained (yield: 30.11% w/w with respect to dried plant material).

Animals

Swiss albino mice weighing 20-30g and Wistar albino rats of 200-250 g were used for the pharmacological studies. They were housed in a clean polypropylene cage and maintained under standard laboratory conditions (temperature $25 \pm 3^\circ\text{C}$ with dark/light cycle 12/ 12 h; 35-60 humidity). They were fed with standard pellet diet (VRK Nutritional solutions, Sangli, Maharastra) and water *ad libitum*. The studies were carried out at Nandha College of Pharmacy and Research Institute, Perundurai, Tamil Nadu, India. The experimental protocol was subjected to the scrutiny of the Institutional Animal Ethics Committee, and was cleared by same before beginning the experiment (688/02/C/CPCSEA).

Acute toxicity

Acute oral toxicity study was performed according to acute toxic class method. Swiss albino mice (n=6) of either sex selected by random sampling technique were used for acute toxicity study. The animals were kept fasting for overnight providing only water. Methanol extract of *S. corymbosa* (suspended in 0.5% carboxy methyl cellulose) were administered orally at a dose of 5, initially to separate groups of mice and mortality was observed for 3 days. If mortality was observed in 4/6 or 6/6 animals, then the dose administered was considered as toxic dose. However, if the mortality was observed in only one mouse out of six animals, then the same dose was repeated with higher doses such as 50, 300, 1000 and 2000 mg/kg. The general behaviors such as motoractivity, tremors, convulsions, straub reaction, aggressiveness, piloerection, loss of lighting reflex, sedation, muscle relaxation, hypnosis, analgesia, ptosis, lacrimation, diarrhea and skin colour were observed for the first one hour and after 24 h of test drug administration.

Analgesic activity

Hot plate method

The hotplate method was used to measure response latencies according to the method described by MacDonald et al [12]. For the experiments, four groups (n=6) of swiss albino mice (20–25g) were placed on a plate maintained at room temperature for 15 min. Group 1: normal control fed with 0.5% carboxyl methyl cellulose (vehicle) 10 ml/kg p. o., and Group 2: treated with pentazocine (25mg/kg, i. p.) whereas Groups 3 and 4 animals received methanol extract of *S. corymbosa* (100 and 200 mg/kg, p. o.). The animals were positioned on Eddy's hot plate kept at a temperature of 55±0.5°C. The time taken by the animals to lick the fore or hind paw or jump out of the place was taken as the reaction time. The latency was recorded at the time of 0 (just before any treatment) and 30, 60, 120, and 240 min after oral administration of samples and intraperitoneal administration of standard. A latency period of 15 s was observed to avoid damage to the paw. The percentage thermal pain stimulus relief or protection was determined by applying the formula:

$$\% \text{ Protection against thermal pain stimulus} = \frac{[(\text{test mean} - \text{control mean}) / \text{test mean}] \times 100}{\text{Acetic acid-induced writhing}}$$

The test was carried out using the method of Siegmund et al [13]. This method was used to preferentially evaluate possible peripheral effects of methanol extract of *S.corymbosa* as analgesic substance. Four groups of swiss albino male mice (n = 6) were fasted overnight prior the start of the experiment, and water *ad libitum*. The peripheral analgesic drug, indomethacin was used as a positive control. Group 1 received the vehicle distilled water (10 ml/kg, p.o.) and group 2 was treated with indomethacin (25 mg/kg, p.o.). Whereas groups 3 and 4 animals were orally administered with

methanol extract of *S.corymbosa* at doses of 100 and 200 mg/kg. Thirty minutes after treatment, the mice were injected (i.p.) with 0.1 ml of 1% acetic acid solution to induce the characteristic writhings. After 5 min, the mice were placed in an observation box and the number of writhes in a 10 min period was counted. The response of the extract and indomethacin treated groups were compared with those of animals in the control group.

