



## Evaluation of pharmacological activities of traditional herbal drug *Ocimum sanctum* in rats

Mahima VEDI, Bharath Loganathan, Gayathri Chandrasekar, Saranya Vinayagam, Kanika Verma, Mahaboobkhan Rasool, Evan Prince Sabina\*

School of Biosciences and Technology, VIT University, Vellore-632014, Tamil Nadu, India

### ABSTRACT

*Ocimum sanctum* is a traditional herbal plant possessing numerous pharmacological and therapeutic properties. The present study was carried out to evaluate antipyretic, analgesic and anti-ulcerogenic properties of *Ocimum sanctum* (500 and 1000mg kg<sup>-1</sup>b.wt<sup>-1</sup>) in model system of rats. Drug Indomethacin (10mg kg<sup>-1</sup>b.wt<sup>-1</sup>) was used as a reference drug. Animals were randomly allocated in four groups consisting of six animals each. Group I was control group which received normal saline 0.09%; Group II received *O.sanctum* (500mg kg<sup>-1</sup>b.wt<sup>-1</sup>) and Group III was administered with 1000mg kg<sup>-1</sup>b.wt<sup>-1</sup> *O.sanctum*; Group IV received Indomethacin (10mg kg<sup>-1</sup>b.wt<sup>-1</sup>). Hot plate reaction test was used for determining the analgesic activity, acetic acid test was used to evaluate the writhing response, yeast induced pyrexia was used for determining the antipyretic activity test in control and experimental rats. Statistical analysis was performed using ANOVA to determine significant differences between groups followed by student's Newman-keul's test \*p<0.05 implied significance. Rats were fasted to induce ulcer and then effect of *O.sanctum* was evaluated. *Ocimum sanctum* at both the doses was found to possess analgesic, antipyretic, anti-ulcerogenic activities experimental rats and *O.sanctum* at a dose of 1000mg kg<sup>-1</sup>b.wt<sup>-1</sup> was found to be significantly effective than the other dose.

**Keywords:** *Ocimum sanctum*; antipyretic; analgesic; ulcer; indomethacin

### INTRODUCTION

*Ocimum sanctum* (Tulsi), which belongs to the family Lamiaceae, is a principal medicinal and most sacred herb of India (Samson et al, 2007). *Ocimum sanctum* is 30-100 cm tall and is found in ample in semitropical and tropical parts of India. It has been in use in India for over 5000 years for rejuvenating body and mind.

*Ocimum sanctum* has many reported pharmacological effects like ulcer ameliorative, antioxidant, anti-carcinogenic, antihelminthic, anti-septic, anti-rheumatic and anti-stress (Dharmani et al, 2004; Samson et al, 2007; Godhwani et al, 1987, 1988; Bhargava and Singh, 1981; Singh and Majumdar, 1999). However, the antipyretic and analgesic activities of this plant has not been reported up to best of our knowledge. The present study was carried out to evaluate analgesic, antipyretic, anti-ulcerogenic activity of the extract of *Ocimum sanctum* using the rats as model.

### MATERIALS AND METHODS

#### Test Drug

In this study, commercially available *Ocimum sanctum* and Indomethacin procured from Himalaya products Ltd., Bangalore, India was used. The other laboratory reagents were of analytical grade.

#### Animals

Rats of either sex, weighing 200-220gm, were used in this study. The animals were well treated and cared for in accordance of the guidelines recommended by the Committee for the Purpose of Control and Supervision of Experiments on Animals, Ministry of Culture, Govt. of India, Chennai, India. Animals were fed with commercially available pelleted feed and water.

#### Analgesic Activity

##### Hot plate reaction test

Hot plate test was done according to the method of Williamson et al (1996). Individually animals were kept in a beaker and placed on a temperature-controlled hot plate which was maintained at 50°C and it is speculated as pain threshold when animals lift and lick their paws or try to leap out of the beaker. The reaction of the rat response to the heat was noted using stopwatch.

