

Original Research Article

Screening of *Euphorbia hirta* Leaves for Antipsoriatic Activity in MiceHarsha Vardhan K¹, Jaikumar S^{2*}, Premlal KR³¹Professor, Department of Dermatology, Sri Lakshmi Narayana Institute of Medical Sciences, Pondicherry²Assistant Professor, Department of Pharmacology, Sri Lakshmi Narayana Institute of Medical Sciences, Pondicherry.³Reader, Division of Oral Pathology & Microbiology, Indira Gandhi Institute of Dental Science, Sri Balaji Vidyapeeth University, Pondicherry***Corresponding author**

Dr. S. Jaikumar

Email: jaipharma2007@gmail.com

Abstract: Psoriasis is a skin disease that causes scaling and inflammation where the skin cells grow deep in the skin and slowly rise to the surface. Worldwide nearly 1 to 3% total population affected with psoriasis. This inflammatory disease is usually involves extensor surfaces and scalp and is now recognized to be mediated in part by immune alterations. Herbal remedies have been used effectively in skin disorders for thousands of years worldwide. *Euphorbia hirta* belongs to the plant family *Euphorbiaceae* and genus *Euphorbia*, widely used in various skin disorders. In order to validate its traditional claim, an attempt was made to evaluate the anti-psoriatic activity of 250 mg/kg ethanolic leaf extract of *Euphorbia hirta* in mouse tail test method. Isoretinoic acid was used as reference drug. Anti-psoriatic activity was assessed by measuring the Degree of Orthokeratosis and Relative Epidermal Thickness. Result shows that, *Euphorbia hirta* produced significant increase in Orthokeratosis and the Relative epidermal thickness compared to control. Present study concludes that, *Euphorbia hirta* exhibited anti-psoriatic activity and it may be considered for psoriasis and other skin disorders.

Keywords: Psoriasis, *Euphorbia hirta*, Orthokeratosis and Isoretinoic acid

INTRODUCTION

Psoriasis is a chronic systemic inflammatory disease affecting about 1% to 3% of the population worldwide. Although psoriasis may occur at any age, the peak periods of onset are 16 to 22 and 50 to 60 years of age, with majority of patients developing the disease before age 40. Psoriasis primarily affects the elbows, knees, scalp, genitals, and trunk but can involve any body location. In addition to cutaneous psoriasis is associated with significant physical and behavioral comorbidities. Moreover, the visibility of the skin lesions creates a strong psychological burden involving relationships, work, social activities and overall wellbeing [1].

Herbal therapy for skin disorders has been used for thousands of years. Even our biologically close relatives, the great apes, use herbal self-medication [2]. Specific herbs and their uses developed regionally, based on locally available plants and through trade in ethnobotanical remedies. *Euphorbia hirta* belongs to the plant family *Euphorbiaceae* and genus *Euphorbia*. It

is a slender- stemmed, annual hairy plant with many branches from the base to top, spreading upto 40 cm in height, reddish or purplish in color. Leaves are opposite, elliptic - oblong to oblong- lanceolate, acute or subacute, dark green above; pale beneath, 1- 2.5 cm long, blotched with purple in the middle, and toothed at the edge. The fruits are yellow, three- celled, hairy, keeled capsules, 1-2 mm in diameter, containing three brown, four-sided, angular, wrinkled seeds [3].

Euphorbia hirta is used in the treatment of gastrointestinal disorders (diarrhea, dysentery, intestinal parasitosis, etc.), bronchial and respiratory diseases (asthma, bronchitis, hay fever, etc.), and in conjunctivitis. The aqueous extract exhibits anxiolytic, analgesic, antipyretic, and anti-inflammatory activities. The stem sap is used in the treatment of eyelid styes and a leaf poultice is used on swelling and boils. Extracts of *Euphorbia hirta* have been found to show anticancer activity. The aqueous extract of the herb strongly reduced the release of prostaglandins I₂, E₂, and, D₂. Methanolic leaf extract of *Euphorbia hirta* have

antifungal and antibacterial activities. The leaves pounded with turmeric and coconut oil are warmed and rubbed on itchy soles [4]. Decoction of dry herbs is used for skin diseases. Decoction of fresh herbs is used as gargle for the treatment of thrush. Root decoction is also beneficial for nursing mothers deficient in milk. Roots are also used for snake bites [5]. The polyphenolic extract of *Euphorbia hirta* has antiameobic and antispasmodic activity [6]. Quercitrin, a flavanoid glycoside, isolated from the herb showed an antidiarrheal activity [7]. It is reported to have a relaxation effect on respiration [8]. The alcoholic extract of whole plant shows hypoglycemic activity in rats [9]. Current study was conducted to evaluate the antipsoriatic activity of *Euphorbia hirta* leaf extract in animal model.

MATERIALS AND METHODS

Plant Material

The *Euphorbia hirta* was collected from the out skirts of Pondicherry and it was identified, authenticated as *Euphorbia hirta* by Prof. Dr. P. Jayaraman, Director, Plant Anatomy Research Centre, Chennai. The voucher specimen was deposited in the herbarium for future reference.

Preparation of Extract

The collected *Euphorbia hirta* leaves were washed, shade dried and then ground into coarse powder. The powder was then subjected to exhaustive extraction by a maceration process using 90% methanol as a solvent at room temperature for 7 days. The methanolic extract was concentrated by vacuum distillation to dry. The collected extract was stored in desiccators and used for further pharmacological study.

