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ORIGINAL ARTICLE

Evaluation of analgesic activity of the ethanolic leaves extract of *Aglaia elaeagnoidea*

G.MahaLakshmi Priya*, K.shanmukha Sai Kumar, T.Manasa, J.Sai Harshini, C.Sirisha, Hima Saila.M
Department of Pharmacology, Sri Padmavathi School of Pharmacy, Tiruchanoor, Tirupathi
Corresponding author's Mail ID:-mahalakshmipriya.g@gmail.com

ABSTRACT
This study aims to evaluate the analgesic activity of the ethanolic leaves extract of Aglaia elaeagnoidea. This plant is acknowledged for its secondary metabolites and it belongs to the family Meliaceae. It has many uses such as anti-inflammatory, anti-oxidant, hepatoprotective, anti-pyretic, astringent etc. Ethanolic extract was prepared from the leaves of this plant and phytochemical screening was done which shows alkaloids, phenols, flavonoids, tannins, coumarins, quinones, leucoanthocyanins, Carbohydrates and fatty acids are present. For the evaluation of analgesic activity three evaluation methods were used acetic acid induced writhing test, tail-immersion test and formalin test. The ethanolic leaves extract was given at the dose of 150 and 300 mg/kg p.o. and Ibuprofen was used as standard drug at the dose of 100mg/kg p.o. The extract showed analgesic activity in dose dependent manner($p<0.05$) indicating that the plant possess analgesic activity. Therefore we can conclude that the phytochemical constituents present in the extract are responsible for analgesic activity
Key words:- *Analgesic activity, Aglaia elaeagnoidea, acetic acid induced writhing test, tail-immersion test, formalin test, Ibuprofen*

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INTRODUCTION

Plants are the main source of drugs in Indian system of medicine and other ancient systems within the world. Rig-Veda (2500–1800 BC) gives the details of healing properties of medicinal plants, while Charka Samhita and Sushruta Samhita unfurl elaborately on several medicinal herbs. At present, the appliance of plant-derived natural products within the synthesis of medicine becomes a drift. Hence, there's a requirement to update data on the properties, uses, effectiveness, and safety of medicinal plant products [1].

The word Algesia is Derived from the Greek word *algesis* which means sensitivity to pain [2].

Pain is a ill-defined, disabling accompaniment of many medical conditions. Pain is the response given to an external or internal noxious stimulus. Analgesics are the drugs which possess significant pain relieving properties by acting within the CNS or on peripheral pain receptors without significantly affecting consciousness. Analgesics are divided into two groups.

1. Narcotic/ Opioid /Morphine like analgesics.
2. Non-narcotic/Non-steroidal anti-inflammatory agents – analgesics – antipyretic agents [3]

Most of the drugs used at present are synthetic in nature and prolonged use of which causes severe side effects and exhibit toxic effects. In this regard novel possibility of evaluating herbs in analgesic activity arises.

Aglaia elaeagnoidea is an evergreen tree. This genus belongs to *Meliaceae* family. They are located from Indo-Malaysia to Pacific Islands. In India they're found within the dense and moist forests in Western and Eastern Ghats. It grows up to 1015 m tall, with greyish brown bark. Leaves are alternate, elliptic, or

compound with 3-7 leaflets. Roundish yellow flowers are borne in panicles, Fruit Berry. Genus *Aglaia* is represented by quite 100 species belonging to the Meliaceae.
Certain species of *Aglaia* such as *A. Lawii. A.elaeagnoidea* may be a traditional medicinal plant having been used for the treatment of bacterial infection, liver, tumour diseases and headaches[4] Among all the known medicinal plants, species of *Aglaia* genus are known for its secondary metabolites. Hence our present study was aimed to evaluate the analgesic activity of ethanolic leaves extract of *Aglaia elaeagnoidea*

