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BIOPROSPECTING OF *EUPHORBIA NIVULIA* Buch.

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ABSTRACT

Nature always stands as a golden mark to exemplify the outstanding phenomenon of symbiosis. The history of herbal medicines is as old as human civilization. Many herbal remedies have been employed in various medicinal systems for the treatment and management of different ailments. *Euphorbia nivulia* Buch. – Ham. a member of *Euphorbiaceae* family is a wild, thorny, xerophytic, succulent plant, commonly used in fencing of the agricultural field and also in dry barren areas. It has different biological activities for the treatment of several ailments of human being. It possesses antimicrobial, wound healing, haemostatic, larvicidal, insecticidal, nematicidal and cytotoxic activity. Chemically, it contains terpenes, glycoproteins, phytoelements and phytochemicals. This article provides informative data on ethnobotanical importance and bioprospecting of *Euphorbia nivulia* Buch. - Ham.

Keywords: *Euphorbia nivulia*, Biological activities, Traditional uses.

INTRODUCTION

Vegetation is one of the precious gifts of nature where plants are intimately related to human being. People have been utilizing plants as medicine since the earliest period of civilization. The use of plants for curing human diseases is an ancient practice, in which interest has been revived at modern age [1]. People of remote villages and tribal areas are dependent upon the practice of folk medicines [2]. In recent times, focus on plant research has increased all over the world and a large body of evidence has been collected to show immense potential of medicinal plants used in various traditional systems [3]. Ethnobotanically plant latex has a great potential with respect to its medicinal value. Latex has been reported to occur in 12000 plant species belonging to 900 genera. A common feature that can be found in the latex of the *Euphorbiaceae* is the presence of noticeable digestive enzyme activity. *Euphorbia* is a large genus consisting of about over 2000 species in the world. Approximately 195 species of of *Euphorbia* have been recorded from India (Aditya, 2010). The genus includes herbs, shrubs and trees in widely diverse habitats [4]. One such plant, *Euphorbia nivulia* Buch.-Ham. invites attention of the researchers worldwide for its biological activities. There is not much literature available on biological activities of *Euphorbia nivulia* Buch.-Ham. Also, the earlier reviews on euphorbian plants lack satisfactory information regarding its biological activities. The aim of the present review is to provide the updated information on biological uses of *Euphorbia nivulia*. Emphasis is being laid on the areas of the most recent interest and those which have not been presented in earlier reports.

Botanical Description

Kingdom: Plantae, Division: Magnoliophyta, Class: Magnoliopsida, Order: Malpighiales, Family: Euphorbiaceae, Genus: *Euphorbia*, Species: *Euphorbia nivulia* Buch. - Ham.

Synonyms

Sanskrit: Patrasnuhi; Patta-karie. Hindi: Sehund. Bengali: Sij. Marathi: Sabar; Sabarkanda; Nivdung. Telgu: Akujimudu. Tamil: Akujemudu.

Habitat

Northern and central India, Planted as hedge plant, also wild in arid soils and often planted in dry areas.

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Part used

Juice of leaves, root bark, stem and latex.

Species of Euphorbia

Its species are widely distributed in tropical Asia, Africa, Europe and Australia. Other reported species of genus Euphorbia are: *Euphorbia acaulia* Rox., *Euphorbia antiquorum* Linn., *Euphorbia clarkeana* Hook f., *Euphorbia cristata* Heyne ex Roth., *Euphorbia cyathophora* Murr., *Euphorbia dracunculoides* Lamk., *Euphorbia fusiformis* Buch.-Ham., *Euphorbia hirta* Linn., *Euphorbia hypericifolia* auct. non Linn., *Euphorbia indica* Lamk., *Euphorbia milii* Desmoul, *Euphorbia neriifolia* auct. non Linn., *Euphorbia nivulia* Buch.-Ham., *Euphorbia notoptera* Boiss., *Euphorbia panchaganiensis* Blatt and McCann., *Euphorbia pilosa* Linn., *Euphorbia prostrata* Ait., *Euphorbia prunifolia* Jacq., *Euphorbia pulcherrima* Willd. ex Klotz., *Euphorbia pycnostegia* Boiss., *Euphorbia resinifera* Berg., *Euphorbia royleana* Boiss., *Euphorbia thomsoniana* Boiss., *Euphorbia thymifolia* Linn. and *Euphorbia tirucalli* Linn. [5,6].