Anti-inflammatory activity

Carrageenan induced paw edema

For the experiment, the male wistar rats (120–150 g) were divided into four groups (n = 6). The animals were fasted overnight prior to the start of the experiment, and water *ad libitum*. The first group received distilled water (10 ml/kg, p.o.), while the second group was treated with indomethacin (25 mg/kg, p.o.). The third and the fourth groups were administered with the methanol extract of *S. corymbosa* (100 and 200 mg/kg, p.o. respectively). Acute inflammation was produced by the subplantar administration of 0.1 ml of 1% carrageenan (in 1% CMC w/v) in the right hind paw of the rats. The vehicle, extracts and the standard drugs were administered 60 min prior to the injection of the phlogestic agent. The volumes of edema of the injected and the contralateral paws were measured at 1, 2, 3, 4, and 5 h after the induction of inflammation. The inflammation was measured by using an electronic vernier caliper (CD-6 CSX; Digimatic caliper, Mitutoyo, Japan) and calculates the percentage of paw edema inhibition [14].

Yeast -induced hyperpyrexia in rats

Before experimentation rectal temperature of rats were recorded by inserting a well lubricated bulb of a thermometer in the rectum. Hyperpyrexia was induced in rat by subcutaneous injection of 10ml/kg, b.w of a 15% aqueous suspension of brewer's yeast in the back below the nape of the rat. Pre-drug control temperatures were taken at 24 h after the yeast injection to determine the pyretic response of yeast. Methanol *S. corymbosa* extract (100 and 200mg/kg, b.w) and paracetamol (150mg/kg, b.w) served as the reference drug given orally 24 h after the yeast injection. The temperatures were recorded at 1-6 h after the drug treatment [15, 16].

RESULTS AND DISCUSSION

The methanol extract of *S. corymbosa* was evaluated for acute toxicity in mice. The extract did not alter the general behaviors and failed to produce any mortality even at the highest dose (2,000 mg/kg, p.o.) studied after 3 days and found to be safe. Based on acute toxicity, 2 doses i.e., 100 and 200 mg/kg were used for further pharmacological studies.

The analgesic activity of methanol extract of *S. corymbosa* assessed using hot plate test in swiss albino mice and the results are presented in Fig. 1.

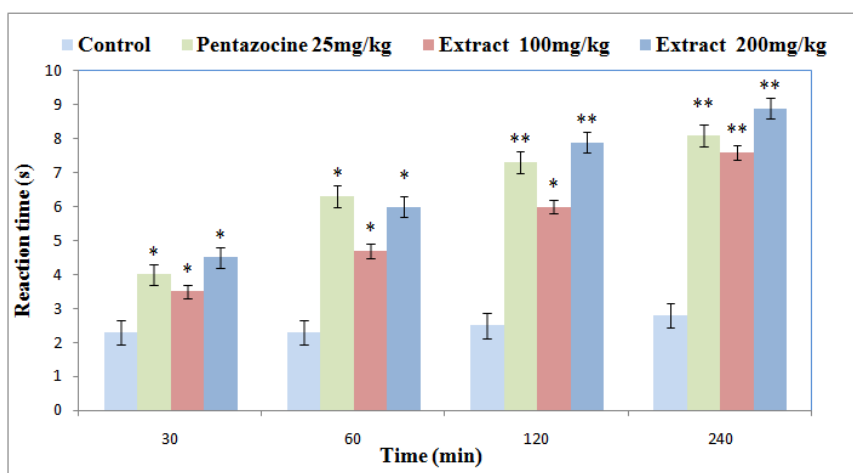


Fig. 1: Analgesic effect of methanol extract of *S. corymbosa* on heat stimulation response in the hot plate test.

Values are the mean ± SD (n = 6), *P < 0.05, **P < 0.01 compared to control group control (vehicle) – distilled water.

In this analgesic testing model, pentazocine significantly prolonged the reaction time of animals with relatively extended duration of stimulation, confirming centrally mediated activity. Methanol extract of *S. corymbosa* showed significant analgesic activity at 100 and 200 mg/kg. Analgesic activity of the latter dose (200 mg/kg) was often higher over the positive drug pentazocine. At the dose level 200 mg/kg and 240 min reaction time, the analgesic activity (8.90 ± 0.40 s) of the test extract was higher than that of the standard drug pentazocine (8.10 ± 0.90 s). Thermic painful stimuli are known to be selective to centrally active drugs [17]. In the present study, the extract showed significant ($P < 0.01$) analgesic activity but among the two doses, 200 mg/kg showed higher analgesic activity.