\* Corresponding Author  
Email: eps674@gmail.com  
Contact: +91-9080494445  
Received on: 13-04-2013  
Revised on: 17-05-2013  
Accepted on: 20-05-2013

The animals used in the experiment were first tested for paw licks or jump response and those which reacted after 4 sec were used. Animals were tested after 30 minutes of administration of Indomethacin ( $10\text{mg kg}^{-1}\text{b.wt}^{-1}$ ) and *Ocimum sanctum* ( $500$  and  $1000\text{mg kg}^{-1}\text{b.wt}^{-1}$ ), which were suspended in 0.09% saline solution prior of use. Control animal were given equal volume of normal saline and the experiment was repeated. The difference between the control and drug treated animals among different groups were compared for statistical significance.

#### Acetic acid test

Acetic acid is used to instigate writhing response in rat. This test was done using the method by Witkin et al (1961) and intra peritoneal injection of 0.6% solution of acetic acid was administered in rats by muscular contraction. Animals were kept in glass cages and number of stretching per animals was noted for next 30 minutes. *Ocimum sanctum* ( $500$  / $1000\text{mg kg}^{-1}\text{b.wt}^{-1}$ ) and Indomethacin ( $10\text{mg kg}^{-1}\text{b.wt}^{-1}$ ) were suspended in 0.09% saline solution and were administrated 30 minutes before acetic acid injection.

#### Antipyretic Test

The rats were fasted overnight with water ad libitum before the experiments. This test was performed in rats by administering subcutaneously 20% aqueous suspension of baker's yeast to induce pyrexia and after 18 hours the rectal temperature of the animals were noted. Animals were given orally *Ocimum sanctum* ( $500$  and  $1000\text{mg kg}^{-1}\text{b.wt}^{-1}$ ) and Indomethacin ( $10\text{mg kg}^{-1}\text{b.wt}^{-1}$ ) and rectal temperature was noted at the interval of 1 hour (Mukerjee et al, 1996).

#### Ulcerogenic Test

The animals were kept fasting for 16 hours and then *Ocimum sanctum* ( $500$  and  $1000\text{mg kg}^{-1}\text{b.wt}^{-1}$ ) and Indomethacin ( $10\text{mg kg}^{-1}\text{b.wt}^{-1}$ ) orally. The animals were sacrificed after 6 hours of the last dose and the stomach was removed, opened along the great curvature and the severity of the ulcer index was measured using the arbitrary scale .0: no lesions, 0.5: hyperaemia, 1: one or two lesions, 2: severe lesions, 4: mucosa full of lesion (Cashin et al, 1997).

#### STATISTICAL ANALYSIS

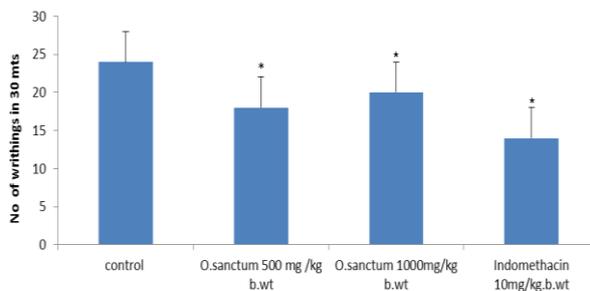
Results were expressed as mean  $\pm$ SD and statistical analysis was performed using ANOVA to determine significant differences between groups followed by student's Newman-keul's test \* $p < 0.05$  implied significance.

#### RESULTS

Pharmacological activities like analgesic, antipyretic, antiulcer activity of *Ocimum sanctum* were determined in rats.