MATERIAL AND METHODS

Plant material:-

Fresh leaves of plant *Aglaia eleagnoidea* were collected locally from a nearby medicinal garden. The taxanomic identification of the plant was confirmed and processed further. Collected leaves were washed thoroughly with water for 2-3 times and dried under shade for 30 days. Dried leaves were powdered and stored in room temperature in sterile bottle

Preparation of ethanolic extract:-

100g of leaf powder was added to 900ml of 95% ethanol. The mixture was covered and shaken every 30 min for 6 hours and then allowed to stand for 48 hours for extraction. The mixture was then separated by passing through whatmann's filter paper, after which the filtrate was evaporated to dryness under air pressure. The dried crude Ethanolic extract of *Aglaia elaeagnoidea* [EAE] were stored in the refrigerator at 4^0C under aseptic conditions for subsequent use[5] and subjected to preliminary phytochemical screening

Procurement of Animals:-

Healthy Male Wistar strain rats of about 150–250 g were used for the study, they were purchased from Biogen laboratory animal facility Bangalore. The animals were caged individually and kept in air conditioned room at temperature of 22±2^{0}C with 50%±10% relative humidity with 12hrs light and dark cycle. Throughout the study animals were maintained at normal laboratory conditions and at standard rat pellet diet and drinking water *ad libitum*. The experimental protocol was approved by the Institutional Animal Ethical Committee of Sri Padmavathi School of Pharmacy, Tiruchanoor, Tirupathi (No:-SPSP:1016/PO/Re/S/06/CPCSEA/2020/016)

Evaluation of Analgesic activity

Experimental design:-

Male Wistar rats were divided into four groups control, Standard, test-1, test-2, the animals were marked individually. Food was withdrawn 12 hours prior to drug administration till the completion of experiment. The test-1 (EAE-150mg/kg), test-2 (EAE-300mg/kg) and standard drug Ibuprofen(100mg/kg)[1] were given orally.
150mg/kg and 300mg/kg were chosen as low dose and high dose respectively based on the data obtained from the study of K. Manikya Kumari and V. Padmaja. "Evaluation of hepatoprotective activity of *Aglaia elaeagnoidea(A. Juss)* benth stem extract against carbon tetrachloride induced hepatotoxicity in albino rat" **2012;**2(3):117-124[4]

Acetic Acid induced Writhing Test:-

After 60 min of drug administration, writhing was induced by intraperitoneal injection of 1% acetic acid in volume of 0.1ml/10g body weight. The writhing episodes were recorded for 30 minutes; stretching movements consisting of arching of back, elongation of body and extension of hind limbs were counted[6]

$$\%In\square = \frac{no.of\ writ\square es\ in\ control\ group - no.of\ writ\square es\ in\ treated\ group}{no.of\ writ\square es\ in\ control} \times 100$$

Formalin test:-

20µl of 2.5% formalin solution made in normal saline was injected intraplantarly under the surface of right hind paw after 1 hr of drug administration. After formalin was given as intraplantar injection, the animals were placed in a glass cylinder, 20cm in diameter and therefore the time spent in licking the injected paw was monitored and recorded for the period of 0-5 min(early phase of licking)and 15-30 min (late phase of licking) [7]

Tail immersion test

After 60 min of administering the drug, the lower 5cm portion of the tail which is marked was immersed in waterbath of exactly of 55^{0}C.Within a few seconds the rat reacts by withdrawing the tail. The reaction time is recorded in seconds by a stop watch. The cut off time is 15 seconds[6]

Statistical analysis

The results are expressed as mean±SEM (n=6). Statistical analysis was performed using one way ANOVA and subsequent Dunnett's test. P values less than 0.05 were considered significant

RESULTS

Phytochemical Screening

The dried extracts of the leaves were subjected to preliminary phytochemical screening which showed Alkaloids, Flavonoids, Tannins, Saponin glycosides and Carbohydrates are present.

