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Research Article

Phytochemical Investigation and Analgesic Activity of Leaf Extracts of *Jatropha Gossypifolia* Linn.

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Abstract

Jatropha gossypifolia L. is evergreen plant belonging to the family *Ephorbeaceae*, common in waste places throughtout India, in the souerthen part it is cultivated chiefly for hedges. The present study was undertaken with the objective to phytochemical investigation and the analgesic activity of the dichloromethan and methanolic leaf extracts of *Jatropha gossypifolia* Linn. in mice and rats at different doses. The preliminary phytochemical study revealed that plant powder contains carbohydrats, gum and mucilage, alkaloid, flavonoid, saponin, phytosterols, tannin and phenolic compound, whereas dichloromethane extract contains carbohydrate, flavonoid, phytosterol, saponin, tannin, phenolic compound and methanolic extract contains carbohydrate, alkaloid, flavonoid, phytosterols, saponin, tannin and phenolic compound.

Analgesic study shows significant activity in 200 and 400 mg/kg dose of methanolic extract but was found insignificant in 200 and 400 mg/kg dose of dichloromethan extract when compared with standard analgesic drug aspirin. The activity may be due to presence of alkaloid, flavonoid, phytosterols, saponin, tannin and phenolic compound in methanolic extract. The results are expressed as mean increase in latency after drug administration ± SEM in terms of seconds and analyzed for ANOVA and post hoc Dunnet's t-test. Differences between groups were considered significant at $P < 0.05$ levels and $P<0.01$ as highly significant.

Key words: *Jatropha gossypifolia* Linn., phytochemical investigation, post hoc Dunnet's t-test.

Introduction

Jatropha is a genus of approximately 175 succulent plants, shrubs and trees (some are deciduous, like *Jatropha curcas* L.), from the family *Euphorbiaceae*. The name is derived from (Greek jatros = physician and trophe = nutrition), hence the common name physic nut. This evergreen plant is common in waste places, It is also common in waste lands, roadsides, poorly tended agricultural fields, and river overflow areas throughtout India, in the sourthen part,it is cultivated chiefly for hedges.

Jatropha gossipifolia Linn. is adapted to a variety of habitats. Often cultivated as an ornamental, grows on nearly all types of soils within its range. It is intolerant of shade. Although it may survive for a season in moderate shade, they need full or nearly full sun for longer-term survival and fruiting.

The seeds are toxic to humans. The plants contain several toxic compounds, including curcin, lectin, saponin etc. Despite this, the seeds are occasionally eaten after roasting, which reduces some of the toxicity. Its sap is a skin irritant, and ingesting as as few as three untreated seeds can be fatal to humans.

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It possesses significant anticancer, hepatoprotective and pesticidal activity [1-2]. The leaf decoction of *Jatropha gossypifolia* is used for bathing wounds. The stem sap stops bleeding and itching of cuts and scratches [3-5]. The roots are employed against leprosy, as an antidote for snakebite and in urinary complaints. A decoction of the bark is used as an emmenagogue and leaves for stomachache, venereal and as blood purifier [6-9].

Therefore, the present study was undertaken with the objective to phytochemical investigation and the analgesic activity of the dichloromethan and methanolic leaf extracts of *Jatropha gossypifolia* Linn. in mice and rats at different doses.

Material and Methods

Collection of plant

The plant material used for investigation was collected locally from Jaipur, Rajasthan, India in the month of August 2009. The plant was identified with the help of available literature and authenticated from Rajasthan university, Jaipur. The specimen (RUBL 20618) of plant has been submitted to the Department of Botany (Herbal Sciences), Raj.university, Jaipur. The plants were dried in shade for 15 days and then the leaf parts of the plants were taken for the study.

Preparation of extracts

The dried plant material leaf were subjected to cold meaceration with dichloromethane and methanol as solvent respectively. About 500 gm of dry powder was deffatted with Petroleum ether .The deffating was continued for 7-8 days with occasional shaking. The Petroleum ether extract was filtered. The marc left after Petroleum ether deffating was dried, then extracted with dichloromethane and methanol respectively. The extracts were collected and concentrated to a semisolid mass under reduce pressure.The residue obtained from extracts are dichloromethane and methanol extract were used for this study. For in vivo studies, the dichloromethane and methanolic extract of *Jatropha gossypifolia* Linn. were prepared by suspending in normal saline.

