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Research Article

PHYTOCHEMICAL SCREENING AND ANALGESIC ACTIVITY ON COCCINIA GRANDIS

D. Satyavati [1] *, SK. Tajuddin [2], B. Himaja [2]

** [1] Principal & Professor, Department of Pharmacy, Brilliant Grammar School Educational Society, Group of Institutions Integrated Campus (Faculty of Engineering & Faculty of Faculty), Abdullapurmet, Hyderabad, Telangana, INDIA.*

[2] Assistant Professor, Department of Pharmacy, Brilliant Grammar School Educational Society, Group of Institutions Integrated Campus (Faculty of Engineering & Faculty of Faculty), Abdullapurmet, Hyderabad, Telangana, INDIA.

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ABSTRACT

In the present study, the analgesic effect of the aqueous extract of coccinia grandis whole plant was investigated. The hydroalcoholic extracts of coccinia grandis whole plant were ingested orally (p.o.) in two different doses, 200 and 400 mg/kg body weight. The analgesic activity of the aqueous effect of coccinia grandis was evaluated in Whister Albino rat by using Analgesiometer and Eddys hot plate method and compared with the standard, Diclofenac (10 mg/kg body weight). The results showed that coccinia grandis has significant ($p<0.01$) reduction in the paw licking for (400 mg/kg), tail flicking for (200 mg/kg) and diclofenac (10 mg/kg) when compared to control. These results expressing that the extracts would possess analgesic properties. All these effects and the changes in the behavioural activities could be suggested as contributory effects to the use of coccinia grandis whole plant in the management of inflammation and painful conditions.

KEYWORDS: *Coccinia Grandis, Analgesic, Diclofenac, Aqueous extract and Phytochemical Screening.*

INTRODUCTION

Plants are one of the most important sources of medicines. India is known as the "Emporium of Medicinal plants" due to availability of several thousands of medicinal plants in the different bioclimatic zones Analgesic diseases including rheumatoid arthritis are still one of the main health problems of the world's population [1]. Several modern drugs are used to treat these disorders but, their prolonged use may cause severe adverse side effects [2], the most common being gastrointestinal bleeding and peptic ulcers [3]. Consequently, there is a need to develop new Analgesic agents with minimum side effects. The uses of natural remedies for the treatment of inflammatory and painful conditions have a long history, starting with Ayurvedic treatment, and extending to the European and other systems of traditional medicines. Plant drugs are known to play a vital role in management of Analgesic diseases is a moderate size tree with small leaves, which falls earlier on the dry season [4]. Leaves of the plant are used in traditional and tribal medicine of Andhra Pradesh to treat painful inflammatory conditions. A perusal of the literature revealed that although of *coccinia grandis* is widely used in traditional medicine as an anti-inflammatory and analgesic agent [5-8] these properties have not been scientifically evaluated [5]. Therefore, the present study is an attempt to investigate the analgesic properties of the hydroalcoholic extract of *coccinia grandis* whole plant in experimental animals [9, 10].

***Corresponding author:**

Dr. D. Satyavati

Principal & Professor,
Department of Pharmacy,
Brilliant Grammar School Educational Society, Group of Institutions Integrated Campus (Faculty of Engineering & Faculty of Faculty), Abdullapurmet, Hyderabad, Telangana, INDIA.
** E-Mail: satyavati.dhulipala@gmail.com*

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From botanical description of C. grandis, it is a dioecious perennial herbaceous vine. Stems are mostly glabrous, produced annually from a tuberous rootstock, tendrils simple, axillary. Leaves are alternate, simple, blade broadly ovate, 5-lobed, (5-9 or 4-9 cm), acute and mucronate at theapex, cordate with a broad sinus at the base. Surfaces areglabrous or scaly, with 3-8 glands near the base, margins denticulate, petiole (1-5 cm) long. Inflorescence is usually of solitary, axillary flowers. Calyx is of 5 subulate, recurved lobes (2-5 mm) long on the hypanthium. Peduncle is (1-5cm) long. Corolla is campanulate, white, (3-4.5 cm) long, deeply divided into 5 ovate lobes. Stamens are 3, presentas staminodes in female flowers. Ovary is inferior. Fruit is a smooth, bright red, ovoid to ellipsoid berry (2.5-6 cm) long.

