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Original Article

Estımatıon Of Aqueous Morınga Oleıfera Extract For Antınoceceptıve Effect In Rat Model By Usıng Hot Plate Test

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Abstract

*Moringaoleifera*is a folk plant of Moringaceae occurring in and around Udupi district in India. It is effectively used in the folklore medicine in the management of low backache and arthritis. The traditional medicines have been used as sources of bioactive compounds, including potential antitumor, antioxidant, antiobesity, and antimicrobial molecules. The present study aimed to evaluate analgesic activityof aqueous extract of leaves of *M. oleifera*in albino rats. Eddy's hot plate test was used for antinoceceptive assay. The animals were divided into four groups control, standard and test. The control group administered with distilled water, standard group with Panadol 100 mg/kg and the test group*received* aqueous leaves extract of *M. oleiferaat* different doses (50 , 100 mg/kg orally). The oral administration of aqueous extract of leaves of *M. oleifera*(50 and 100mg/kg) exhibited significant ($p<0.05$) antinociceptive activity and dose dependent results at all the doses and standard group (Panadol 100 mg/kg) when compared to the control group. Also the extract was found to have a significant ($p<0.001$) reduction of painful sensation on hot plate method at all doses. potential. This study reveals that the leaves of *M. oleifera*are a potent analgesic which supports the traditional claim.

Keywords: *M. oleifera*, Panadol, Antinoceceptive, Hot plate test.

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Introduction

Moringaoleifera Lam (Moringaceae) is well known for its different therapeutic uses. The plant has been reported to known by regional names such as drumstick tree, sajiwan and sajna, and is a multipurpose tree known as nature's medicine cabinet. It possesses antitumor, antipyretic, antiepileptic, anti-inflammatory, antiulcer, antispasmodic, diuretic, antihypertensive, cholesterol lowering, antidiabetic, renal [1] and hepatoprotective activities [2].Many of the currently used medicines originate from natural products, especially plants. Drugs and plants are closely related to each other through the use of traditional medicines or ethnomedicines that are mainly prepared from plants. [3] Various types of plants have been used for several centuries worldwide not only as dietary supplements but also as traditional treatments for many diseases [4], [5], [6]. Indeed, the fact that traditional medicines have been widely used worldwide demonstrates the potential of plants as sources of bioactive compounds, including potential antitumor, antioxidant, antiobesity, and antimicrobial molecules. Among these plants, the widely cultivated *Moringaoleifera* (Moringa or drumstick tree), a rapidly growing perennial tree, was used by the ancient Romans, Greeks, and Egyptians, and has been naturalized from the tropics to the sub-Himalayan regions (e.g., India, Pakistan, Bangladesh, and Afghanistan), Oceania, Latin America, Africa and tropical Asia [7], [8], [9], [10]. Since ancient times for relieving pain caused by disease, injury and surgery Plants and herbs are the sources of not only crude drugs, but also bioactive compounds that could lead to novel drug structures. Because medicinal plants and herbs have been used, some of them contributed to the development of modern anesthesia to reduce pain [11]. *M. oleifera* has been used as an analgesic and anti-inflammatory in Indian folk medicine since centuries. Mechanism of action of analgesic effect is by the phytochemical components of leaf which have alkaloids, glycosides, phenols, saponins and tannins. The analgesic effect is due to inhibition of the activity of cyclooxygenase-2 (cox-2) which results in the inhibition of prostaglandins synthesis. The extract may also have interfered with G-protein mediated signal transduction, an analgesic mechanism unrelated to inhibition of prostaglandin synthesis. It also may have augmented the peripheral mechanism through interference with the formation of prostaglandins in the central nervous system. These mechanisms have been implicated in the forms of analgesia induced by non-steroidal anti-inflammatory drugs (NSAIDS), including Aspirin and Diclofenac. Are associate with greater side effects so there is arising scope for traditional medicines have lead to increase emphasis on the use of plant materials as a sources of medicines for wide variety of human ailments[12].Hence the present research work has been carried out to evaluate the analgesic of aqueous extract of leaves of *M. oleifera*to prove this activity scientifically by using various animal model.

Materıals And Methods

Collection of plant and extract preparation

Thefresh leaves of *M. oleifera* were collected in May 2021from Swani Benadem (Tripoli, Libya). The leaves were shade dried under appropriate condition. Shade dried powdered leaves (100 grams) mixed with 1 L of distilled water and macerated for 3 days with continuous shaking. After completion of maceration the dark green solution was filtred through filter paper according toYadu in 2010 with slight modifications[13].The filtrated solution was transferred to Buchner funnel fitted with Whatman No.1 filter paper where the filtration using by vacuum units.The homogeneous solution was kept in freezer over nightat -20 °C in tightly sealed jars after that were used in the feeding experiments.