The hot plate test was selected to investigate central anti-nociceptive activity because it had several advantages, particularly the sensitivity to strong anti-nociceptives and limited tissue damage [18]. Prostaglandins and bradykinins were suggested to play an important role in analgesia [19]. In mice treated with the extract, there was a dose dependant increase in latency to thermal

stimulation compared with control mice. This effect started 60 min after the treatment and persisted throughout the 240 min duration of the experiment. For the extract and Pentazocine (25 mg/kg), the maximum antinociceptive effect was produced almost at about 240 min after treatment. The central analgesic action of the extract may be mediated through inhibition of central pain receptors.

The oral administration of methanol extract of *S. corymbosa* induced a dose dependent analgesic activity and the values \pm SD for the extract are shown in Table 1. Injection of acetic acid into the control mice resulted in 58.67 ± 1.69 writhes. Pretreatment with methanol extract of *S. corymbosa* at doses of 100 and 200 mg/kg reduced the number of writhes to 34.65 ± 3.19 (55.64% inhibition) and 19.25 ± 4.66 (75.01% inhibition) respectively. Interestingly, the extract dose 400 mg/kg registered higher levels of analgesic activity than the standard drug indomethacin (21.94 ± 5.76 writhes; 71.91% inhibition). It was also observed that the onset of writhing was delayed and duration of writhing was shortened with extract pretreatment.

Table 1: Analgesic effect of methanol extract of *S. corymbosa* on acetic acid-induced writhing test.

Group	Dose	Number of writhes (in 10 min)	Inhibition (%)
Control	10ml/kg	78.12 \pm 2.08	---
Indomethacin	25 mg/kg	21.94 \pm 5.76**	71.91
Extract	100 mg/kg	34.65 \pm 3.19*	55.64
	200 mg/kg	19.25 \pm 4.66**	75.01

The data represent the mean \pm standard deviation (n = 6).

* $P < 0.05$ ** $P < 0.01$ compared to corresponding control. Control (vehicle): distilled water.

Peripheral analgesic activity was assessed by acetic acid-induced writhing test, which showed significant ($P < 0.01$) suppression of writhes (Table 1). Acetic acid is known to trigger the production of noxious substances within the peritoneum, which induces the writhing response [20]. The effect of the extracts against the noxious stimulus may be an indication that it depressed the production of irritants and thereby bringing a reduction in the number of writhes in animals. The writhing induced by chemical substances is due to sensitization of nociceptors by prostaglandins [14]. The abdominal constriction response induced by acetic acid is a sensitive procedure to establish peripherally acting anti-nociceptives. The results indicate that the analgesic effect of methanol extract of *S. corymbosa* might be mediated by its peripheral effects by inhibiting the synthesis or action of prostaglandins.

The anti-inflammatory activity of *S. corymbosa* against acute pedal edema (induced by carrageenan) is shown in Table 2 and the results are comparable to that of the standard drug indomethacin, a potent inhibitor of the prostaglandins. Carrageenan induced paw edema remained even 6 h after its injection into the subplantar region of rat paw. Indomethacin as a reference standard drug inhibited the edema formation due to carrageenan to an extent of 64.44 % (at 6h) at the dose of 25 mg/kg. The methanol extract of *S. corymbosa*

significantly inhibited edema formation in rats ($P < 0.01$) in a dose dependant manner. The methanol extract at the dose of 200 mg/kg inhibited edema formation to the extent of 63.75% (at 6h) and the edema was found to be reduced to 3.15 ± 0.24 mm. The presence of edema is one of the prime signs of inflammation [21]. It has been documented that carrageenan induced rat paw edema is suitable *in vivo* model to study anti-inflammatory drugs both steroidal and non-steroidal since it involves several mediators [22]. This method was chosen for the present study since edema induced by carrageenan is the most prominent acute experimental model in search for new anti-inflammatory drugs [21].

