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Investigation of Analgesic Activity of Roots and Flowers of *Plumeria Pudica* Linn.

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Abstract

The plant *Plumeria pudia* Linn. known as Nagchampa is one of the aromatic and medicinal flowering plant family Apocynaceae is one of the species belongs to genus *Plumeria*. The plant is used by the tribal and rural people of our country as a source of medicine. Some of the remote peoples also used the various part of the plant in the treatment and management of various microbial and inflammations conditions. The present work was undertaken to investigate the analgesic activity of extracts of P. pudica. Results indicate that the AE possesses higher activity than other extract and found to be significant when compared with the standard drug

Key-words: *Plumeria pudica*, Root, Flowers, analgesic activity

Introduction

India has a rich treasure of medicinal plants due to the diversity of agro-climatic conditions spread over the country from tropical to temperate zones, costal plains to high attitudes and semi-arid to highly humid evergreen forests, therefore, it is an advantageous position to produce a number of crude drugs. It is evident that many valuable herbal drugs have been discovered by knowing that a particular plant was used by the ancient folk healers for the treatment of some kind of ailments. Moreover, the medicinal plant wealth are our national heritage and it seems to be the first and foremost line of defense for the treatment of various diseases mostly tribal and rural communities and is a worth scientific study [1-2]

Plumeria pudica L. is a fast-growing, medium size tree that is botanically belongs to family Apocynaceae. Plant possess, iridoid glycosides, sterols, carbohydrates, tannins, triterpenoids and alkaloids as active constituents. The plant is used for the cure of rheumatism, diarrhoea, blennorhea, venereal disease, leprosy, psychosis and diuresis etc. Plumeria species have also been investigated for isolation of irridoids and triterpenoids, which exhibited algicidal, antibacterial and cytotoxic activities. [3-5] So far, no any systematic was done to evaluate the analgesic activity, therefore, the present work was undertaken.

Material and Methods

Acute Toxicity Studies of Extracts

The mice were used for acute toxicity study as per OECD guidelines 423. The animals were fed with standard pellet diet (Hindustan lever Ltd. Bangalore) and water ad libitum. All the animals were housed in polypropylene cages. The animals were kept under alternate cycle of 12 hours of darkness and light. The animals were acclimatized to the laboratory condition for 1 week before starting the

experiment. The experimental protocols were approved by Institutional Animal Ethics Committee after scrutinization. [6]

Analgesic Activity

Animals

Female Wistar rats of (200-250 gm) were procured and = maintained under ideal feeding and management practices in the laboratory. The animals were fed with standard pellet diet (Hindustan lever Ltd. Bangalore) and water *ad libitum*. All the animals were housed in polypropylene cages. The animals were kept under alternate cycle of 12 hours of darkness and light. The animals were acclimatized to the laboratory condition for 1 week before starting the experiment. The experimental protocols were approved by Institutional Animal Ethics Committee after scrutinization.

Study Design

Hot plate

Animals were divided into different groups, each group containing six animals each. Group I served as the positive control with no protection. Group II animals received the standard drug of Indomethacine 5 mg/kg body weight, whereas group other groups were orally administered the various plant extracts viz., Pet. Ether, Chloroform, ethanolic and aqueous extract of *Plumeria pudica* Linn. at the dose of 250 and 500 mg/kg body weight respectively. The temperature of the hot plate was maintained 55±1 ºC, mice were placed on the hot plate and time in seconds for paw licking or jumping was recorded as basal reaction time. Cut off time in the absence of response was 15sec to prevent the animals being burnt. The reaction time in seconds (latency period) was observed on hot plate, the time taken for mouse to react to the thermal pain by licking its paw or attempting to jump out. Observations were made before and after administration of respective drugs at an interval of 60 min. [7-8]

Tail Flick Method

The animals were tested for tail flick by Analgesiometer (Techno Electronics, Lucknow, India) as it was described earlier (Miranda et al, 2003)21 . The basal time was noted at first for each animal. Current through the naked nichrome wire was set at 5 Amp over which 1-2 cms from the tip of the tail was exposed to check out the response. The cut off time was set at 10 sec to prevent any tissue damage. The time (in second) required for the animal to withdraw (flick) its tail from the heat source was measured. The reaction time was noted in minutes after the animals were treated orally with various doses of extract and with Indomethacine (5 mg/kg). Normal saline (0.1ml/10gm) served as control group. [7-8]

Statistical analysis

All the values ware statistically analyzed by one-way analysis of variance (ANOVA) followed Bonferroni's post hoc test. Comparison between control and drug treated groups were considered to be significant (*$P<0.01$). All values are expressed as mean ± SEM.

