AMERICAN JOURNAL OF PHARMTECH RESEARCH

Journal home page: http://www.ajptr.com/

ANALGESIC AND ANTI-INFLAMMATORY ACTIVITY OF *DESMODIUM OOJEINENSE*(ROXB.)H.OHASHI

Jayadevaiah K.V[*1], Ishwar Bhat K[2], A.B.Joshi[3], Vijaykumar M.M.J[1], G.Swetha[1]

1. Department of Pharmaceutical Chemistry, SJM College of Pharmacy, Chitradurga-577502, Karnataka, India.
2.Department of Pharmaceutical Chemistry, NGSM Institute of Pharmaceutical Sciences, NITTE University, Mangalore - 575018, Karnataka, India.
3. Department of Phyto Chemistry, Goa College of Pharmacy, Goa.

ABSTRACT

The study was aimed to investigate the analgesic and anti-inflammatory activities of ethanolic extract of *Desmodium oojeinense*(Roxb.) in experimental rats. Analgesic activity was evaluated using acetic acid induced writhing method, while anti-inflammatory activity was evaluated using carrageenan induced rat paw oedema model. Various doses of ethanolic extract of the plant (100, 200, 400 mg/kg body weight) were tested for its analgesic activity and anti-inflammatory activity and the results were compared with the standard drug aspirin and diclofenac sodium respectively. Results indicate that the ethanolic extract of the plant significantly inhibited writhing movements in analgesic activity and carrageenan induced hind paw oedema in anti-inflammatory activity in a dose dependent manner. The results suggest that there exists a potential benefit in utilizing *Desmodium oojeinense*(Roxb.) in treating conditions associated with pain and inflammation.

Key Words: *Desmodium oojeinense* (Roxb.), analgesic, anti-inflammatory, carrageenan.

*Corresponding Author Email: jayadevkv@rediffmail.com

Received 23 January 2012, Accepted 28 January 2012

Please cite this article in press as: Jayadevaiah KV *et al.*, Analgesic and Anti-Inflammatory Activity of *Desmodium oojeinense*(Roxb.)H.Ohashi. American Journal of PharmTech Research 2012.

INTRODUCTION:

Pain is the unpleasant and aversive feeling common to such experiences as a stubbed toe, a headache, a burnt finger, and salt in a wound. Typically, pain is characterized by its intensity, location and duration[1]. It is initiated by stimulation of nociceptors in the peripheral nervous system, or by damage to or malfunction of the peripheral or central nervous systems. Pain is broadly classified as acute or chronic[2]. The drugs that relieve pain due to multiple causes at multiple sites are called Analgesics. Analgesics are classified as Narcotics which act on CNS and cause drowsiness and sleep e,g. opioids and Non-narcotics which basically act on peripheral parts of body and do not produce sleeping effect. The non steroidal anti-inflammatory drugs fall in this group[3].

The discovery of medicinal drugs from natural products is not new. Some of the best drugs we have today are from natural products. Herbal or natural products are becoming popular which can be attributed to the belief that they are safe.

Desmodium Oojeinense (Roxb.), belonging to the family Fabaceae. It is found distributed in the sub and outer Himalayan valleys and slopes up to altitude of 5000 ft, from Punjab to Bhutan, Chota Nagpur, Central India, Orissa, Bombay, Marvar of Rajputan, forest of Ganjam, and Vizag. The bark is acrid and hot, anthelmintic, astringent to the bowels, cures “kapha” and “vata”, dysentery, leucoderma, urinary discharges, ulcers, blood diseases, skin diseases, burning sensations and anemia (Ayurveda). In the central prominence the bark is used as a febrifuge. The bark when incised furnishes a Kino-like exudation, which is used in cases of dysentery and diarrhoea[4].

Based on ancient practices and traditional uses of this plant, an effort has been made to establish the analgesic and anti-inflammatory activity of ethanolic extract of *D.oojeinense*.