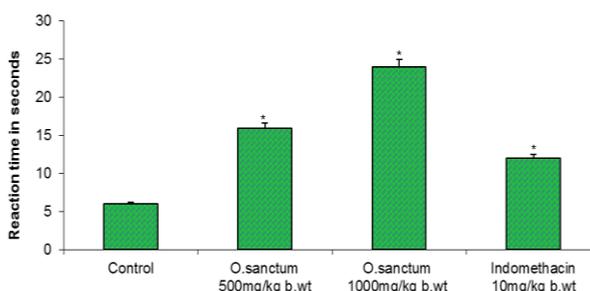
#### Analgesic activity

After the treatment of animals with *Ocimum sanctum* ( $500$  and  $1000\text{mg kg}^{-1}\text{b.wt}^{-1}$ ) there was a significant inhibition in the abdominal writhes. Similar results were noted in case of treatment with Indomethacin ( $10\text{mg kg}^{-1}\text{b.wt}^{-1}$ ) (Fig 1). In hot plate method, the rats treated with *Ocimum sanctum* ( $500$  and  $1000\text{mg kg}^{-1}\text{b.wt}^{-1}$ ) withstood on hot plate reactions for longer period compared to the reference drug Indomethacin ( $10\text{mg kg}^{-1}\text{b.wt}^{-1}$ ) (Fig. 2).



**Figure 1: Effect of *Ocimum sanctum* and Indomethacin on acetic acid induced writhing response in rat.**

Results are compared with control groups. Values are expressed as mean  $\pm$ S.D. (n=6). Symbols represent statistical significance at \*  $p < 0.05$ .

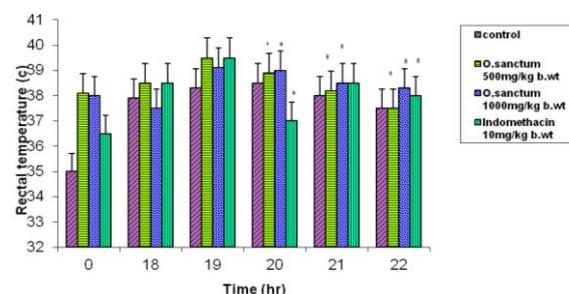


**Figure 2: Effect of *Ocimum sanctum* and Indomethacin on hot plate reaction time in rats.**

Results are compared with control groups. Values are expressed as mean  $\pm$ S.D. (n=6). Symbols represent statistical significance at \*  $p < 0.05$ .

#### Antipyretic activity

The aqueous suspension of *Ocimum sanctum* ( $500$  and  $1000\text{mg kg}^{-1}\text{b.wt}^{-1}$ ) was administered in rats and as compared to standard drug indomethacin ( $10\text{mg kg}^{-1}\text{b.wt}^{-1}$ ), *Ocimum sanctum* shows significant reduction in reaction in rectal temperature (Fig 3).

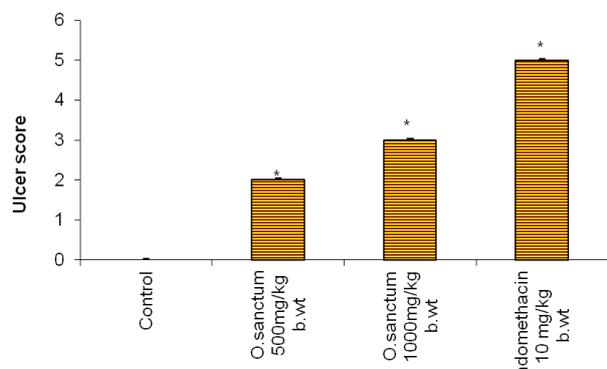


**Figure 3: Antipyretic effects of *Ocimum sanctum* and Indomethacin**

Results are compared with control groups. Values are expressed as mean  $\pm$  S.D. (n=6). Symbols represent statistical significance at \* p<0.05

#### Antiulcer activity

After administration of *Ocimum sanctum* (500/1000 mg kg<sup>-1</sup>b.wt<sup>-1</sup>) in rats, there was significant reduction in the ulcer index as seen in Fig.4.



**Figure 4: Ulcerogenic effects of *Ocimum sanctum* and Indomethacin in rats.**

Results are compared with control groups. Values are expressed as mean  $\pm$  S.D. (n=6). Symbols represent statistical significance at \* p<0.05

#### DISCUSSION

In the present scenario, some diseases like arthritis are treated with a number of anti-inflammatory drugs available in modern medicine. These drugs are always associated with various pyretic activities resulting in gastric damage. Effects of *Ocimum sanctum* in analgesic activity were evaluated by acetic acid test and hot-plate method. The acetic acid writhing test is known as a non-selective antinociceptive model. After intraperitoneal injection of acetic acid in rats, a painful response and acute inflammation develop in the peritoneal area due to which nerve endings are excited (Gyires and Torna, 1984) and an elevation of prostaglandin levels takes place in peritoneal fluid (Daud and Habib, 2006). Thus the analgesic effect of *Ocimum sanctum* may be due to prevention of the local level of prostaglandins. However, the interpretation of this writhing test alone does not confirm that this effect is related with central analgesic substances.