Fig. 1: C. Grandis Flowers and Fruits

MATERIALS AND METHODS

Chemicals Required: Carrageenan (1%w/v suspension), Diclofenac (10 mg/kg-standard dose).

Instruments Required: Analgesiometer, Vernier caliper, Heating mantles.

Collection and Authentication of Plant Material:

The plant material i.e coccinia grandis was collected in the month of August 2011 from Wonder Herbals Pvt Ltd, Vanastalipuram, Hyderabad, 500076, Andhra Pradesh. Around 1kg of plant was collected. The plant material was taxonomically identified by Dr. S.K. Mahmood, Department Of Botony, Nizam University-Hyderabad and a specimen was deposited in their Herbarium against issue of Voucher no: 51236.

Preparation of Powder:

The plant material of coccinia grandis were shade dried and then powdered with a grinder to form a coarse powder. The powder was passed through sieve no 40 and was stored in an air tight container until further use. The powder was used for the extraction process.

Preparation of Aqueous extract:

The aqueous extract of the plant was prepared using Maceration process. The coarse powder of plant (100g) was taken inn a beaker with the water quantity of 1000ml and was Macerated for 72hrs. During the Maceration occasional stirring and warming were carried out. After 72 hrs, the suspension was filtered through a fine muslin cloth. The solvent was removed by heating it and a greenish black residue was obtained. (Yield: 9.14%w/w w.r.to dried plant material).

Fig. 2: Dried aqueous extract powder of Coccinia Grandis

Acute Toxic Studies: [11]

The toxic studies have performed upto the range from 100-2000mg/kg of aqueous extract of *Coccinia grandis*. We have observed that there is no sedation, convulsions & no death. But weight loss is observed. So according WHO guidelines $1/5^{th}$ (400mg) &$1/10^{th}$ (200mg) of the extract.

Phytochemical Screening:

1. Test for Glycosides:

A portion of the extract was hydrolysed with HCl and the hydrolysate was subjected to Legal's and Borntrager's test to detect the prescence of different Glycosides.

Legal's Test: To the extract, 1ml of Pyridine and few drops of Sodiumnitropruside were added and it was made alkaline with NaOH. Appearance of pink to red colour shows the prescence of glycosides.

Borntrager's test: Extract was treated with Chloroform and then the chloroform layer was separated. To this equal quantity of dilute Ammonia solution was added. Ammonia layer aquires pink colour showing the prescence of Glycosides.

2. Test for Saponins:

Froth Test: Place 2ml of extract in water in a test tube. Shake well, stable froth (Foam)is formed hence saponins.

3. Test for Alkaloids:

Dragendorff's Reagent: Drug extract when treated with Potassium Bismuth Iodide Solution gives reddish brown ppt. Hence Alkaloids are present.

Mayer's Reagent: Drug extract when treated with Potassium Mercuric Iodide solution gives cream color ppt. Hence alkaloids are Present.

4. Test for Tannins:

To the extract few ml of Chromic acid was added. No ppt was found. Hence Tannins are present.

5. Test for Flavonoids:

Shinoda Test: To the extract add few Magnesium turnings and con HCl dropwise . Pink scarlet, crimson red or occasionally green to blue colour appears after few mins. Hence Flavonoids are present.

Zinc Hydrochloride test: To the extract add a mixture of Zn dust and con HCl. Gives red colour after few min. Hence Flavonoids are present.

6. Test for Mucilage:

To the extract Ruthenium red solution is added, pink colour is obtained. Hence mucilage is present.

7. Test for Carbohydrates:

Molisch Test: To the extract add few drops of alcoholic α-naphthol, then add few drops of con. H_2SO_4 through sides of the test tube, violet colour ring is appeared at the junction. Hence Carbohydrates are present.