MATERIALS AND METHODS

Collection of plant material: Bark was collected from the medicinal garden of Sri.Ragavendra Ayurvedic Medical College Malladihalli, Karnataka, India .The plant was authenticated by Prof. Gopal Krishna Bhatt, Department of Botany, Poornaprajna College, Udipi, Karnataka, India. A voucher specimen (No.106a) was deposited in NGSM Institute of Pharmaceutical Sciences, Paneer, Mangalore-5, Karnataka.

Preparation of the ethanolic extract: The dried powder material of stem bark of *D.oojeinense* was extracted with ethanol (95%) in a soxhlet extractor. The process was repeated for six times.

The solvent from the total extract was distilled off and the concentrate was evaporated on a water bath to a syrupy consistency and then evaporated to dryness.

Preliminary phytochemical screening:

The ethanolic extract obtained by *D.Oojeinense* was subjected to systematic qualitative analysis for the identification of various plant phytoconstituents.

Animals: Albino wistar rats of either sex (180-260g) were obtained time to time from the laboratory of K.S Hegde Medical Academy (KSHEMA), Deralakatte, Mangalore. Animals were housed in a cage in a climate-controlled room under standard conditions with 12:12h light/dark cycles and free access to water and food. All the experiments were performed within the guidelines of the institutional ethical committee of KSHEMA, Deralakatte, Mangalore (KSHEMA/AEC/NO.051/2007).

Acute Toxicity Studies: The acute toxicity study was carried out in adult female albino rats weighing about 150-200g by up and down method as per OECD 425 guidelines[5]. Overnight fasted animals received drug at a dose of 2000mg/kg body weight orally. Then the animals were observed continuously once in half an hour for next 4h and then after 24h for general behavioral, neurological, autonomic profiles and to find out mortality.

The effective dose of 50% ethanolic extract of *D.oojeinense* was decided as one-tenth of the maximum dose (2000mg/kg). So we used the 50% ethanolic extract D.Oojeinense ***as*** such 100, 200 and 400mg/kg body weight p.o. for analgesic and anti-inflammatory activity.

Statistical analysis: The data were expressed as mean ± SEM of six animals in each group. Data was analyzed by one way analysis of variance (ANOVA) followed by post hoc schiffe's test. $p<0.05$ was considered as statistically significant.

Evaluation of analgesic activity:

Acetic acid induced writhing method: In this method the pain was induced by injecting the acetic acid (0.6% v/v) in to peritoneal cavity of a mouse at a dose of 10ml/kg body weight, i.p. The animals react with characteristic stretching behavior, which is called writhing[6].

In this method healthy albino mice of either sex weighing 25-30g were selected for the study. The animals were divided in to 5 groups of 6 animals each.

Group I: Animals served as control and received 0.5ml of 1 % solution of tween 80.

Group II: Animals served as positive control and received acetyl salicylic acid (aspirin) (50 mg/kg body weight, p.o.) as a suspension in tween 80 (0.5ml of 1 % solution).

Group III: Animals were treated with ethanolic extract of the plant ***D.oojeinense*** at a dose of 100 mg/kg body weight, p.o. as a suspension in tween 80 (0.5ml of 1 % solution).

Group IV: Animals were treated with ethanolic extract of the plant at a dose of 200 mg/kg of body weight p.o. as a suspension in tween 80 (0.5ml of 1 % solution).

Group V: Animals were treated with ethanolic extract of the plant at a dose of 400mg/kg body weight, p.o. as a suspension in tween 80 (0.5ml of 1 % solution).

Further the animals in all the groups were administered with 1ml/100g body weight of 0.6% v/v acetic acid, i.p. 30 min after the drug administration. The writhing movements such as extension of hind limb, abdominal constriction and trunk twisting were observed and counted for 30 min. The percentage inhibition was calculated using the formula.

$$\% \text{ Inhibition} = (1-Rt/Rc) \times 100$$

Where Rt= mean reaction time in treated group

Rc= mean reaction time in control group

Evaluation of anti-inflammatory activity:

Carrageenan induced rat paw oedema method: The method of winter *et. al.*[7] was used to study the anti-inflammatory activity using plethysmograph apparatus to measure the paw volume.

