Plant Archives Volume 20(2), 2020 pp. 7976-7978 **e-ISSN:2581-6063 (online), ISSN:0972-5210**

ANTINOCICEPTIVE ACTIVITY OF ETHANOLIC EXTRACT OF *WEDELIA CHINENSIS*

Nitin Verma[1*] and R.L. Khosa[2]

[1*]Chitkara University School of Pharmacy, Chitkara University, District-Solan-173205 (H.P.), India.
[2]Department of Pharmacy, Bharat Institute of Technology, Bye-Pass Road, Partapur, Meerut-50 103 (U.P.), India.

Abstract

The objective of the present study is to evaluate the antinociceptive activity of ethanolic extract of *Wedelia chinensis* whole plant in Swiss albino mice. The ethanolic extract of *Wedelia chinensis* (WCEE) at a dose of 200, 300 and 500 mg/kg was used for the antinociceptive studies using writhing response induced by acetic acid and hot plate method in Swiss albino mice. Ethanolic extract of *Wedelia chinensis* at a dose of 200,300 and 500 mg/kg inhibited the writhing response induced by acetic acid in a significant and dose-dependent manner, by 7.5%, 57.7% and 78.2% respectively. Using the same doses its ethanolic extract elicited a reduction of licking time during the first phase (neurogenic) at the highest dose of its ethanolic extract. In conclusion, its ethanolic extract possesses antinociceptive properties.

Key words : *Wedelia chinensis*; antinociceptive; acetic -acid writhing; hot plate.

Introduction

The genus *Wedelia* comprises over 60 species distributed in tropical and warm temperate regions, including India, Burma, Ceylon, China and Japan of which nearly two dozen species are reported to be medicinally active (Verma and Khosa, 2015). Among these *Wedelia chinensis* Merrill (Syn. *Wedelia calendulaceae*) (Asteraceae), is a small much branched annual herb commonly known as "Pilabhamgara" or "Bhringraj' is a reputed herbal medicine in both Ayurvedic and Unani system of medicine. The herb contains wedelolactone and demethylwedelolactone (Coumestans derivatives) possessing potent anti-hepatotoxic effect and is incorporated as a major ingredient in a number of developed potent anti-hepatotoxic phytopharmaceuticals formulations. *Wedelia chinensis* is used as traditional herbal medicines throughout the world and they have been reported to possess hepatoprotective, bactericidal, molluscicidal, hypoglycemic and antitumor activities (Jiangsu, 1977) whereas antioxidant (Verma and Khosa, 2008) wound healing (Verma *et al.,* 2008) antistress activity (Verma and Khosa, 2009) and hepatoprotective activity on *W. chinensis* have been reported by our research group in recent years (Verma *et al*., 2009) It is useful in the treatment of osteoporosis of knee and also possesses anti-inflammatory activity (Anonymous, 2005; Yuan *et al*., 2013) As it contains large amount of phenolic constituents and it is also effective in the treatment of inflammatory conditions, so its antinociceptive activity was studied in detail.

Materials and Methods

Plant material

The whole plant of *Wedelia chinensis* was procured from the Plant Physiology Division, Jawaharlal Nehru Krishi Vishwa Vidyalaya, Krishi Nagar, Jabalpur, M.P. and authenticated by the taxonomic division, National Herbarium of Cultivated Plants, National Bureau of Plant Genetic and Resources, New Delhi. A voucher specimen (NHCP/NBPGR/2007/99/2225 dated 22/08/2007) was retained in our laboratory for further reference.

Plant extract

The plant material was dried under shade, reduced to moderately coarse powder and was extracted successively with petroleum ether (60-80^{o}C) and ethanol using soxhlet apparatus. The ethanolic extract was dried under vacuum (yield 6.78%) and its qualitative analysis showed the presence of phenolic compounds, saponins,

reducing sugars and flavonoids. The ethanolic extract of *Wedelia chinensis* (WCEE) was used for the antinociceptive studies.