Results and Discussion

The PEE, CE, EE and AE of PRR= *Plumeria pudica* Linn. Roots and PPF= *Plumeria pudica* Linn. Flowers were screened for acute toxicity study by OECD guideline no. 423 for determination of

$LD_{50.}$ The results showed that the PEE, CE, EE and AE of PRR= *Plumeria pudica* Linn. Roots and PPF= *Plumeria pudica* Linn. Flowers were belonging to category-5(unclassified). Hence, LD_{50} was 5000 mg/kg, therefore, ED_{50} was 500 mg/kg. Therefore doses of 500 mg were selected for present investigation. The results were presented in table 1 & 2.

Table 1: Determination of LD_{50} and ED_{50} of Extract of *Plumeria pudica* Linn. Roots

S/No.	No. of Animals	Extract Dose (mg/kg)	No. of death of animals			
			PEE	CE	EE	AE
1.	3	5	0	0	0	0
2.	3	50	0	0	0	0
3.	3	300	0	0	0	0
4.	3	2000	0	0	0	0
5.	3	5000	0	0	0	0

Table 2: Determination of LD_{50} and ED_{50} of Extract of *Plumeria pudica* Linn. Flowers

S/No.	No. of Animals	Extract Dose (mg/kg)	No. of death of animals			
			PEE	CE	EE	AE
1.	3	5	0	0	0	0
2.	3	50	0	0	0	0
3.	3	300	0	0	0	0
4.	3	2000	0	0	0	0
5.	3	5000	0	0	0	0

The PEE, CE, EE and AE of PRR= *Plumeria pudica* Linn. Roots and PPF= *Plumeria pudica* Linn. Flowers were screened for analgesic activity in animal models and the results are summarized in Table. The result obtained indicates that the extract found to have significant analgesic activity. The PEE, CE, EE and AE of PPF= *Plumeria pudica* Linn. Flowers at the test doses 250 and 500 mg/kg b.w. produced profound analgesic activity and is more potent, when compared with the PEE, CE, EE and AE of PPF= *Plumeria pudica* Linn. Roots at the test doses 250 and 500 mg/kg b.w. when compared to standard drug and control group.

Table 3: Analgesic effect of *Plumeria pudica* Linn. Roots

Group	Reaction time (Sec)					
	Hot Plate			Tail Flick		
	Pre treatment	1 hr	3 hr	Pre treatment	1 hr	3 hr
Control	11.8 ± 0.8	11.1 ± 0.7	12.3 ± 0.9	2.1 ± 0.2	2.5 ± 0.2	2.3 ± 0.1
Standard 5mg/kg	10.5 ± 1.2	18.3 ± 1.7***	19.4 ± 1.4***	2.2 ± 0.1	3.3 ± 0.1***	3.5 ± 0.1***
PEEPPR 250 mg/kg	12.1 ± 0.4	12.1 ± 0.7	12.9 ± 0.7	2.1 ± 0.1	2.2 ± 0.1	2.2 ± 0.5

