Evaluation of Analgesic Activity of the ethanolic extract of *Butea Monosperma* flower in Albino Wistar Mice

[1]Sumreen Mujahid, [2]Safia Abidi, [3]Shaima Sultana Memon, [4]Muneeza Lodhi, [5]Muhammad Aitmaud uddolah Khan, [6]Tajwar Sultana

[1]Sumreen Mujahid
Department of Pharmacology, Baqai Medical College, Baqai University, Karachi, Pakistan.

[2]Safia Abidi
Department of Pharmacognosy, Faculty of Pharmacy and Pharmaceutical Sciences, University of Karachi, Karachi, Pakistan.

[3]Shaima Sultana Memon
Department of Pathology, Dow Medical College, Dow University of Health Sciences, Karachi, Pakistan.

[4]Muneeza Lodhi
Department of Pharmacology, Faculty of Pharmacy, Ziauddin University, Karachi, Pakistan.

[5]Muhammad Aitmaud uddolah Khan
Department of Pharmacology, Liaquat National Hospital & Medical College, Karachi, Pakistan.

[6]Tajwar Sultana
Department of Pharmacology, Jinnah Medical College, Peshawar, Pakistan.

Author's Contribution:

S.M. and S.A. designed the model and the computational framework and analysed the data. S.M. and S.S.M carried out the implementation. S.M. performed the calculations. S.M. and M.L. wrote the manuscript with input from all authors. M.A.K and T.S. conceived the study and were in charge of overall direction and planning.

Corresponding Author:

Sumreen Mujahid
Department of Pharmacology, Baqai Medical College, Baqai University, Karachi, Pakistan.

ABSTRACT

Background:

Butea monosperma, also known as palash, is a species of *Butea* that is native to tropical and subtropical regions of the Indian Subcontinent and Southeast Asia. It is frequently used to treat a variety of illnesses including pain.

Aim:

The present study was planned to evaluate to evaluate the analgesic activity of *Butea Monosperma* extract and standard drug Morphine

Material and Method:

The analgesic effect was assessed using tail flick method to induce pain in mice and the latency period was noted. The animals were divided into four groups, with each consisting of seven albino mice, viz. control, standard drug, extract-treated with two different dosage groups.

Results:

Butea Monosperma produces a dose-dependent analgesic effect using tail flick method in mice.

Conclusion:

The results denote *B. Monosperma* extract has significant analgesic. These results affirm the traditional use of *B.Monosperma* for the treatment of pain along with morphine.

Keywords: *Butea monosperma*, pain, analgesic, morphine, albino mice

INTRODUCTION:

Pain is a sensory modality that is mainly defensive in nature but frequently causes distress. It is the predominant symptom which draws the patient to the doctor. Analgesics alleviate pain symptoms but have little effect on the underlying cause (1).

Opioids are frequently used as painkillers during the perioperative period to reduce pain from surgery and labor (2). Morphine is the most widely used opioid analgesic for pain control(3). While used properly, it will provide sufficient pain relief for about 80% of patients (4). Nevertheless, several patients sometimes have to switch to an alternative opioid because of the intolerable side effects associated with morphine (5). These additional opioid substitutes are also combined with other drugs to boost their effectiveness with fewer side effects, though they may have dose-restriction effects owing to their combination (6).

The usage of medicinal herbs and plant compounds, as well as nutritional supplements, is becoming increasingly popular around the world. The research emphasis on herbal medicines has also increased in recent years, owing to the better efficacy of new plant-derived drugs and growing concerns about the side effects of traditional medicine (7, 8). Ancient medicine is widely used in Pakistan, and it has emerged as an important part of the country's culture and traditions (9).

Butea monosperma (Lam.) Taubert (Synonym *Butea frondosa Koen.ex Roxb*., Family: Fabaceae), also recognized as flame of the forest, is a medium-sized tree native to the subtropical and tropical region Subcontinent, Southeast Asia, and South Asia extending through Pakistan (10), India, Bangladesh, Nepal, Sri Lanka, Myanmar, Thailand, Laos, Cambodia, Vietnam, Malaysia and western Indonesia (11). Because of its antiseizure, anxiolytic, hypoglycemic, and anti-aging characteristics, this genus is frequently used in traditional medicine. Additionally, it has been found to be helpful as a laxative, antihelminthic, anti-inflammatory, and for neuropathic pain (12-14).