The hot plate test is extensively applied method in the analgesic investigations for several decades. This test along with the writhing test, usually differentiates between central and peripheral effects (Srinivasan et al, 2003). A significant analgesic action was shown by *Ocimum sanctum* (500 and 1000 mg kg<sup>-1</sup>b.wt<sup>-1</sup>) in hot plate method after 30 minutes administration. The results showed significant analgesic effect in acetic acid writhing response and hot plate reaction test by *Ocimum sanctum*. This confirms *Ocimum sanctum* has been acting through both peripheral and central mechanism in both models (Fig 2).

Antipyretic activity is usually implicated as an aspect of drugs or compounds that have a restraining effect on prostaglandin-formation (Panthong et al, 2007; Vane, 1987). The anti-pyretic effort is investigated by yeast inducing pyrexia test in rat. Subcutaneous injection of yeast induces pyrexia by causing an increment in production of prostaglandin that sequentially elevates the body temperature since the region in the hypothalamus controls body temperature is stimulated by pyretic activity (Ghamdi, 2001; Zacaria et al, 2008; Shukla et al, 2010). After 15-18 hours of yeast injection and the administration of antipyretic drugs is a method followed by many researchers. Results obtained using *Ocimum sanctum* showed a significant (p<0.05) reduction in rectal pyrexia, similar to standard drug Indomethacin (fig3).

Production of gastric lesions and thus ulcers is a common side effect associated with nonsteroidal anti-inflammatory compounds (Pegalla et al, 1983). For Indomethacin, already it has been seen to have ulcerogenic action in an empty stomach (Rasool et al, 2008). In this study it was found that *Ocimum sanctum* possesses significant (p<0.05) anti-ulcerogenic activity but gastric lesions were seen in Indomethacin treated rat.

#### CONCLUSION

Numerous analgesic compounds are available in market having several side effects so there is a need to evaluate the therapeutic potential of natural compounds. The result of the study shows *Ocimum sanctum* has antipyretic, analgesic and anti ulcerogenic properties, however further studies are required to study the mechanism of *Ocimum sanctum* to confirm these activities. Hence, our research contributes towards traditional use of *Ocimum sanctum* with scientific support.

#### CONFLICT OF INTEREST STATEMENT

There is no conflict of interest between the authors

#### REFERENCES

- Al-Ghamdi, M.S. 'The anti-inflammatory, analgesic and antipyretic activity of *Nigella sativa*'. J Ethnopharmacol, Vol.76, 2001 pp. 45–48.
- Bhargava, K.P. and Singh, N. 'Anti-stress activity of *Ocimum sanctum* Linn'. Indian Journal of Medical Research, Vol. 73, 1981 pp. 443–451.
- Cashin, C.H., Dawson, W. and Kitchen, E.A. 'The pharmacology of benoxaprofen (2-(4-chlorophenyl)- $\alpha$ -methyl-5-benzoxazole acetic acid), LRCL 3794, a new compound with anti-inflammatory activity apparently unrelated to inhibition of prostaglandin synthesis' J.Pharmacol.,Vol. 29,1977 pp. 330-336.
- Daud, A., Habib, N. and Sanchez, R.A. 'Anti-inflammatory, anti-nociceptive and antipyretic effects of extracts of *Phrygilanthus acutifolius* flowers'.