Carrageenan-induced edema has been commonly used as an experimental animal model for acute inflammation and is believed to be biphasic [23]. The early phase (1-2) of the carrageenan model is mainly mediated by histamine, serotonin and increased synthesis of prostaglandins in the damaged tissue surroundings. The late phase (2.5-6) is due to the over production of prostaglandin and nitric oxide with peak at 3 h, produced by inducible isoforms of COX (COX-2) and nitric oxide synthase (iNOS) Panthong et al [24]. However, treatment with the methanol extract of *S. corymbosa* significantly reduced carrageenan induced inflammation in both the phases (1-6 h) of the experiment.

Table 2: Anti-inflammatory effect of *S. corymbosa* methanol extract on carrageenan induced paw edema in rats.

Treatment	Dose (mg/kg b. w.)	Swelling thickness (mm) \pm SD (inhibition %)				
		1h	2h	3h	4h	5h
Control	10 ml/ kg	4.23 \pm 0.65	5.78 \pm 0.43	6.28 \pm 0.45	7.53 \pm 0.43	8.69 \pm 0.10
Indomethacin	25	3.27 \pm 0.15 (22.69%)	3.48 \pm 0.76 (39.79%)	3.51 \pm 0.45 (44.10)**	3.42 \pm 0.43 (54.58%)***	3.09 \pm 0.71 (64.44%)***
Methanol extract	100	3.89 \pm 0.18 (8.03%)	5.10 \pm 0.67 (11.76%)	5.12 \pm 0.21 (18.47%)	5.58 \pm 0.41 (25.89%)	5.14 \pm 0.22 (40.85%)**
	200	3.12 \pm 0.41 (26.24%)	3.98 \pm 0.87 (31.14%)	3.78 \pm 0.43 (39.80)**	3.69 \pm 0.61*** (50.87%)	3.15 \pm 0.24 (63.75%)***

The data represent the mean \pm standard deviation (n = 6).

** $p < 0.05$ *** $p < 0.01$ compared to corresponding control. Control (vehicle): distilled water.

According to the result of our study, it may be concluded that the extract has a non-selective inhibiting effect on the release or actions of these mediators of inflammation. Based on this, it may

be concluded that the suppression of the 1st phase may be due to inhibition of the release of early mediators, such as histamine and serotonin and the action in the 2nd phase may be explained

by an inhibition of cyclooxygenase. The phytochemical investigation of genus *Swertia* revealed that xanthenes, flavonoids, terpenoids, iridoids, secoiridoid glycosides and saponin [25]. Of these, xanthenes are well known for their ability to inhibit pain perception. xanthenes also have anti-inflammatory properties due to their inhibitory effects on enzymes involved in the production of the chemical mediator of inflammation [26-30]. The synergetic effect of the anti-inflammatory in *S. corymbosa* may be responsible for the higher anti-inflammatory activity of this extract.

Subcutaneous injection of yeast suspension markedly elevated the rectal temperature after 24 h of administration. Treatment with the methanolic *S. corymbosa* aerial parts extract at the doses of 100 and 200 mg/kg significantly decreased the rectal temperature of the rats in a dose-dependent manner. The antipyretic effect stated as from the first and the effect was maintained for 4h, after administration of the extract. The result obtained from both the standard (paracetamol) and methanolic extract treated rats were compared with that of control and a significant reduction in the yeast elevated rectal temperature was observed (Table 3).

Table 3: Effect of *S. corymbosa* methanol extract on Brewer's yeast -induced pyrexia in rat model.

Treatment	Dose	Rectal temperature						
		Before treatment		After treatment				
		24h	0h	1h	2h	3h	4h	5h
Control	-	37.45±0.23	40.56±0.15	41.41±0.23	41.37±0.43	41.20±0.13	41.17±0.11	41.10±0.21
Paracetamol	150	37.61±0.32	41.20±0.32	39.95±0.11*	38.87±0.22*	38.52±0.19*	38.46±0.32*	37.85±0.31**
Methanol extract	100	37.11±0.12	40.97±0.31	39.91±0.19	38.72±0.32*	37.67±0.87**	37.45±0.43**	37.25±0.32**
	200	37.21±0.41	40.78±0.51	39.67±0.28*	38.86±0.50*	38.45±0.65*	38.21±0.33**	37.12±0.11***

The data represent the mean ± S.D. (n = 6).