The yield was calculated according to the doses and diluted with distilled water (dH_2O) to use as experimental doses.

Experimental Rats (model):

Male albino rat (n = 12), weighing between 120-180 g were obtained from local animal house of the National Medical Research Center, Alzawia, Libya. They were kept at standard laboratory conditions (at 20±25 C in both dark and light environments consecutively). Food and water were provided ad libitum. The animal were adapted to the condition for 10 days in the laboratory before examination .

Animal Grouping: The rats were divided randomly into four groups of three rats each: group 1: a control group which received the vehicle (distilled water) only. Group 2: a standrad groupwere treated orally with 100 mg of panadol (500 mg/kg). The two sample or treatment groups (groups 3, and 4) which received 50 and 100 mg/kg, respectively, of M. oleifera extract orally.

Procedure used for testing analgesic activity

Eddy's Hot plate method

The analgesic activity of M.oleifera was assessed using hot plate method of Eddy and Leimbach[14]. The temperature was maintained at 55 ± 0.2 c^0. This is hot enough to cause discomfort without tissue damage. Animals licked their forelegs and jumped as an indication of pain. These rats were treated with suspensions as follows: control group received distilled water. The test groups received 50 mg/kg and 100mg/kg ofM. oleifera extract. The standard group received Panadol 100 mg/kg by the oral route. One hour later they were placed on hot plate. The time taken by the animal to lick the fore or hind paw or jump out of the plate was taken as the latency time.

Statıstıcal Analysıs

Data in this study were analysed using Graph pad prism 5.01 soft ware (Graph pad soft ware Inc). One –way ANOVA of variance with Bonferroni post-boc testing (with correction for mltiple test) was performed . results were viewed as statistically significant with (p value 0.05).

Results

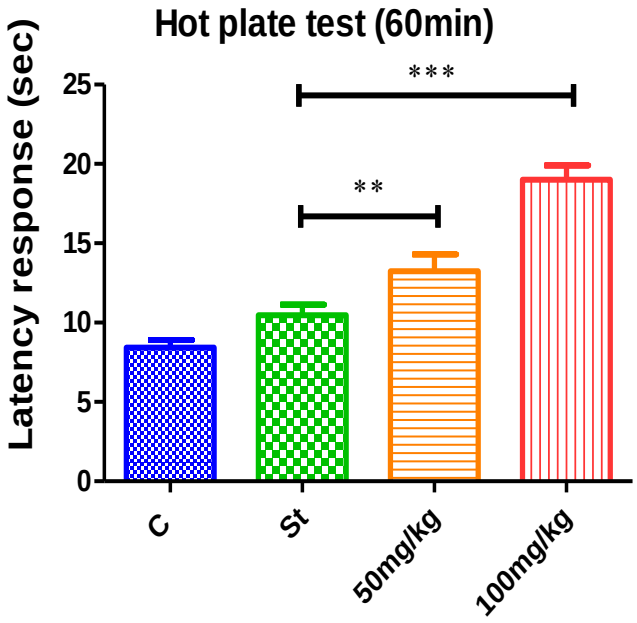
The experimental data were expressed as Mean ± SEM (standard error of mean). Pain induced by application of radiant heat in rats. The results of the hot plate method is given in [Table.1].M.oleifera

extract in dose of 100 mg/kg showed high significant increase in latency response(P***< 0.001) (19.00±0.52)compared to control and standard at 60 minutes onwards following administration of drug. Similar result was seen after 90 minutes. The doses of 50 mg/kg mildly increased the latency of hot plat response (*p < 0.05) (3.43±0.29) at 60 min compared to control and standrad . Whereas showed deccreasing the response at 90 min in the groups were fed both 50mg/kg and 100mg/kg of extract . The lowest values of analgesic activity was noticed in the group that receiving 50 mg of *aqueous extract of M.oleifera leaves* at 120min after treated (1.60±0.25) at the end of experiment.

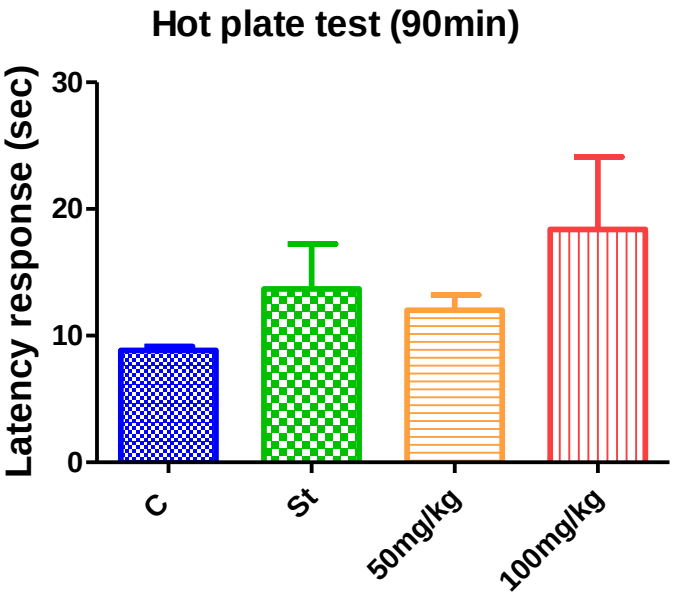
Table.1. *Analgesic activity: Effect of aqueous extract of M.oleifera leaves on thermal stimuli induced pain in rats by using hot plate test.*