- Journal of Ethanopharmacology , Vol. 108,2006 pp. 198–203.
- Dharmani, P., Kuchibhotla, V.K., Mauryab, R., Srivastava, S., Sharma, S. and Palit, G. 'Evaluation of anti-ulcerogenic and ulcer-healing properties of *Ocimum sanctum* Linn.' Journal of Ethanopharmacology, Vol. 93, 2004 pp. 197–206.
- Godhwani, S., Godhwani, J.L. and Vyas, D.S. '*Ocimum sanctum*—a preliminary study evaluating its immunoregulatory profile in albino rats'. Journal of Ethanopharmacology, Vol.24, 1988 pp. 193–198.
- Godhwani, S., Godhwani, J.L. and Vyas, D.S. '*Ocimum sanctum*—an experimental study evaluating its anti-inflammatory, analgesic and antipyretic activity in animals'. Journal of Ethanopharmacology, Vol.21, 1987 pp. 153–163.
- Gyires, K. and Torna, Z. 'The use of the writhing test in mice for screening different types of analgesics'. Arch Int Pharmacodyn ,Vol. 267, 1984 pp. 131-40.
- Mukherjee, P.K., Das, J., Saha, K., Giri, S.N., Pal, M. and Saha BP. 'Antipyretic activity of *Nelumbo nucifera* rhizome extract'. Indian J Exp Biol, Vol. 34, No. 3, 1996 pp. 275-6.
- Panthong, A., Norkaew, P. and Reutrakul, V. Anti-inflammatory, analgesic and antipyretic activities of the extract of gamboges from *Garcinia hanburyi* Hook f. Journal of Ethanopharmacology, Vol. 111, 2007 pp. 335–340.
- Pegalla, P.G. and Bellavite, O. 'Pharmacological studies of imidazole z-hydroxybenzoate (ITE-182) an anti-inflammatory compound with an action on thromboxane A2 production'. Arzneimitte forschung, Vol.33, 1983 pp.716-726.
- Rasool, M., Sabina, E.P., Nithya, P. and Lavanya, K. 'Studies on analgesic, antipyretic and ulcerogenic properties of *Spirulina fusiformis* in mice'. Journal of pharmacology and toxicology, Vol.3, no.1, 2008 pp. 47-52.
- Samson, J., Sheeladevi, R. and Ravindran, R. 'Oxidative stress in brain and antioxidant activity of *Ocimum sanctum* in noise exposure'. NeuroToxicology, Vol. 28, 2007 pp. 679–685.
- Shukla, S., Mehta, A., Mehta, P., Vyas, S.P., Shukla, S. and Bajpai, V.K. Studies on anti-inflammatory, antipyretic and analgesic properties of *Caesalpinia bonducella* F. seed oil in experimental animal models. Food and Chemical Toxicology, Vol. 48, 2010 61–64.
- Singh, S. and Majumdar, D.K. 'Effect of *Ocimum sanctum* fixed oil on vascular permeability and leucocytes migration'. Indian Journal of Experimental Biology, Vol. 37, 1999 pp. 1136–1138.
- Srinivasan, K., Muruganandan, S., Lal, J., Chandra, S., Tandan, S.K., Raviprakash, V. and Kumar, D. Anti-nociceptive and antipyretic activities of *Pongamia pinnata* leaves. Phytotherapy Research, Vol.17, 2003 pp. 259–264.
- Vane, J.R. 'The evolution of non-steroidal anti-inflammatory drugs and their mechanisms of action'. Vol.33, 1987 pp. 18-27.
- Williamson, E.M., Okpako, D.T. and Evans, F.J. 'In pharmacological methods in phytotherapy research: selection, preparation and pharmacological evaluation of plant material'. Wiley Chichester, 1996 pp. 131-154.
- Witkin, L.B., Heibner, C.F., Gald, F., Kefee, E.O., Spilatetta, P. and Plummer, A.J. 'Pharmacology of 2-amino-indane hydrochloride (Su-8629). A potent non-narcotic analgesic'. J.pharmacol Exp Ther, Vol. 133, 1961 pp. 400-408.
- Zacaria, Z.A. and Abdul gani, Z.D.F. 'Antinociceptive, anti-inflammatory and antipyretic properties of an aqueous extract of *Dicranopteris lineareis* leaves in experimental animal model'. J Nat med, Vol.62, 2008 pp.179-187.