PEEPPR 500 mg/kg	12.6 ± 0.7	12.9± 0.5	13.5 ± 0.2	2.2 ± 0.4	2.2 ± 0.5	2.3 ± 0.1
CEPPR 250 mg/kg	12.5 ± 0.4	12.7 ± 0.7	12.9 ± 0.6	2.1 ± 0.3	2.1 ± 0.2	2.1 ± 0.4
CEPPR 500 mg/kg	12.8 ± 0.7	12.9 ± 0.5	13.2 ± 0.1	2.2 ± 0.4	2.4 ± 0.5	2.5 ± 0.4
EEPPR 250 mg/kg	11.6 ± 0.1	12.8 ± 0.7	12.8 ± 0.5	2.0 ± 0.3	2.0 ± 0.1	2.2 ± 0.5
EEPPR 500 mg/kg	11.9 ± 0.7	12.9 ± 0.5	13.0 ± 0.1	2.1 ± 0.4	2.3 ± 0.5	2.3 ± 0.2
AEPPR 250 mg/kg	11.5 ± 0.2	13.4 ± 0.8	14.1 ± 0.3	1.8 ± 0.6	2.8 ± 0.7	2.8 ± 0.1
AEPPR 500 mg/kg	11.0 ± 1.2	13.9 ± 0.6	14.1 ± 0.7	1.9 ± 0.3	2.9 ± 0.4	3.1 ± 0.4

All values are expressed as mean ± S.E.M (n=6), ***P<0.001 as compared control, **P<0.01 as compared control, One-way ANOVA followed by Bonferroni multiple comparison test

Table 4: Analgesic effect of *Plumeria pudica* Linn. Flowers

Group	**Reaction time (Sec)**					
	Hot Plate			**Tail Flick**		
	Pre treatment	**1 hr**	**3 hr**	**Pre treatment**	**1 hr**	**3 hr**
Control	11.8 ± 0.8	11.1 ± 0.7	12.3 ± 0.9	2.1 ± 0.2	2.5 ± 0.2	2.3 ± 0.1
Standard 5mg/kg	10.5 ± 1.2	18.3 ± 1.7***	19.4 ± 1.4***	2.2 ± 0.1	3.3 ± 0.1***	3.5 ± 0.1***
PEEPPF 250 mg/kg	12.2 ± 0.4	12.5 ± 0.7	13.4 ± 0.7	2.0 ± 0.3	2.1 ± 0.1	2.1 ± 0.5
PEEPPF 500 mg/kg	12.5 ± 0.7	12.8± 0.5	13.4 ± 0.3	2.1 ± 0.4	2.2 ± 0.5	2.4 ± 0.2
CEPPF 250 mg/kg	12.4 ± 0.5	12.6 ± 0.7	12.9 ± 0.8	2.0 ± 0.3	2.1 ± 0.1	2.1 ± 0.5
CEPPF 500 mg/kg	12.9 ± 0.7	12.9 ± 0.5	13.3 ± 0.2	2.1 ± 0.4	2.2 ± 0.5	2.4 ± 0.2
EEPPF 250 mg/kg	11.5 ± 0.3	12.6 ± 0.7	12.4 ± 0.5	2.0 ± 0.3	2.1 ± 0.1	2.1 ± 0.5
EEPPF 500 mg/kg	11.9 ± 0.7	12.8 ± 0.5	13.1 ± 0.2	2.1 ± 0.4	2.2 ± 0.5	2.4 ± 0.2
AEPPF 250 mg/kg	11.4 ± 0.2	13.5 ± 0.8	14.2 ± 0.6	1.9 ± 0.6	2.9 ± 0.7	2.9 ± 0.2
AEPPF 500 mg/kg	10.9 ± 1.2	14.2 ± 0.6	14.3 ± 0.9	2.0 ± 0.1	3.1 ± 0.6	3.2 ± 0.6

All values are expressed as mean ± S.E.M (n=6), ***P<0.001 as compared control, **P<0.01 as compared control, One-way ANOVA followed by Bonferroni multiple comparison test

Conclusion

The PEE, CE, EE and AE of root and flowers of plant of *Plumeria pudica Linn.*. were screened for acute toxicity study by OECD guideline no. 423 for determination of LD_{50} and ED_{50}. No mortality in either extract was observed at the dose of 5000 mg/kg, therefore, the LD_{50} was 5000 mg/kg and ED_{50} was 250 mg/kg. Hence, two doses of 250 and 500 mg were selected for further investigation. The PEE, CE, EE and AE of root and flowers of plant of *Plumeria pudica Linn.*. were employed study analgesic, anti-inflammatory, *in vitro* anthelmintic activity and anti-microbial activity of the extract were determined and it was found that AE possesses higher activity than other extract and found to be significant when compared with the standard drug

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