Consequently, the current study was performed in order to evaluate the analgesic potential of Butea Monosperma flower.

MATERIAL AND METHODS:

Plant Material: The fresh flowers of *Butea Monosperma* used in this study were purchased from the local market place of Karachi. The flowers were identified and authenticated by Prof. Dr. Iqbal Azhar, Department of Pharmacognosy, Faculty of Pharmacy and Pharmaceutical Sciences Karachi. The voucher specimen number is EZ186/93 and is kept in the Department of Pharmacognosy, University of Karachi**.**

Preparation of Extract: The flowers of *Butea Monosperma* were shade dried. After drying, the plant material was crushed in a crusher and powdered before being stored in amber-colored glass bottles for later use. The powdered material was sieved through a 40 mesh sieve. 300gm of *Butea Monosperma* powdered flower were soaked in Ethanol + water in a 70/30 percent (ethanol/water) ratio for 30 days to evaluate all constituents present. The mixture will then be filtered through

Whatmann filter paper. The solvent filtrate was evaporated using a rotary evaporator. The ethanolic extract was kept at 4 degrees Celsius in an amber-colored glass bottle for future research.

Experimental Animals: Albino Wistar mice (20-25g) of both sexes obtained from the Laboratory Animal House, Department of Pharmacognosy, Faculty of Pharmacy and Pharmaceutical Sciences, University of Karachi, were used for the experiment. The animals were housed under standard laboratory conditions at room temperature with relative humidity of 70–80%. The animal were acclimatize to customary research laboratory 12 hours day and night cycle. They were fed with standard commercial mice food diet and water *ad libitum*. Prior to the experiment, the animals were fasted for 12 h with water given *ad libitum* and weighed.

Grouping of Experimental Animals: Animals are separated into four sets and each set consists of 10 animals

Group 1: Control (0.5ml of Normal Saline)

Group 2: Standard dose of Morphine (10mg/kg)

Group 3*: Butea Monosperma* dose (50mg/kg)

Group 4: *Butea Monosperma* dose (100mg/kg)

The tail-flick technique was employed to evaluate the analgesic activity. Each mice was submerged in warm water kept at 51°C for about 2-3 cm from the distal end of its tail. The reaction time (in seconds) was the amount of time it took the mouse to flick its tail in pain. After administration of analgesic Morphine, the first reading was omitted and reaction time. The reaction time was recorded at 30 min after the administration of the treatments. The procedure was then repeated and take 5 to 7 readings of 30 mins interval.

RESULTS:

Traditional medicine used medicinal herb that are said to relieve pain and inflammation. Traditional practitioners use different parts of *Butea monosperma* to treat inflammation, pain, and convulsions (15).

The results of the analgesic activity of the ethanolic extract *Butea Monosperma* are shown in **Table 1**. The ethanol extracts of Butea monosperma flower is non-toxic and has a fatality rate of up to 2 g/kg in Swiss albino mice. Albino Mice treated with normal saline (negative control) did not show any significant difference in the reaction time on tail-flick throughout the 180 min observation. In comparison with the baseline values within the same treatment groups, the increase in reaction time at different time points significantly differed for morphine sulfate. Two different doses of *B. monosperma* flower extract were used, and it was discovered that the increased in tail flick method reaction time in a dose-dependent manner. Morphine sulphate and *B.Monosperma* extract-treated animals had significantly longer reaction times than saline-treated animals. The morphine reaction time was 2.48 seconds at 120 minutes, while the saline reaction time was 0.92 seconds and the *B.Monosperma* 1.16 sec (50mg/kg) and *B.Monosperma* 1.23 sec (100mg/kg) groups, respectively.