*p < 0.05 **p < 0.01*** p < 0.001*** compared to corresponding control. Control (vehicle): distilled water.

The data revealed that the rectal temperature of 39.56±0.15°C at 0h was markedly elevated to temperature for control and 40.56±0.15°C for standard 24 h after the subcutaneous injection of yeast suspension. The animal treated with methanol extract at 100 and 200 doses showed a decrease in the rectal temperature by 1.06 and 1.11°C respectively within 1 h. On the other hand, after 5 hour the temperature was reduced by 3.66°C for 200 mg/k treated rats.

The results of the present study exhibited that methanol extract of *S. corymbosa* possesses analgesic, anti-inflammatory and antipyretic activities which may be mediated by the central and peripheral mechanisms. Further investigation on the isolation and characterization of individual compounds will be required to elucidate their different analgesic, anti-inflammatory and antipyretic mechanism and existence of possible synergism among the compounds.

REFERENCES

- Kalkan A, Bulut V, Erel O, Avci S, Bingol NK. Adenosine deaminase and guanosine deaminase activities in sera of patients with viral hepatitis. Mem Inst Oswaldo Cruz 1999; 94: 383-6.
- Hambleton P, Miller P. Studies on carragenin air pouch inflammation in the rat. Br J Exp Pathol 1989; 70:425-33.
- Cawthorn WP, Sethi JK. TNF-alpha and adipocyte biology. FEBS Lett 2008; 582:117-31.
- Bhatia M, Wong FL, Cao Y, Lau HY, Huang J, Puneet P. Pathophysiology of acute pancreatitis. Pancreatology 2005; 5:132-44.
- Costa EA, Lino RC, Gomes MN, Nascimento MVM, Florentino IF, Galdino PM, Andrade CH, Rezende KR, Magalhães LO, Menegatti R. Anti-inflammatory and antinociceptive activities of LQFM002-A4-nerolidylcatechol derivative. Life Sci 2013; DOI http://dx.doi.org/10.1016/j.lfs.2012.12.003.
- Hajhashemi V, Sajjadi SE, Heshmati M. Anti-inflammatory and analgesic properties of *Heracleum persicum* essential oil and hydro alcoholic extract in animal models. J Ethnopharmacol 2009; 124: 475-480.
- Dharmasiri M, Jayakody J, Galhena G, Liyanage S, Ratnasooriya W. Anti-inflammatory and analgesic activities of mature fresh leaves of *Vitex negundo*. J Ethnopharmacol 2003; 87: 199-206.
- Gupta M, Bisht D, Khatoon S, Srivastava S, Rawat AKS. Determination of Ursolic Acid a Biomarker in Different *Swertia* Species through High Performance Thin Layer Chromatography. Chinese Medicine 2011; 2: 121-124.
- Williamson EM .2002. Major herbs of ayurveda. Compiled by Dabur Res. Foundation & Dabur Ayurveda Ltd. India. Elsevier Sc. Ltd. London.
- Brahmachari G, Monda S, Gangopadhyay A, Gorai D, Mukhopadhyay B, Saha S, Brahmachari AK. *Swertia*