Animals

Swiss albino mice of both sexes (20±2g) were used for the present studies. They were housed in clean polypropylene cages (38X23X10 cm) with not more than six animals per cage and maintained standard laboratory condition (temperature25±2°C) with dark and light cycle (12/12 h). They were allowed free access to standard pellet diet (Hindustan Lever, Kolkata, India) and water *ad libitum*. The Institutional Animal Ethics Committee, (IAEC) approved the use of animals for the present study**.**

Writhing test

The mice were pretreated with WCEE (200,300and 500 mg/kg i.p) and Aspirin (100mg/kg i.p). After 30 min, 0.6% (v/v) solution of acetic acid was injected i.p. (10ml/kg). The numbers of writhes were noted for 15 minutes beginning 5 minutes after acetic acid injection (Collier *et al.,* 1968; Omonkhelin and Eric 2007).

$$\%\,\text{Inhibition} = \frac{W_C - W_T}{W_C} \times 100$$

Where,

W_C = Mean number of writhes in control group

W_T = Number of writhes in test group

Hot plate test

Each mice was dropped on the heated plate (55±0.5°C), separated by 30 min interval from each other. The first trial familiarized the animals with the test procedure and second served as control reaction time (licking the paw or jumping). Animals showing a reaction time greater than 10 sec. were discarded. Immediately after the second trial (control reaction time), groups of six mice each received i.p. saline, WCEE (200,300 and 500 mg/kg) and Pentazocine (10mg/kg) (Goverdhan Puchchakayala et al 2008). Reaction time were measured at time zero (0 time), 30, 60,120 and 180 min after compounds administration with a cut-off time of 40 sec (Turner, 1965; Goverdhan *et al.,* 2008).

Statistical analysis

All the data obtained were expressed as mean ± standard error. The results were analyzed using student t-test.

Results

Antinociceptive activity was investigated by the acetic acid induced writhing test and Hot plate method in mice. Results of writhing studies in mice are presented in (Table 1). The maximum writhes were produced by saline treated mice. The ethanolic extract of *W. chinensis* (200, 300, 500 mg/kg i.p.) showed a significant dose dependent reduction in the number of writhing with 7.5%, 57.7% and 78.2% of inhibition respectively. The maximum inhibition was observed at a dose of 500mg/kg, which was statistically similar to the standard drug, diclofenac sodium (10 mg/kg).

The ethanolic extract of *W. chinensis* (200, 300, 500 mg/kg i.p.) elicited a significant analgesic activity in the hot plate as evidenced by increase in latency time in seconds table 2 as compared with vehicle control. The increase in latency time was dose dependent. The latency time was noted at 0, 30, 60, 120 and 180 minutes after the administration of vehicle, standard and plant extract. WCEE at doses of 300 and 500 mg/kg showed significant increase in latency time in mice which is comparable with standard drug.

Discussion

Inhibition of acetic acid-induced writhing in mice suggests that the analgesic effect of the extract may be peripherally mediated via the inhibition of the synthesis

Table 1: Effect of Ethanolic extract of *Wedelia chinensis* on the acetic acid induced writhing in mice.

S.No.	Groups	Dose (mg/kg)	No. of Writhings (Mean ± SEM)	% Inhibition
1	Control		39.83 ± 1.5584	-
2	Aspirin	100	6.83 ± 0.4773**	82.85
3	WCEE	200	36.83 ± 0.6010*	7.53
4	WCEE	300	16.83 ± 0.7033**	57.74
5	WCEE	500	8.66 ± 0.6147**	78.25

Values are expressed as Mean ± S.E.M. n=06

* $p<0.02$ vs control

** $p<0.001$ vs control

and release of prostaglandins (Koster *et al.,* 1959) Writhes can be described as a wave of constriction and elongation passing caudally along the abdominal wall with twisting of the trunk and extension of the hind limbs in mice. This is due to the nociceptive property of acetic acid (Surender and Mafumdar 1995). The percentage of inhibition, clearly shown in Table 1, also indicates that the extract at 300 and 500 mg/kg produced a significant inhibition when compared to aspirin (100 mg/kg) a known standard analgesic drug.

Hot plate test has been found to be suitable for evaluation of centrally but not of peripherally acting analgesics. The results shown in table 2 clearly indicates that the extract at 300 and 500 mg/kg showed significant

Table 2: Effect of Ethanolic extract of *Wedelia chinensis* on the hot plate test in mice.

S.No.	Groups	Dose (mg/kg)	Time of Response (Seconds)				
			0	30	60	120	180
1	Control		6.33±0.1666	6.76±0.2092	7.28±0.1301	7.33±0.0918	7.48±0.0792
2	Pentazocine	10	6.26±0.0792	10.76±0.1763***	14.71±0.3736***	19.41±0.5364***	21.36±0.6587***
3	WCEE	200	6.41±0.2007	7.16±0.1308	7.88±0.1905*	8.06±0.2275**	7.73±0.1706
4	WCEE	300	6.63±0.2044	8.60±0.2221***	9.08±0.1833***	9.38±0.1851***	9.48±0.3081***
5	WCEE	500	6.75±0.1544	9.5±0.1844***	10.98±0.2040***	14.78±0.3944***	19.06±0.6946***

Values are expressed as Mean ± S.E.M. n=06

* $p<0.05$ vs control

** $p<0.02$ vs control

*** $p<0.001$ vs control

analgesic activity which is comparable with the standard drug, Pentazocine (10 mg/kg).