Morphological Characters

Tall shrubs or small trees with cylindrical stem and branches. Stipular spines glabrous, straight, paired, often blackish. Leaves appear only during rainy season, 8.5 – 20 x 3.5 – 6.5 cm, crowded at the end of branches, obovate-oblong or spathulate, glabrous. Cymes – 3- flowered, born from above the leaf scars on the tubercles. Capsules glabrous, trigonous, seeds globose, dorsally lined, smooth. Flowering and fruiting period is March to July [5,6].

Uses of plant

All parts of the plant possess medicinal properties. The juice of the leaf is used as a purgative, diuretic etc. The paste of the leaf, made with neem oil is applied externally in rheumatism. Plant latex is used for treating jaundice, dropsy, enlargement of liver and spleen, and applied to hemorrhoids. Coagulated latex is used for bronchitis [7]. It is warmed in mustard oil; resulting mixture is applied in cold and headache. Root bark is boiled in rice-water and arrack is given in dropsy. In north eastern region of India, the leaf juice is utilized in pains and boils by Boro community of Assam [8]. The latex of this plant possesses vesicant, wormicidal and purgative properties [9]. Milky juice i.e. latex of this plant is reported for its bronchodilating activity [10]. Fleshy part of stem is roasted in extremely hot ash for 20-30 minute; juice is extracted, 1-2 tablespoonful juice (for 3-7year aged children) per day is recommended dose for curing cough with in 7 -10 days (Mahajan and Badgujar, 2008). Leafy latex and root is applied in skin disorders, ear disorders, retention of urine, swelling and worm infection [11]. Kumar and Chaturvedi (2010) [12], reports the application of ethnomedicine derived from stem of this plant in curing the bone fractures and antiseptic utility of latex. Although there is a wide range of potential useful medicinal phytoconstituents of the plant, the research in this area is infantile. Some of experimentally proved biological activities of *Euphorbia nivulia* are given below.

Antimicrobial

Aqueous alcohol extracts of the leaves of *Euphorbia nivulia* is reported for antimicrobial activity. Nineteen different bacterial strains and two fungal cultures were used for antimicrobial activity. The results indicate that the aqueous alcohol extract is active at concentrations ranging from 3-12.5 µg/ml for Gram-negative and 12.5-200 µg/ml for Gram-positive bacteria, in comparison with known antibiotics such as streptomycin and nystatin. The latex has several triterpenes, which exhibit significant antimicrobial activity against *Staphylococcus aureus* and *Escherichia coli*. This information is given by Khare (2007) [5] in his book, "Indian Medicinal Plants". The remarkable effect on bacterial growth inhibition with increasing concentration of Nivulian, a cysteine protease of *Euphorbia nivulia latex* is reported by us. It shows comparable inhibitory activity against both Gram positive and Gram negative bacteria tested. Nivulian had a strong antibacterial activity against *Escherichia coli* and *Staphylococcus aureus,* moderate against *Pseudomonas aeruginosa* and lesser activity against *Klebsiella pneumoniae*, *Proteus vulgaris* and *Bacillus subtilis* [13]. The observations made by Nagarathnam et al., 2010 [14] regarding antimicrobial activity of cysteine protease of plant origin present in *Curcuma longa* is also similar to Nivulian. Another member of the Euphorbiaceae i.e. *Euphorbia prostrata* is also reported for antibacterial activity [15]. Therefore we believe that, plant latex may find a position in preparing antimicrobial compounds with a great potential in abatement of infectious diseases in the near future.

Wound healing

Recently, Badgujar et al., 2009 [16] reported that the centrifugal fraction of latex of *Euphorbia nivulia* enhanced the rate of wound contraction and period of epithelization in mice. The surface area of latex treated wound was reduced by 93 to 98 per cent on the 18^{th} day as compared to control (90 per cent). It is interesting to note that, similar type of wound healing activity is present in other members of Euphorbiaceae family viz., *Euphorbia hirta* and *Euphorbia nerifolia*. Ethanolic extract of whole plant of *Euphorbia hirta* is reported for burn wound healing purpose [17]. Plant latex of *Jatropha curcas* is ethnobotanically reported for curing the wound. Active ingredient i.e. curcain, isolated from the latex of *Jatropha curcas* has profound effect on wound healing phenomenon, as it is evident from the report of Nath and Dutta (1992) [18]. Aqueous extract of latex of *Euphorbia nerifolia* is

evaluated for wound healing effect in guinea pig. This extract facilitated the healing process as evident in increase in tensile strength, DNA content, epithelization and angiogenesis [19]. Thus, the plant latex contains not only cysteine protease but also other active ingredient responsible for wound healing activity.