A mark was made on both the hind paws just below the tibio-tarsal junction, so that every time the paw could be dipped in the mercury column of plethysmograph up to the mark to ensure constant paw volume. After 30 min of above treatment, the inflammatory oedema was induced in the left hind paw by injecting 0.1ml 1 % carrageenan in the plantar tissue of the paw of all the animals. The right paw served as a reference to non-inflamed paw for comparison. The initial paw volume was measured plethysmographically within 30 sec of the injection. The relative increase in the paw volume was measured in control, standard and treated groups 4 hrs after carrageenan injection. The % increase in paw volume over the initial reading was also calculated. This increase in the paw volume in animals treated with standard drug and the different doses of ethanolic extracts of plant *D. oojeinense* were compared with the paw volume of untreated control animals after 4 hrs.

In this method healthy albino rats of either sex weighing between 150-200g were selected for the study. The animals were divided in to 5 groups of 6 animals each.

Group I: Animals served as control and received vehicle tween 80, 3ml of 1 % solution orally.

Group II: Animals were administered with standard drug diclofenac sodium at a dose of 20 mg/kg body weight, p.o. as a suspension in tween 80.

Group III: Animals were treated with ethanolic extract of the plant *D.oojeinense* at a dose of 100 mg/kg body weight, p.o. as a suspension in tween 80.

Group IV: Animals were treated with ethanolic extract of the plant at a dose of 200 mg/kg of body weight p.o. as a suspension in tween 80.

Group V: Animals were treated with ethanolic extract of the plant at a dose of 400 mg/kg body weight, p.o. as a suspension in tween 80.

The % inhibition of oedema volume was calculated using the formula,

$$\% \text{ Inhibition} = [1 - Vt/Vc] \times 100$$

Where,

Vt = mean increase in the paw volume in test animals group

Vc = mean increase in the paw volume in control group

RESULTS AND DISCUSSION

Preliminary phytochemical investigation

Qualitative test for phytoconstituents revealed the presence of alkaloids, carbohydrates, steroids, triterpenoids, and flavonoids. Results are shown in Table 1.

Table 1: Preliminary phytochemical investigation

Sl.no	Phytoconstituents	Inference
1	Alkaloids	+ve
2	Carbohydrates	+ve
3	Steroids	+ ve
4	Triterpenoids	+ ve
5	Flavonoids	+ve

The presence of these phytoconstituents may be responsible for the analgesic and anti-inflammatory activities.

Table 2: Analgesic activity of ethanolic extract of the plant *D.oojeinense*

Group	Treatment	Dose (mg/kg, p.o)	Mean writhing per 30 min	Inhibition %
I	Tween 80(1%)	-	80.66±2.12	-
II	Aspirin	50	20.18±0.88*	74.98
III	ethanolic extract	100	45.29±0.42*	43.86
IV	ethanolic extract	200	39.43±0.47*	51.12
V	ethanolic extract	400	33.71±0.88*	58.21

Values are expressed in mean ± SEM (n=6) $p < 0.05$ considered as significant when compared with control group.