Phytochemical test

The extract were subjected to preliminary phytochemical investigation to identify various phytoconstituent i.e carbohydrate, protein, amino acid, alkaloid, glycoside, gum and mucilage, tannin and phenolic compound, fats and oil, steroid etc. present in leaves by using standard tests.[10,11]

Experimental animals

Thirty six Wistar rats of both sexes, weighing 150 – 200 gm were used for the study. The animals were kept in polypropylene cages in a room maintained under controlled atmospheric conditions. The animals were fed with standard diet (Hindustan liver, Mumbai, India) and had free access to clean drinking water. Pharmacological study was approved by Animal Ethical Committee of School of Pharmacy; Suresh Gyan Vihar University, with CPCSEA Reg no. 1234/a/.08/CPCSEA.

Acute toxicity studies

The acute oral toxicity studies were performed to study the acute toxic effects and to determine minimum lethal dose of the drug extracts. Swiss albino mice of either sex weighing 18-25 g were used for the study. The dichloromethan and methanol extracts were administered orally to different groups of overnight fasted mice at the doses of 30, 100, 300, 600 and 1000 mg/kg body weight. After administration of the extracts, animals were observed continuously for the first three hours for any toxic manifestation. Thereafter, observations were made at regular intervals for 24 hrs. Further the animals were under investigation up to a period of one week [12]. This studies show that drug is safe up to the dose of 1000 mg/kg with both the extracts.

Analgesic activity

Analgesic activity of aqueous and alcohol extracts of *Jatropha gossypifolia* Linn. was studied by tail immersion method.

Tail immersion method

The study was conducted by "Tail Immersion Method" . Thirty six healthy Wistar rats weighing around 150-200 gm were distributed in 6 groups each consisting of 6 rats with 1:1 sex ratio. Group I served as control (received normal saline), Group II received the standard drug (received Aspirin 25mg/kg), Group III received dichloromethane extract (DEJG 200mg/kg), Group IV received dichloromethane extract (DEJG 400mg/kg), Group V received methanolic extract (MEJG 200mg/kg) and Group IV received methanolic extract (MEJG 400mg/kg). The tails of the rats up to 5 cm were dipped into hot water maintained at 55±0.5°c. The time taken by the rat to flip its tail clearly out of water was taken as the reaction time. The reaction time in seconds before administration of the drugs was recorded for all the rats from each group. Each extract was studied in two different doses of 200 and 400 mg/Kg b.w. Oral route was followed for the administration of the extract. Reaction time in seconds after the administration of drugs was recorded at an interval of 30 min, 60 min, 90 min and 120 min. Mean was compared with Aspirin.[13,14]

Statistical analysis

All the values are expressed as mean increase in latency after drug administration ± SEM in terms of seconds and analyzed for ANOVA and post hoc Dunnet's t-test. Differences between groups were considered significant at $P < 0.05$ levels and $P<0.01$ as highly significant. The statistical analysis was carried out using Graph pad Instat 3.0 software.

Results and discussion

Phytochemical analysis

The preliminary phytochemical study revealed the presence of alkaloids, carbohydrates, phytosterols, tannins, flavonoids. The results are tabulated in table no.1. It helps to undertake further studies on the isolation and identification of studies carried out with reference to traditional uses of the drug mentioned in *Ayurveda* to justify the claim. In conclusion, this study provides evidences for the analgesic activity of *Jatropha gossypifolia Linn.* which could partly contribute to its ethnomedical use.

Analgesic activity

The results were expressed as a reaction time in sec.(TABLE 2)

Analgesic study shows significant activity in 200 and 400 mg/kg dose of methanolic extract but was found insignificant in 200 and 400 mg/kg dose of dichloromethan extract when compared with standard analgesic drug aspirin. The activity may be due to presence of alkaloid, flavonoid, phytosterols, saponin, tannin and phenolic compound in methanolic extract may be works by blocking chemicals (prostaglandins), that sensitize the peripheral pain receptors to send a pain signal to the central nervous system (CNS).

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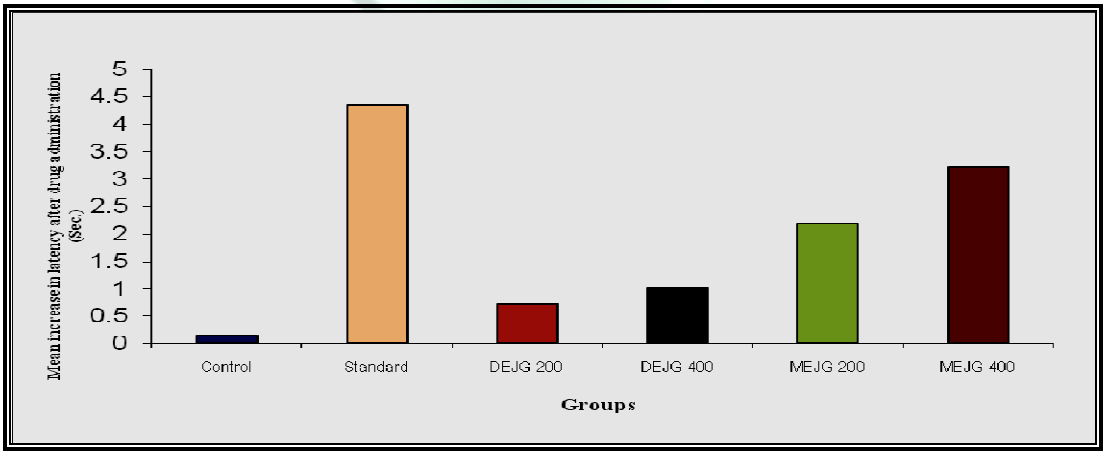