8. Test for Proteins:

Xanthoproteic test: To 5ml of extract add 1ml of con Nitric acid and boil. Yellow ppt was obtained. After cooling add 40% NaOH solution. Orange colour was obtained. Hence proteins are present.

9. Test for Phytosterols:

Salkowski Test: To the extract add few drops of con H_2SO_4, re colour at the lower layer indicate the prescence of sterol and yellow colour prescence indicate. Hence Phytosterols are present.

Analgesic activity:

36 Albino rats (Whister Strain) were taken and divided into 6 groups i.e.6 in each group (Head, Body, Tail, Head-Body, Body-Tail, Head-Tail). Every rat in each group was weighed and their weights were in the range of 150-200mg and as per the weight the standard dose of Diclofenac (10mg/kg) and coccinia grandis for each Rat was calculated. Both Diclofenac and coccinia grandis were given orally. Analgesic activity was studied by using Eddy's Hot Plate. The time for paw licking was noted in different groups as given below. The temperature in Eddy's Hot Plate was maintained at 60^oC (Note: This experiment was conducted after a recovery period of 1 week).

Table No. 1: Analgesic activity of different groups with drugs dose

GROUP	DRUG GIVEN
GROUP I	SALINE (Control)
GROUP II	DICLOFENAC (Standard-10mg/kg)
GROUP III	LOW DOSE OF COCCINIAGRANDIS (200mg/kg)
GROUP IV	HIGH DOSE OF COCCINIAGRANDIS(400mg/kg)
GROUP V	LOW DOSE +DICLOFENAC
GROUP VI	HIGH DOSE+DICLOFENAC

The analgesic activity with time was tabulated

RESULTS AND DISCUSSIONS

Phytochemical Screening:

Table No. 2: Data showing phytochemical screening of aqueous extract of *coccinia grandis*

Carbohydrates	Glycosides	Alkaloids	Proteins	Phytosterols	Flavonoids	Tannins	Saponins	Mucilage
+	+	+	+	+	+	+	+	+

Note: + (Present); - (Absent)

Table No. 3: Data showing % inhibition of Paw Licking in each group

DRUG	0H	1H	2H	3H	4H
GROUP 1	3.74	3.36	3.2	3.28	3.36
GROUP 2	4.24	6.10	8.18	8.16	9
GROUP 3	5.1	6.18	6.9	8.6	8.77
GROUP 4	5.1	5.02	6.2	7.03	8.06
GROUP 5	5.1	7.38	7.29	7.9	7.8
GROUP 6	3.7	5.8	6.64	7.84	7.9

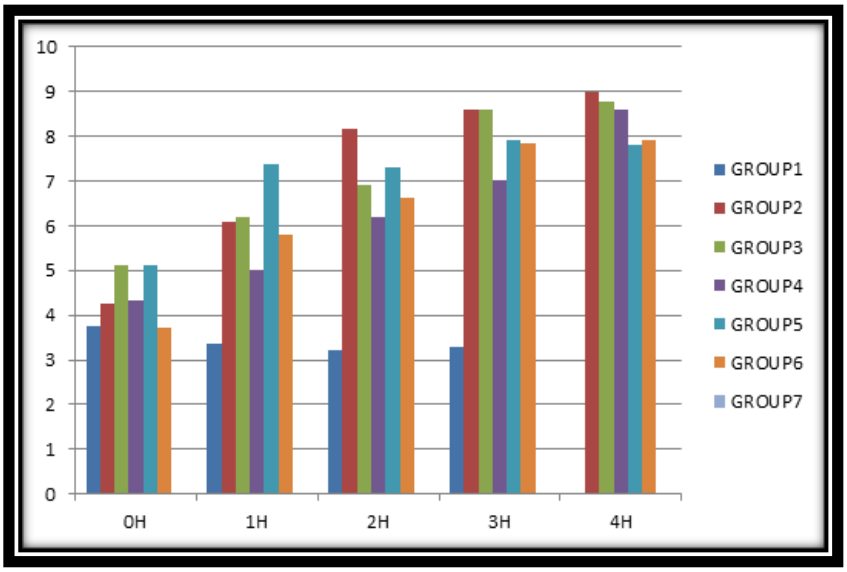

Fig. 3: Showing % inhibition of Paw Licking in each group

The above graph illustrates that there is a delay in paw licking in Groups V and VI when compared to other groups which also infer that Coccinia grandis itself also acts as analgesic agent.