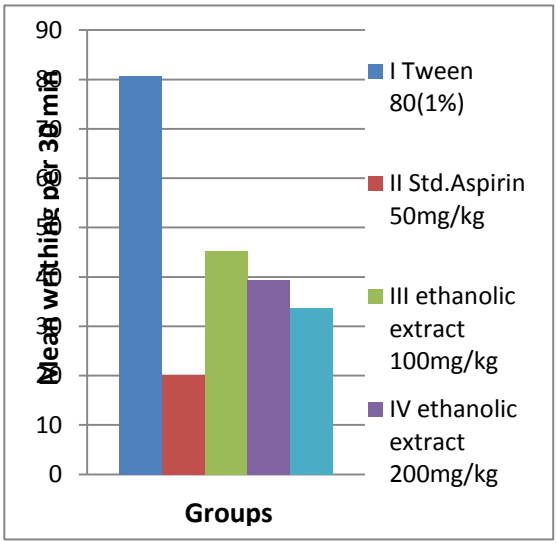

Figure 1

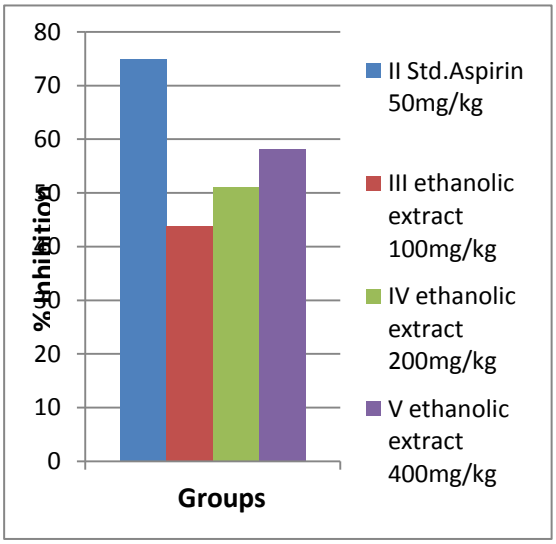

Figure 2

Analgesic activity of ethanolic extract of the plant *D.oojeinense*

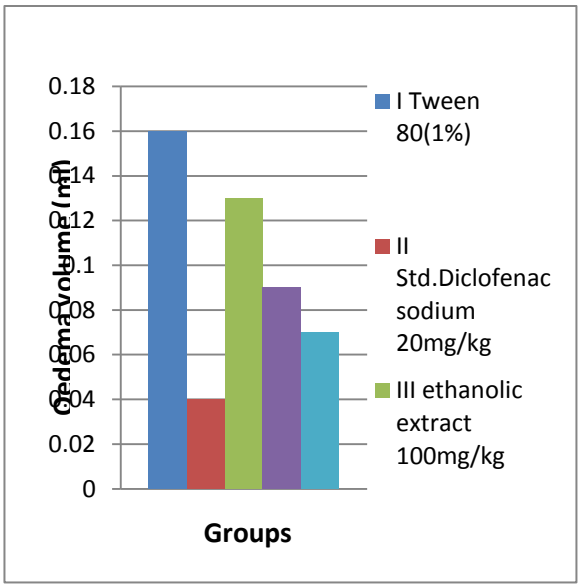

Figure 3

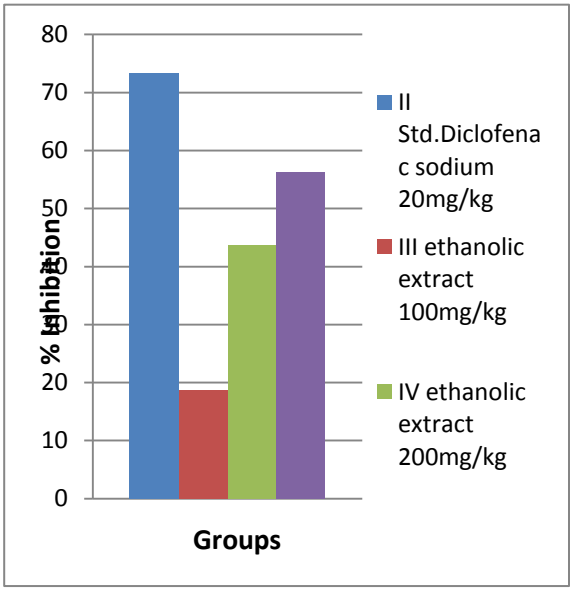

Figure 4

Anti-inflammatory activity of ethanolic extract of the plant *D.oojeinense*

The results of analgesic studies of the ethanolic extract of *D.oojeinense* showed a significant dose dependent reduction in number writhes. The ethanolic extract at a dose of 100, 200, 400 mg/kg body weight showed 43%, 51 %, 58 % inhibition respectively when compared to the standard drug aspirin that was 74.98 %. The results are summarized in the Table 2 and graphical representation of the results is indicated in Figure 1 and Figure 2.

The results of anti-inflammatory studies showed significant decreased paw volume in comparison with the standard group. The ethanolic extract at a dose of 100, 200, 400 mg/kg body weight showed 18 %, 43% and 56 % inhibition respectively when compared to standard drug Diclofenac sodium that was at 73%. It exhibited dose dependent anti-inflammatory activity. The

results are presented in Table 3 and graphical representation of the results is indicated in Figure 3&4.