DISCUSSION

The EAE suppressed the pain by showing significant decrease in the number of writhings at the doses of 150mg/kg *p.o.* and 300mg/kg *p.o.* when compared with control and standard which may due to the suppression of prostaglandins, lipoxygenases, serotonin, bradykinins, histamine or Substance P.

In the tail immersion model EAE increased the tolerance capacity to pain produced by thermal stimuli and hence also indicates the possible actions of it on the higher centre. Both the test doses 150mg/kg *p.o.* and 300mg/kg *p.o.* showed significant increase in reaction time when compared to control and standard.

Formalin test is a tonic pain model that is often used in analysing antinociceptive activity which is very effective model in evaluating the analgesic activity of the plant extract. The early phase of the formalin test indicates the transmission of nociceptive impulses while the second phase of the formalin test represents the events of central. Centrally acting analgesics have effects on both phases whereas peripherally acting analgesics affects only the first phase. This is because the injection of formalin resulted in the release of various neurotransmitters including glutamate and aspartate in the dorsal horn[8] This study shows that EAE inhibits pain in both the phases. There is a significant decrease in the time spent in pain at the doses 150mg/kg *p.o.*and 300mg/kg *p.o.* when compared with control and standard

Table .1:-Phytochemical screening

S.no	Test	Result
1	Test for Alkaloids	+
2	Test for Flavonoids	+
3	Test for Athraquinone glycosides	_
4	Test for Cardiac glycosides	_
5	Test for Saponin glycosides	+
6	Test for Tannins	+
7	Test for Carbohydrates	+
8	Test for Amino acids	_
9	Test for Triterpenoids and Steroids	_

Table .2:-Evaluation of analgesic activity of EAE using acetic acid induced writhing model

Treatment	Dose	Number of writhes in 1 hr	%Inhibition
Control(Normal)	-	69.7 ± 1.80#	0%
Std(IBUPROFEN)	`100mg/kg	34.7 ± 1.82*#	50.21%
EAE	150mg/kg	54.2 ± 1.08*#	22.23%
EAE	300mg/kg	44.3 ± 1.41*#	36.44%

Each value represents mean ± SEM, n=6, Experimental group was compared with control #p<0.05, considered significant. Experimental group was compared with standard *p<0.05, considered significant

Table no.3:-Evaluation of analgesic activity of EAE using tail immersion test

Treatment	Dose	Reaction time(in sec)
Control(Normal)	-	0.993 ± 0.0456#
Std(IBUPROFEN)	`100mg/kg	4.71 ± 0.460*#
EAE	150mg/kg	2.32 ± 0.131*#
EAE	300mg/kg	2.40 ± 0.190*#

Each value represents mean ± SEM, n=6, Experimental group was compared with control #p<0.05, considered significant. Experimental group was compared with standard *p<0.05, considered significant.

Table 4:-Evaluation of analgesic activity of EAE using Formalin induced paw licking test

Treatment	Dose	Time spent in pain [early phase]in seconds	Time spent in pain [Late phase] in seconds
Control	-	141±1.32#	84.7±1.25#
Std(Ibuprofen)	`100mg/kg	34.1±0.762*#	25.2±2.01*
EAE	150mg/kg	88.2±2.84*#	41.9±0.698*#
EAE	300mg/kg	75.7±0.942*#	34.9±1.46*#

Each value represents mean ± SEM, n=6, Experimental group was compared with control #p<0.05, considered significant. Experimental group was compared with standard *p<0.05, considered significant.

CONCLUSION

Analgesic activity of ethanolic leaves extract of the *Aglaia elaeagnoidea* was studied using Acetic acid induced writhing test, Formalin induced paw licking test, tail immersion test. The present study shows that phytochemical constituents of the *Aglaia elaeagnoidea* are responsible for the analgesic activity. Further studies are necessary to isolate particular active phytochemical ingredients which helps in designing a potent medication for Algesia

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COMPETING INTERESTS

The authors have declared that no competing interest exists

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