Table No. 4: Data showing % inhibition of Tail Flicking in each group

DRUG	0H	1H	2H	3H	4H
GROUP 1	1.25	1.46	1.53	1.258	2.326
GROUP 2	1.094	1.186	1.52	1.094	1.924
GROUP 3	0.835	0.92	1.146	1.26	1.33
GROUP 4	0.98	0.148	1.20	1.21	2.964
GROUP 5	1.020	1.594	2.062	2.356	2.78
GROUP 6	0.842	1.33	1.428	1.478	2.202

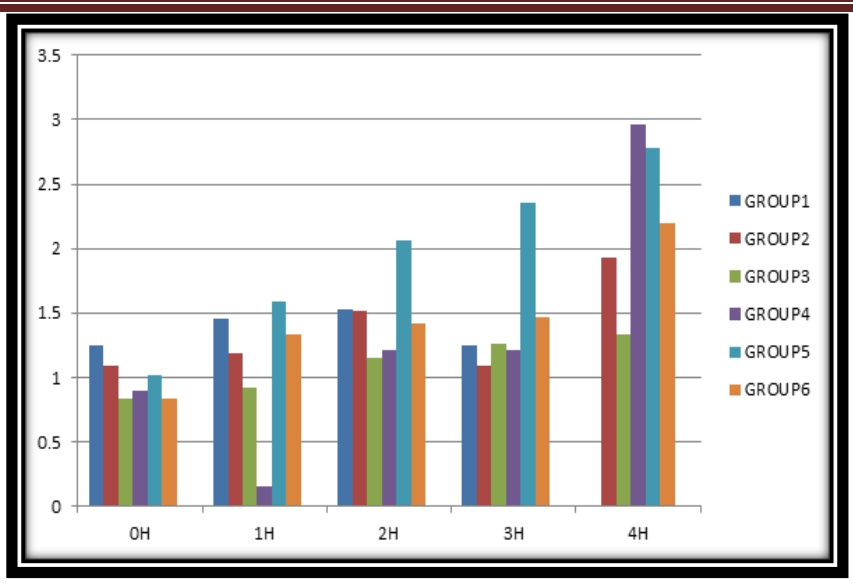

Fig. 4: Showing % inhibition of Tail Flicking in each group

The above graph illustrates that % inhibition of Tail Flicking was more in groups V and VI, when compared with the other groups. It also infer that Coccinia grandis itself acts as Analgesic agent but not as good as the standard drug (Diclofenac). Coccinia grandis shows synergistic effect when given in combination with standard drug.

CONCLUSION

The phytochemical screening of aqueous extract of *coccinia grandis* showed the presence of Saponins which were considered to be responsible for its pharmacological activity (Analgesic activities). Therefore *coccinia grandis* was considered to possess analgesic activities.

The literature clearly suggests that *coccinia grandis* has been widely used as Analgesic. In order to evaluate its analgesic activities, in-vivo studies of aqueous extract of *coccinia grandis* were conducted on rats.

The investigations on *coccinia grandis* were found to produce positive results towards the evidence of Analgesic activities. The data obtained from Analgesic activities experiments clearly suggested that the analgesic activities of *coccinia grandis* were dose dependent. It also can be noted that the combination of *coccinia grandis* and Diclofenac had a synergistic effect in curing algesia. Finally our studies concluded that *coccinia grandis* has Analgesic activities.

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