Haemostatic

A significant reduction in bleeding/clotting time in mice is reported by the treatment of Nivulian [20]. Also, it significantly decreased the coagulation time of whole blood in mice. The Nivulian arrested bleeding from fresh wounds by reducing bleeding/clotting and whole blood coagulation time which are important indices of haemostatic activity. The reduction in coagulation time of whole blood by the Nivulian indicates that it may also enhance the blood coagulation pathways. Thus, this shrub could be a promising haemostatic agent [13]. Haemostatic function of plant product is also reported from the latex of Euphorbiaceae member i.e. *Jatropha gossypifolia* [21]. Traditional report of hemostatic activity of *Jatropha curcas* is experimentally proved by Osoniyi and Onajobi (2003) [22]. Plant latex remarkably reduces the whole blood clotting and bleeding time. However, active ingredient of plant latex of *Euphorbia nivulia* is yet unknown and has to be worked out.

Larvicidal

The larvicidal activity of centrifugal fraction of latex against fourth instar stage of *Anopheles* sp is very recently reported by us [13]. Mortality rate of larvae was high at higher concentration of proteinous fraction of latex. But lower concentration caused several morphological changes of larvae. The Nivulian (centrifugal fraction of latex) had a moderate larvicidal activity as it was 2.5 times less potent than pyrethrin, when fourth instar larvae of *Anopheles* sp. are exposed to both for a period of 24 hrs. The larvicidal activity of ethyl acetate, butanol, and petroleum ether extracts of five species of Euphorbiaceae plants, viz., *Jatropha curcas*, *Pedilanthus tithymaloides*, *Phyllanthus amarus*, *Euphorbia hirta*, and *Euphorbia tirucalli*, is evaluated against the early fourth instar larvae of *Aedes aegypti* L. and *Culex quinquefasciatus* (Say). The larval mortality is observed after 24 h of exposure. The LC_{50} value of petroleum ether extracts of *Jatropha curcas*, *Pedilanthus tithymaloides*, *Phyllanthus amarus*, *Euphorbia hirta*, and *Euphorbia tirucalli* are 8.79, 55.26, 90.92, 272.36, and 4.25 ppm, respectively, against *Aedes aegypti* and 11.34, 76.61, 113.40, 424.94, and 5.52 ppm, respectively, against *Culex quinquefasciatus.* Of the various ratios tested, the petroleum ether extracts of *Jatropha curcas* and *Euphorbia tirucalli* are more efficient than the other plant extracts [23]. It is, therefore, suggested that plant latex may have an ideal potential larvicidal property against *Aedes aegypti* and *Culex quinquefasciatus*. This may be an ideal ecofriendly approach for the control of the dengue vector, *Aedes aegypti*, and the lymphatic filariasis vector, *Culex quinquefasciatus.*

Insecticidal

The *Euphorbia nivulia* aqueous leaf extract exhibited toxic and insect growth regulatory (IGR) effects on cabbage diamondback moth (DBM), *Plutella xylostella* L. At 96 h after treatment, mortality is observed only in 10 and 20 per cent concentration with a maximum of 10 per cent mortality. At adult emergence, the highest mortality was observed at 2.5 per cent concentration (57.7 % mortality) followed by 10, 20 and 5 per cent concentrations of extract. The mortality is observed to increase from 96 h after treatment till adult emergence in all the concentrations. A change in mortality pattern with variable time is observed such that the initial mortality is largely due to toxic effect while at the time of adult emergence the IGR effect was more compared to toxic effects [24].