Table 1: Analgesic activity of ethanolic extracts of *Butea Monosperma* by using tail flick method

Group	Treatment	Dose (mg/kg per-oral)	Reaction Time (in Seconds) (mean±SEM)						
			0 min	30 min	60 min	90 min	120 Min	150 min	180 min
Control	Distilled water	0.5	0.89±0.01	0.90±0.01	0.91±0.01	0.91±0.01	0.92±0.01	0.90±0.00	0.90±0.00
Standard	Morphine	10	0.86±0.02	1.17±0.01***	1.79±0.01**	2.26±0.03*	2.48±0.02*	2.35±0.02*	2.08±0.04*
Test 1:	BM	50	0.90±0.01	0.97±0.01***	1.05±0.01**	1.11±0.01**	1.16±0.01**	1.11±0.00***	0.98±0.00***
Test 2:	BM	100	0.90±0.01	0.99±0.02***	1.14±0.01**	1.21±0.02*	1.23±0.02*	1.14±0.01**	0.99±0.01***

(N=7) each data suggest Mean±SEM.

One-way Analysis of Variance (ANOVA) followed by post hoc tukey's multiple comparison test is applied for statistical analysis.

*Significant at P<0.05, ** Significant at P<0.01 vs. Normal control

DISCUSSION:

An analgesic is a drug that relieves pain selectively without obstructing nerve impulse conduction, significantly altering sensory perception, or affecting consciousness (16). Centrally acting analgesics work by increasing the pain threshold and modifying the physiological reaction to pain. Peripherally acting analgesics, in contrast hand, work by preventing impulse production at the chemoreceptor site of pain (17). In this study, pain-state models using stimuli such as tail-flick method was used to evaluate for analgesic activity. This useful for demonstrating centrally mediated antinociceptive responses, which are primarily concerned with changes above the spinal cord level (18).

In the tail-flick model, the ethanolic extract from the flower *of B. Monosperma* showed considerable analgesic activity by prolonging the reaction time of albino mice compared to control (saline treated mice) at all points of time. Morphine sulphate was used as a standard drug, which is classified as a moderate to severe analgesic. In contrast to the control, morphine generated the most significant antinociception impact during all observation times, followed by the extract. The tail-flick method is based on the finding that morphine-like compounds selectively lengthen the response time of the typical tail-withdrawal effect in mice. This method is also beneficial for distinguishing central opioid-like analgesics from peripheral analgesics (19).

Sharma et al. discovered that when compared to the conventional drug ibuprofen and an untreated control, methanolic extract of Butea monosperma gum created dose dependent and substantial (P 0.01) analgesic activity (20). This other research on the stem bark of *B. monosperma* methanolic extract revealed anti-inflammatory and analgesic activity in a dose-dependent manner that is comparable to the conventional drug diclofenac sodium for carrageenin-induced paw edoema and acetic acid-induced writhing and Pentozocine for hot plate test model (21). According to another study by Arefin et al indicates that *BM* root extract has analgesic activity but may not have a

peripheral analgesic effect, which is facilitated by inhibiting the production and release of prostaglandins and other intrinsic substances (22). Moreover, using various animal models, an Indian study established the antinociceptive and anti-inflammatory activity of a petroleum ether extract of *Butea monosperma* leaves (23). Numerous studies have been conducted on *Butea Monosperma*, but to our knowledge, none of them have evaluated the analgesic potential of the flower extract.

CONCLUSION:

Finally, the ethanolic extract of *B.Monsperma* demonstrated analgesic activity, positively contributing to the traditional use medicinal plant in pain relief. More research is needed to identify the active compounds or constituent in this extract and to understand the mechanisms underlying its analgesic properties.

CONFLICTS OF INTEREST:

There was no conflict of interest among the authors.

REFERENCES:

1. Lynch ME, Watson CPN. The pharmacotherapy of chronic pain: a review. Pain Research and Management. 2006;11(1):11-38.
2. Koepke EJ, Manning EL, Miller TE, Ganesh A, Williams DG, Manning MW. The rising tide of opioid use and abuse: the role of the anesthesiologist. Perioperative Medicine. 2018;7(1):1-10.
3. Murphy PB, Bechmann S, Barrett MJ. Morphine. StatPearls [Internet]: StatPearls Publishing; 2021.
4. Karamchandani K, Klick JC, Dougherty ML, Bonavia A, Allen SR, Carr ZJ. Pain management in trauma patients affected by the opioid epidemic: a narrative review. Journal of trauma and acute care surgery. 2019;87(2):430-9.
5. Rigal M, Martino R, Hain R. Opioids and the World Health Organization pain ladder. Oxford Textbook of Palliative Care for Children. 2021:176.
6. Ehieli E, Yalamuri S, Brudney CS, Pyati S. Analgesia in the surgical intensive care unit. Postgraduate medical journal. 2017;93(1095):38-45.
7. Geneva S. WHO Guide lines on Safety Monitoring of Herbal Medicines in Pharmacovigilance Systems.[Google Scholar]. 2004.
8. Ekor M. The growing use of herbal medicines: issues relating to adverse reactions and challenges in monitoring safety. Frontiers in pharmacology. 2014;4:177.
9. Hussain S, Malik F, Khalid N, Qayyum MA, Riaz H. Alternative and traditional medicines systems in Pakistan: history, regulation, trends, usefulness, challenges, prospects and limitations. A compendium of essays on alternative therapy London: InTech. 2012:67.
10. Akram M, Akhtar N, Asif H, Shah PA, Saeed T, Mahmood A, et al. Butea monosperma Lam.: A review. J Med Plants Res. 2011;5:3994-6.
11. Tandon R, Shivanna K, Mohan Ram H. Reproductive biology of Butea monosperma (Fabaceae). Annals of Botany. 2003;92(5):715-23.

12. GURAV SS, Gulkari VD, Duragkar NJ, Patil AT. Antimicrobial activity of Butea monosperma Lam. gum. 2008.
13. Maurya R, Yadav DK, Singh G, Bhargavan B, Murthy PN, Sahai M, et al. Osteogenic activity of constituents from Butea monosperma. Bioorganic & medicinal chemistry letters. 2009;19(3):610-3.
14. Thiagarajan VR, Shanmugam P, Krishnan UM, Muthuraman A, Singh N. Ameliorative potential of Butea monosperma on chronic constriction injury of sciatic nerve induced neuropathic pain in rats. Anais da Academia Brasileira de Ciências. 2012;84:1091-104.
15. Rahmatullah M, Noman A, Hossan MS, Rashid M, Rahman T, Chowdhury MH, et al. A survey of medicinal plants in two areas of Dinajpur district, Bangladesh including plants which can be used as functional foods. American Eurasian Journal of Sustainable Agriculture. 2009;3(4):862-76.
16. Patidar A, Birla D, Patel V, Chaturvedi M, Manocha N. A Review on advantages of Natural Analgesics over Conventional Synthetic Analgesics. International Journal of Pharmacy & Life Sciences. 2014;5(5).
17. Tripathi K. Essentials of medical pharmacology: JP Medical Ltd; 2013.
18. Wakeel OK, Abe AI, Awosan OB, Olapade MK, Olatoyan-Layonu TJ, Olowe OA, et al. Anti-Nociceptive and Anti-Inflammatory Effects of Stem Bark Extract of Ficus Capensis Thunb (Moraceae) by Bioactivity Fractionation. Anti-Inflammatory & Anti-Allergy Agents in Medicinal Chemistry (Formerly Current Medicinal Chemistry-Anti-Inflammatory and Anti-Allergy Agents). 2021;20(2):206-18.
19. Bilel S, Azevedo N, Arfè R, Tirri M, Gregori A, Serpelloni G, et al. In vitro and in vivo pharmacological characterization of the synthetic opioid MT-45. Neuropharmacology. 2020;171:108110.
20. Kumar SA, Maya C, Rajesh K, Bhupendra C, Kovidendra K, Vipin A. Evaluation of antiinflammatory and analgesic activity of Butea monosperma gum.
21. William CM, Krishna Mohan G. Antiinflammatory and analgesic activity of Butea monosperma (Lam) stem bark in experimental animals. Pharmacologyonline. 2007;2:88-94.
22. Arefin SMA, Islam MA, Rashid STB, Rayhan MA, Hossen MM, Alam MJ, et al. Analgesic, anti-inflammatory, anticonvulsant and CNS effect of the ethanolic extract of Butea monosperma roots. Jahangirnagar University Journal of Biological Sciences. 2015;4(1):9-18.
23. Rathod VD, Bhangale JO, Bhangale PJ. Preliminary evaluation of antinociceptive and anti-inflammatory activities of petroleum ether extracts of Butea monosperma (L.) leaves in laboratory animals. World Journal of Pharmaceutical Sciences. 2017:246-52.