The fact that WCEE showed analgesic activity in both the models studied, indicated that the analgesic effect of WCEE could possess two components viz. central and peripheral (Panthong *et al.,* 1998).

Conflict of interest statement

We declare that we have no conflict of interest.

Acknowledgement

The authors are thankful to Dr. Anjula Pandey, Scientist, Taxonomic Division, National Herbarium of Cultivated Plants, National Bureau of Plant Genetic and Resources, New Delhi for plants identification and its authentication.

References

Verma, N. and R.L. Khosa (2015). Chemistry & biology of genus *Wedelia*: A review. *Indian J. Nat. Prod. Resour.,* **62(2):** 71-90.

Jiangsu, New Medical College, NA *Comprehensive Dictionary of Traditional Chinese Materia Medica*, Shanghai Peoples Press, Shanghai, 1977.

Verma, N. and R.L. Khosa (2008). Antioxidant potential of some Indian medicinal plants. *Nar. Phay. J.*, **1:** 99-101.

Verma, N., R.L. Khosa and V.K. Garg (2008). Wound healing activity of *Wedelia chinensis* leaves. *Pharmacologyonline*, **2:** 139-145.

Verma, N. and R.L. Khosa (2009). Effect of alcoholic extract of *Wedelia chinensis* on cold immobilization stress induced tissue lipid peroxidation. *Pharmacologyonline,* **1:** 493-497.

Verma, N. and R.L. Khosa (2009). Effect of *Costus speciosus and Wedelia chinensis* on brain neurotransmitters and enzyme monoamine oxidase following cold immobilization stress. *J. Pharm. Sci. Res.,* **1:** 22-27.

Verma, N., G. Mishra, R. Sinha, R.L. Khosa, V.K. Garg and P. Singh (2009). Hepatoprotective activity of alcoholic and aqueous extracts of *Wedelia chinensis*. *Pharmacologyonline,* **1:** 345-356.

Anonymous, *The Wealth of India-A Dictionary of Indian raw materials & industrial products*, Vol. X (Sp-W), Publication and information directorate, CSIR, New Delhi, 2005.

Yuan, F., J. Chen, P.P. Sun, S. Guan and J. Xu (2013). Wedelolactone inhibits LPS-induced pro-inflammation via NF-kappaB pathway in RAW 264.7 cells. *J. Biomed. Sci.*, **31:** 20-28.

Collier, H.O.J., L.C. Dinneen, C.A. Johnson and C. Scheider (1968). The abdominal contraction response and its suppression by antinociceptive drugs in the mouse. *Br. J. Pharmacol. Chemother.,* **32:** 295-310.

Omonkhelin, J.O. and K.I.O. Eric (2007). Analgesic and anti-inflammatory activities of the ethanolic stem bark extract of *Kigelia africana* (Bignoniaceae). *Afr. J. Biotechnol.,* **6:** 582-585.

Turner, R.A. (1965). *Screening method in Pharmacology*. Academic Press New York and London 1965, 26-34.

Goverdhan, P., P. Laxmi, B. Diwakar, K. Thirupathi, M.B. Krishna, R.Y. Narasimha, K.B. Ravi, M.G. Krishna and R.P. Rajeshwara (2008). Antinociceptive and anti-inflammatory effects of *Cleome chelidonni* Linn. roots in experimental animals. *Pharmacogn. Mag*., **4:** 32-36.

Koster, R.M., M. Anderson and E.J. De Beer (1959). Acetic acid for analgesic screening. *Federation Proceedings*, **18:** 412.

Surender, S. and D.K. Mafumdar (1995). Analgesic activity of *Ocimum sanctum* and its possible mechanism of action. *Int. J. Pharmacog.,* **33:** 188-192.

Panthong, A., D. Kanjanapothi, Y. Thitiponpunt, T. Taesotikul and D. Arbain (1998). Anti-inflammatory activity of the alkaloid bukittinggine from *Sapium baccatum*. *Planta Med*., **64:** 530-535.