Nematicidal

The nematicidal effect of Nivulian showed that the reproduction of root knot nematode, *Meloidogyne incognita* in plants grown in soil (treated with Nivulian) is significantly suppressed. However, nematode population declined only in pots treated with Nivulian and standard nematicide whereas, in untreated – inoculated pots it had almost doubled. It is, therefore, reasonable to believe that plants grown with Nivulian develop certain degree of resistance against nematode attack. It may be due to the latex containing phenolic substances or due to the absorption of substances liberated during decomposition of Nivulian by soil microbial flora [25]. This type of nematicidal property is already reported in some latex bearing plants namely, *Carica papaya*, *Artocarpus heteropyllus*, *Ficus carica*, *Ficus elastica*, *Ficus glomerata*, *Ipomoea fistulosa*, *Nerium odorum* and *Tabernaemontana coronaria* by Siddiqui et al., 1987. Incorporation of chopped shoots of these laticiferous plants significantly suppressed the population of root knot nematode i.e. *Meloidogyne incognita*. Very interestingly, *Euphorbia prostrata* possesses antiparasitic activity against *Meloidogyne incognita.* It is mentioned for its nematicidal potency [15]. Therefore, we believe that, the plant latex may occupy the place of bio-pesticide for controlling the population of root knot nematodes.

Cytotoxic activity

The latex of *Euphorbia nivulia* afford ingol diterpenes, 3-acetyl-8-methoxyl-7-angolyl-12-hydroxylingol (6), 3, 12-diacetyl-7-hydroxy-8-methoxylingol (7), and 3,12-diacetyl-7-angolyl-8-hydroxylingol (8) as shown in figure 1 [26] along with three macrocyclic ingol diterpenes derivatives: 3, 7, 12-triacetyl-8-benzoylingol (1), 3, 12-diacetyl-7-angeloyl-8-methoxyingol (2), and 7-angeloyl-12-acetyl-8-methoxyingol (3) and two ingol diterpenes derivatives 3, 12-diacetyl-8-benzoylingol (4) and 3, 12-

diacetyl-7-benzoyl-8-nicotinylingol (5) [27]. The structures of above natural products are deduced on the basis of spectroscopic analysis (Figure 1). Mostly all diterpenes exhibit cytotoxic activity.

These authors also reported that, the ingol derivatives (**1 – 5**) of *Euphorbia nivulia* latex are reported for prostaglandin E2 (PGE2) inhibitory activity, only isolate **3** ingol derivative showed significant inhibition. These diterpenes are known for their diverse biological activities [28-30]. Ingol derivatives of compounds **1 – 8** are notably reported for their potential anticancer activity [26]. Of these, compounds **2**, **3**, and **7** showed significant cytotoxic activity against Colo 205, MT2, and CEM cell lines and others showed moderate or no activity. Toxicity potential of compounds **2**, **3**, and **7** is almost same in the three cell lines [26].

Chemical constituents

Terpenes

The new tetracyclic triterpenes: Cycloart-25-en-2β-ol and cyclonivulinol and three ingol diterpenes 3,12-*O*-diacetyl-7-*O*-angeloyl-8-methoxyingol**;** 3,7,12-*O*-triacetyl-8-*O*-benzoylingol and 7-*O*-angeloyl-8-methoxy-12-*O*-acetylingol [26] identified from the latex of *Euphorbia nivulia.*

Glycoproteins

High molecular weight lectin (44 kDa) was purified by affinity chromatography from the latex of *Euphorbia nivulia*. It is a type of glycoprotein containing 9% of carbohydrate and appears to be a dimer with subunits of M_r of 22 000 Da. Its sedimentation coefficient is 4.8S [31]. Recently, we isolated two glycoproteins from the latex of this plant viz., Nivulia-II and Nivulian-III (43.42 and 52.96 kDa). Nivulian-II has notable proteolytic activity and it belongs to cysteine protease category [32].