- (Gentianaceae): Chemical and Pharmacological Aspects. Chemistry & Biodiversity 2004; 1:1627-1651.
- Abraham Z. 1981. Ethnobotany of the Todas, the Kotas, the Irulas of the Nilgiris. In Jain SK. (Ed.), Glimpses of Indian Ethnobotany. Oxford & IBH, New Delhi. pp. 26-30.
- MacDonald AD, Woolfe G, Bergel F, Morrison AL, Rinderknecht H. Analgesic action of pethidine derivatives and related compounds. Br J Pharmacol 1946; 1:4-14.
- Siegmund E, Cadmus R, Lu G. A method for evaluating both non-narcotic and non narcotic analgesics. Proc Soc Exp Biol Med 1957; 95: 729-731.
- Vasudevan M, Gunnam KK, Parle M. Antinociceptive and anti-inflammatory properties of *Daucus carota* seeds extract. J Health Sci 2006; 52: 598-606.
- Nisar M, Khan I, Simjee SU, Gilani AH, Perveen OH. Anticonvulsant, analgesic and antipyretic activity of *Taxus wallichiana* Zucc. J Ethanopharmacol 2008; 116: 490-494.
- Aiyalu R, Ramasamy A, Shanmugasundram M. Evaluation of antipyretic activity of ethyl acetate extract of *Adenema hyssopifolium* G. Don in rat mode. Asian Paci J Trop Med 2010; 3: 523-526.
- Chau T. 1989. In: Alan R. (Ed.), Modern Methods in Pharmacology, vol. 5. Liss Inc. New York. pp. 195.
- Kou J, Ni Y, Li N, Wang J, Liu L, Jiang ZH. Analgesic and anti-inflammatory activities of total extract and individual fractions of Chinese medicinal plants *Polyrhachis lamellidens*. Biol Pharm Bull 2005; 28: 176-180.
- Vinegar R, Schreiber W, Hugo R. Biphasic development of carrageenan edema in rats. J Pharmacol Exp Ther 1969; 166: 96-103.
- Bartolini A, Galli A, Ghelardini C, Giotti A, Malcangio M, Malmberg-Aiello MP, Zucchi PL. Antinociception induced by systematic administration of local anaesthetics depends on a central cholinergic mechanism. Br J Pharmacol 1987; 92: 711-721.
- Yonathan M, Asres K, Assefa A, Bucar F. In vivo anti-inflammatory and anti-nociceptive activities of *Cheilanthes farinosa*. J Ethnopharmacol 2006; 108: 462-470.
- Mequanint W, Makonnen E, Urga K. In vivo anti-inflammatory activities of leaf extracts of *Ocimum lamiifolium* in mice model. J Ethnopharmacol 2011;134: 32-36
- Carey MW, Rao NV, Kumar BR, Mohan GK. Anti-inflammatory and analgesic activities of methanolic extract of *Kigelia pinnata* DC flower. J Ethnopharmacol 2010; 130:179-182.
- Panthong A, Supraditaporn W, Kanjanapothi D, Taesotikul T, Reutrakul V. Analgesic, anti-inflammatory and venotonic effects of *Cissus quadrangularis* Linn. J Ethnopharmacol 2007; 110:264-270.
- Tan P, Hou CY, Liu YL. Swertiapunicoside, the first bisxanthone C-glycoside. J Org Chem 1991; 56 (25): 7130-7133.

26. Banerjee S, Sur TK, Mandal S, Das PC, Sikdar S. Assessment of the anti-inflammatory effects of *Swertia chirata* in acute and chronic experimental models in male albino rats. Indian J Pharmacol 2000; 32: 21-24.
27. Chen LG, Yang LL, Wang CC. Anti-inflammatory activity of mangostins from *Garcinia mangostana*. Food Chem Toxicol 2007; 46: 688-693.
28. Nakatani K, Yamakuni T, Kondo N, Arakawa T, Oosawa K, Shimura S, Inoue H, Ohizumi Y. γ -Mangostin inhibits inhibitor-KB kinase activity and decreases lipopolysaccharide-induced cyclooxygenase-2 gene expression in C6 rat glioma cells. Mol Pharmacol 2004; 66: 667-674.
29. Firdous SM and Koneri R. *In vivo* and *in vitro* antiinflammatory activity of leaves of *Ipomoea staphylina*. Int J Pharm Pharm Sci 2012; 4:339-343.
30. Amutha D, Shanthi S and Mariappan. V. Antiflammatory effect of *Tarenna asiatica* (L) in carrageenan induced lung inflammation. Int J Pharm Pharm Sci 2012; 4:344-347.