Animals

Swiss albino mice of either sex, weighing between 20 - 25 gm were used for this study. The animals were obtained from animal house, of Sri Lakshmi Narayana Institute of Medical Sciences, Pondicherry. The animals were placed at random and allocated to treatment groups stainless steel cages. Animals were housed at a temperature of $24 \pm 2^\circ\text{C}$ and relative humidity of 30 - 70 %. A 12:12 light: day cycle was followed. All animals were allowed to free access to water and fed with standard commercial pelleted rat chaw (M/s. Hindustan Lever Ltd, Mumbai). All the experimental procedures and protocols used in this study were reviewed by the Institutional Animal Ethics Committee.

Antipsoriatic Activity – Mouse Tail Test [10]

Euphorbia hirta leaf extract was evaluated for antipsoriatic activity by the mouse tail test for psoriasis. Eighteen animals were divided into 3 groups of six

each. Group I served as normal control (0.1% CMC), group II served as reference control (Isoretinoic acid, 0.5 mg/kg) and group III was treated with the methanolic leaf extract of *Euphorbia hirta* at 250 mg/kg body weight. Test drugs were administered once daily for 14 days, by suspending in 0.1 % CMC solution. At the end of the 14th day treatment, mice were sacrificed by phenobarbitone anesthesia and the proximal parts of their tails were cut and each group tails stored in separate containers containing 10 % formalin in saline.

Histopathological Examination

Longitudinal histological sections were prepared from the tail skin and stained with hematoxylin eosin.

The specimens were histometrically analyzed for:

(I) The horizontal length of an individual scale lying in between adjacent hair follicles including sebaceous glands (n = 10 scales per animal, n = 6 animals per treatment group; i.e. a total of 60 measurements per treatment),

(II) The horizontal length of the fully developed granular layer within an individual scale (n = 10 scales per animal, n = 6 animals per treatment group; i.e. a total of 60 measurements per treatment), and

(III) The vertical epidermal thickness between the dermo-epidermal junction and the lowest part of the stratum corneum (n = 5 measurements per scale, n = 10 scales per animal, n = 6 animals per treatment group; i.e. a total of 300 measurements per treatment).

From these raw data (I to IV) the following parameters were calculated according to the method Bosman et al., 1992 [11].

(IV) The degree of orthokeratosis of an individual scale defined as the percentage ratio of (2) divided by (1) (n = 60 data per treatment condition),

(V) The control related 'drug activity' upon epidermal differentiation,

$$\text{Drug activity} = \frac{\text{OKs} - \text{OKc}}{100 - \text{OKc}} \times 100$$

with OK (i.e. orthokeratosis) as the mean of the parameter explained under (IV) for a test substance (s) and the untreated control condition (c), respectively, and

(IV) the relative epidermal thickness of individual scales as the percentage ratio of the measure under (III),

for a given treatment in relation to the mean of untreated controls set to 100% (n = 300 data per treatment condition).

The three overall parameters namely, the degree of orthokeratosis, drug activity and relative epidermal thickness were eventually used for the evaluation of drug effects.

Statistical Analysis

Data obtained was presented as Mean \pm Standard Error Mean. In the mouse tail test for statistical comparisons, explorative probabilities were obtained by the Mann–Whitney U test. Statistical calculations were performed using GraphPad prism software. Values with $P < 0.05$ are considered significant.

RESULTS

Table-I: Effect of methanolic leaf extract of *Euphorbia hirta* on the degree of orthokeratosis and relative epidermal thickness in the mouse tail test

Drug Treatment	Degree of Orthokeratosis (%)	Drug Activity (%)	Relative Epidermal Thickness (%)
Vehicle Control (0.1% CMC)	17.33 \pm 1.12	-	100
Reference Control (Isoretinoic Acid, 0.5 mg/kg)	65.52 \pm 2.43***	56.97	119.34
<i>Euphorbia hirta</i> (250mg/kg)	57.44 \pm 2.71***	51.73	107.72

Values are expressed as mean \pm S.E.M.

* $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$ Vs Control

Anti-psoriatic activity of the methanolic leaf extract of *Euphorbia hirta* was evaluated in the mouse tail test method, the % Degree of Orthokeratosis and the % Relative Epidermal Thickness were observed and the result were given in table I. Keratotic condition was seen in the adult mouse tail which is one of the main features of psoriasis. Induction of orthokeratosis in the adult mouse tail is the basis behind the mouse tail test. Isoretinoic acid was used as reference drug in the study. Isoretinoic acid and the test drug *Euphorbia hirta* significantly ($P > 0.001$) enhanced the degree of Orthokeratosis and Relative Epidermal Thickness in the mouse tail method. The % drug activity of the reference control Isoretinoic acid and *Euphorbia hirta* were 56.97% and 51.73% respectively.

CONCLUSION

Psoriasis is a chronic, cutaneous-articular disease affecting 2-3% of the worldwide population. This inflammatory disease is usually involves extensor surfaces and scalp and is now recognized to be mediated in part by immune alterations. *Euphorbia hirta* traditionally used in various skin infections and disorders. From the results of present study, it was concluded that methanolic leaf extract of *Euphorbia hirta* exhibited antipsoriatic activity in mouse tail test method.

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