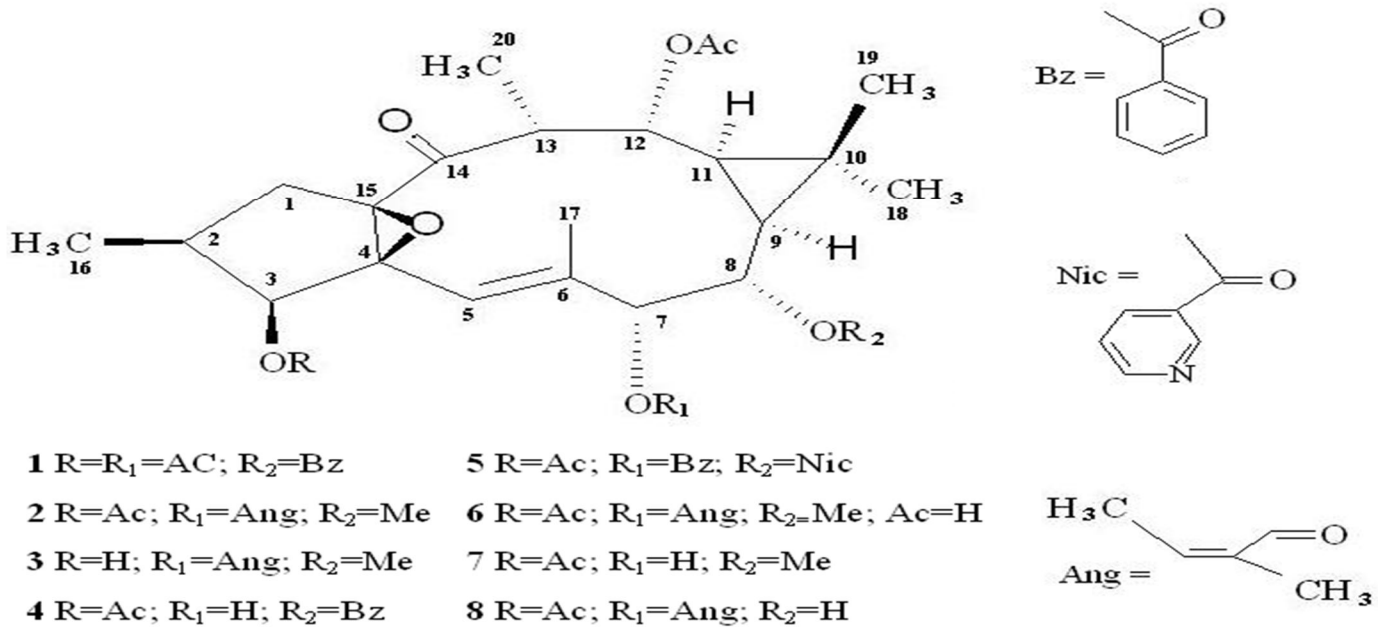
Phytoelements

Phytoelements like Fe (1.484), Cu (0.072), Zn (0.384), Mn (0.173), Mg (0.204), Na (2.085) and Ca (1.031) are detected in ppm quantities from the ash sample of plant latex by using atomic absorption spectroscopy [13]. All these minerals are also found in latex bearing unripe fruit pulp of *Carica papaya* [33] and *Ficus carica* [32]. Only difference is the per cent occurrence of these minerals with great variation in its content.

Phytochemicals

The latex of *Euphorbia nivulia* contains phenolic compounds, alkaloids, cynogenic glycosides, terpenes and tannins [34]. Miscellaneously, it also contains caoutchouc-like substance, fat, albuminoids, hydrolytic enzymes, sugar, pectin, citric, tartaric and mallic acid, dextrin, euphol, nerifloiol etc [13]. Similar phytochemicals are reported in the plant latex samples of *Carica papaya* and *Hevea brasiliensis* [26].

Figure 1: Ingol derivatives of *Euphorbia nivulia* latex



1 R=R_1=AC; R_2=Bz
2 R=Ac; R_1=Ang; R_2=Me
3 R=H; R_1=Ang; R_2=Me
4 R=Ac; R_1=H; R_2=Bz
5 R=Ac; R_1=Bz; R_2=Nic
6 R=Ac; R_1=Ang; R_2=Me; Ac=H
7 R=Ac; R_1=H; R_2=Me
8 R=Ac; R_1=Ang; R_2=H

DISCUSION AND CONCLUSION

In present global scenario, natural medicines are gaining prominence, because they are economical, easily available and relatively free from side effects. The increased global demand for polyherbal formulations is reflective of positive impact of consolidated efforts aimed at reviving science of phytopharmacy. *Euphorbia nivulia* possesses a wide spectrum of biological activities applicable in both medicine as well as agricultural practices. This plant has potential for various health related activities such as antimicrobial, antimalerial, cytotoxic, wound healing, haemostatic and many more. As this plant exhibit soil-pest controlling property and also apply in post harvest management process. The benefits of the vigorous nature of *Euphorbia nivulia* and its wide geographical distribution offer an opportunity to agro and pharma industries.

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