Fig:1 Analgesic activity of Dichloromethane and Methanolic extracts of *Jatropha gossypifolia* leaves in rats by tail immersion method.

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Table:2 Analgesic activity of Dichloromethane and Methanolic extracts of *Jatropha gossypifolia*Linn. leaves in rats by tail immersion method

Group	Dose (mg/kg) b. w.	Reaction time in sec. before drug administration (Mean ± SEM)	Reaction time in sec. (Mean ± SEM)			
			30 min	60 min	90 min	120 min
Control	-	3.12 ± 0.26	3.26 ± 0.18	3.33 ± 0.42	3.28 ± 0.35	3.17 ± 0.24
Standard**	25	3.29 ± 0.35	5.72 ± 0.40	6.54 ± 0.37	7.12 ± 0.22	7.65 ± 0.18
DEJG^{ns}	200	3.41 ± 0.18	3.63 ± 0.33	3.97 ± 0.24	4.16 ± 0.53	4.23 ± 0.67
DEJG^{ns}	400	3.09 ± 0.23	3.27 ± 0.15	3.55 ± 0.33	3.92 ± 0.28	4.11 ± 0.46
MEJG**	200	2.88 ± 0.44	3.04 ± 0.21	4.22 ± 0.27	4.91 ± 0.33	5.06 ± 0.4
MEJG**	400	3.66± 0.36	4.08 ± 0.67	5.78 ± 0.48	6.47 ± 0.29	6.88 ± 0.16

**** Extremely sigmificant (P<0.01), * Significant (p< 0.05), ns- Not significant (P> 0.05)**

Table:1 preliminary phytochemical study of *Jatropha gossypifolia*Linn. leaves

S.No.	Name Of Test	Observation		
		Powder	Dichloromethan extract	Methanolic extract
1.	**Test for Carbohydrate**			
	Molish Test	+ve	+ve	+ve
	Fehling Test	-ve	-ve	-ve
	Benidict Test	+ve	+ve	+ve
	Barfoed Test	+ve	+ve	+ve
2.	**Test for Protien and amino acid**			
	Biuret Test	-ve	-ve	-ve
	Xanthoproetic Test	-ve	-ve	-ve
	Ninhydrin Test	-ve	-ve	-ve
	Test with heavy metals	+ve	+ve	+ve
3.	**Test for fixed oil and fats**			
	Spot test	-ve	-ve	-ve
	Saponification test	-ve	-ve	-ve
4.	**Test for Gum and Mucilage**			
	Swelling test	+ve	-ve	-ve
	Ruthenium red test	+ve	-ve	-ve
5.	**Test for Alkaloid**			
	Dragendroff test	+ve	+ve	-ve
	Mayer's test	+ve	-ve	-ve
	Wagner's test	+ve	+ve	-ve
	Hager' test	+ve	+ve	-ve
6.	**Test for Glycoside**			
	Legal's Test	-ve	-ve	-ve
	Brontrager test	-ve	-ve	-ve
	Killer-killani's test	-ve	-ve	-ve
	Test for cynogenetic glycosides	-ve	-ve	-ve
7.	**Test for Phytosterols**			
	Liebermann's Test	+ve	+ve	+ve
	Salkowaski test	+ve	-ve	+ve
	Liebermann-Burchard's Test	+ve	+ve	-ve
8.	**Test for flavanoid**			
	Ferric Chloride test	-ve	-ve	-ve
	Shinoda test	-ve	-ve	-ve
	Lead acetate test	+ve	+ve	+ve
	Test with acid and alkali	+ve	+ve	+ve
9.	**Test for tannin and phenolic compound**			
	Ferric chloride test	-ve	-ve	-ve
	Copper sulphate test	+ve	+ve	+ve
	Lead acetate test	+ve	+ve	+ve
	Potassium dichromate test	+ve	+ve	+ve
10.	Test for Saponins			
	Foam test	+ve	_ve	_ve
11.	Test for volatile oil	-ve	-ve	-ve