Table 3: Anti-inflammatory activity of ethanolic extract of the plant *D.oojeinense*

Group	Treatment	Dose (mg/kg, p.o)	Oedema volume (ml)	Inhibition %
I	Tween 80(1%)	-	0.16±0.004	-
II	Diclofenac sodium	20	0.04±0.004**	73.3
III	ethanolic extract	100	0.13±0.003*	18.75
IV	ethanolic extract	200	0.09±0.003**	43.75
V	ethanolic extract	400	0.07±0.007**	56.25

Values are expressed in mean ± SEM (n=6) p< 0.05 considered as significant when compared with control group.

CONCLUSION

The ethanolic extract of the plant *D.oojeinense* was subjected to analgesic and anti-inflammatory activity studies. The results indicated that the ethanolic extract of the plant was found to have significant analgesic and anti-inflammatory activity.

ACKNOWLEDGEMENT

The authors are thankful to Dr.Shivamurthy Murugha Sharanaru, President, SJM college of Pharmacy, Chitradurga for providing all necessary facilities through the Principal. The authors are also thankful to the Principal, NGSMIPS, Mangalore and NITTE educational trust.

REFERENCES

1. Finkel, Richard et al. Drugs Affecting the Central Nervous System, Lippincott's Illustrated Reviews, Pharmacology 2009; 4:159.
2. Brunner L et al. Textbook of Medical Surgical Nursing, JB Lippincott Company, 1988; 6: 242 - 258.
3. Rang HP. *et al,* Analgesics, Rang and Dale's Pharmacology, 2007; 6:596-607.
4. Kirthikar KR, Basu BD. Indian Medicinal Plants.Vol-1.Allahabad: 1935, 755-756.
5. OECD 425 guidelines. OECD guidelines for testing of chemicals. 2001 Dec; 1/26:1-26.
6. Koster R, Anderson M, Debeer EI. Acetic acid for analgesic screening. Fed. Proc. 1959; 18: 412.
7. Risely CA, Nuss EA.Carrageenan induced oedema in hind paw of the rat as an assay for anti-inflammatory drugs. Pro Soc Exp Bio Med 1962; 111: 544-547.
8. Jena PK, Chakraborty AK. Evaluation of analgesic activity studies of various extracts of leaves of *Eupatorium odoratum* Linn. Ind J Pharm Tech 2010; 2(3): 612-616.
9. Panda BK, Patra VJ, Mishra US, Kar S, Panda BR, Hati MR. Analgesic activities of the stem bark extract of *Spondias piñata* (Linn.f)Kurz. J PharmRes 2009; 2(5): 825-827.

10. Hajare SW, Chandra S, Tandan SK, Sarma J, Lal J, Telang AG. Analgesic and anti-pyretic activities of *Dalbergia sissoo* leaves. Ind J Pharmacology 2000; 32: 357-360.
11. Alam K, Pathak D, Ansari S. H. Evaluation of Anti-inflammatory activity of *Ammomum subulatum* fruit extract. Int J Pharm Sci Drug Res 2011; 3(1): 35-37.
12. Behera GM, Baidya M, Satish BN, Sayed Bilal, Panda S. Analgesic and anti-inflammatory effect of different extracts of *Ocimum canum*. Res J Pharm Bio Chem Sci 2011; 2(1) : 283-296
13. Adeolu A. Adedapo, Margaret O. Sofidiya, Viola Maphosa, BusaniMoyo, Patrick J. Masika and Anthony J. Afolayan. Anti-inflammatory and analgesic activities of the aqueous extract of *Cussonia paniculata* stem Bark. Rec Nat Prod 2008; 2(2): 46-53.
14. Nain Parminder, Saini Mamta, Malik Manisha. Evaluation of anti-inflammatory and analgesic activity of *Punica granatum* linn leaves. Int J Res Ayur Pharm.2011; 2(3) : 987-990
15. Saraswathi R, Lokesh Upadhya, Venkatakrishnan R, Meera R, Devi P. Phyto chemical investigation, analgesic and anti inflammatory activity of *Abutilon indicum* linn. Int J Pharm Pharm Sci, 3(2), 2011, 154-156