Group / Dose (mg/kg)	Duration of latency of jumping response in (sec) at various time interval		
	x60min	90min	120min
Control	8.43±0.27	8.83±0.17	8.66±0.14
standrad	10.47±0.38	13.70±2.04	11.53±0.63
sample (50mg/kg)	13.23±0.61**	12.00±3.29**	12.23±1.21
sample(100mg/kg)	19.00±0.52***	18.40±0.15**	12.67±0.43

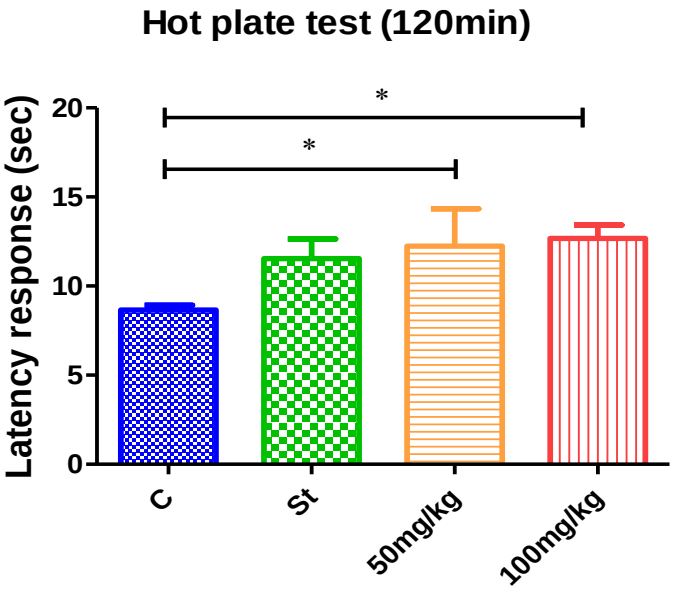
Each value represented in Mean ± SEM, *p < 0.05, **p < 0.01, ***p < 0.001 in comparison with control group (one way ANOVA).



Graphic.1. Analgesic activity (latency response) of *aqueous extract of M.oleifera*leaves at60min after treated.
c= control group. st=standrad group were fed panadol. 50mg/kg, 100mg/kg=sample group were fed *aqueous extract of M.oleifera*leaves.



Graphic .2. Analgesic activity (latency response) of *aqueous extract of M.oleifera*leaves at90min after treated.



Graphic.3. Analgesic activity (latency response) of *aqueous extract of M.oleifera*leaves at120min after treated.

Discussion

A study of analgesic efficacy of aqueose extract of M.oleifera leaves in male albino rats has been done in models of pain for study of both central and periferal pain. The stimulus was thermal (hot plate tests) [15].

NSAIDs act primarily on peripheral pain mechanisms but also in CNS to raise pain threshold.They are the most commonly used anti-inflammatory, antipyretic, analgesic drugs. Most NSAIDs block prostaglandin synthesis by inhibiting COX1 and COX2 non-selectively, but now some selective COX2 inhibitors has been developed. Of the common toxicities caused by NSAIDs due to inhibition of prostaglandin synthesis, gastric mucosal damage is most troublesome. This sometimes limits the use of this group of drugs in patients with chronic pain. The hot plate is a suitable for evaluation central visceral pain[16]. In the method of pain induction by application of radiant heat on hot plate, M.oleifera extract at the dose of 100mg. /kg showed highly significant increase in latency of which is comparable to that of panadol at the dose of 100 mg/kg of body weight [Tables 1].In a similar study by Manaheji et al; found significant reductions in both thermal hyperalgesia and mechanical allodynia in adult Male Wistar rats with CFA-induced arthritis compared to indomethacin 5mg/Kg [17]. Another study by Nitin G et al; found that the seeds of Moringa oleirera Lam. possess marked analgesic activity and is equipotent to standard drug (Panadol) [18].On contrast higher latency response values have been shown by Manoj in 2011 on albino mice treated with *ethanolic M.oleifera* extract at 400 mg/kg from 15 min to 90 min[19]. From this study, it can be concluded that the seeds of *Moringaoleirera*Lam. possess marked analgesic activity [Graphic. 1and 2] .The present study establishes the use of *Moringaoleifera*.leaves as regular analgesic.

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