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A REVIEW ON WOUND HEALING ACTIVITY OF GHAMRA WITH THEIR PHYTOCHEMICAL SCREENING, COMPOSITIONS AND PHARMACOLOGICAL ACTIVITY

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ABSTRACT: The wound healing method is thought of as mutually beneficial cellular and organic chemistry stages that classify measures to enhance the wound. Wound healing is outlined as stages that are finished by the body and delayed wound healing will increase the likelihood of microorganism infection. The improved wound healing method is performed by shortening the time required for healing or lowering the inappropriate happens. World Health Organization (WHO) and India have been promoting the use of traditional medicine because they are less expensive, easily available, strong belief among the community in developing countries, etc. Literature reveals that simple traditional plants are beneficial in treating several skin-related problems and for wound healing. *Tridax procumbens* Linn (Asteraceae) growing courtallam widely and surrounding regions peoples used who were suffered from minor cuts, scrapes. This plant easily available in any region of the world. Tridax daisy plant grows at all types of land escapes. In rainy seasons, it grows rapidly. This is an evergreen plant widely used for wound healing, which is helpful for researchers to develop new Wound healing formulations for human use.

INTRODUCTION: The current trend is leading to the development of innovative wound care therapies, combining traditional healing agents and loading silver nanoparticles into nano cabs, aloe Elgin hydrogels, the use of advanced products / techniques in propolis and honey dressing films hydrogel sheets. India has a rich flora, which is widely distributed across the country. Herbal medicines are the basis for treating and preventing various ailments and physical conditions in traditional practices such as Ayurveda, Unani and Siddha.

Components from plants play an important role in traditional and Western medicine. Drugs derived from plants have been a part of human and health care development for thousands of years. Plant-based medicines are commonly used in India and China. A survey conducted by the WHO reported that more than 80% of the world's population still relies on traditional medicines for various ailments. About 25 percent of medicinal plants in developed countries are based on plants and their derivatives. The use of medicinal plants by indigenous peoples in rural areas of developing countries is well known^{1,2}.

Wound: Wound has been outlined because of the disruption of anatomical or useful continuity of living tissue due to physical, chemical, microbial or electrical insult. Wound healing is that the body's process of making dermal and epidermic tissue.

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The healing cascade is activated once blood platelets are available in Contact with exposed albuminoid resulting in platelet aggregation and unharness of natural process factors to deposition of protein clot at the location of injury ³.

Wounds square major measure issues for the patient and practician alike; wounds affect the oversized variety of patients and seriously scale back their quality of life. Current estimates indicate that just about 6 million individuals suffer from chronic worldwide. There classify measure only a few Indian studies on the medical specialty of chronic wounds. In one study, the prevalence of chronic wounds within the community was rumored as 4.5/1000 population wherever as that of acute wounds was nearly doubled at 10.5/1000 populations ⁴. In recent years, there has been a major increase in the usage of natural remedies with notable healing potential for numerous sorts of skin disorders, like cuts, burns and wounds. Medicative herbs have provided higher safety wound healing products compared to chemical medicine with an additional affordable worth ⁵. Wound healing is the method that the skin goes through because it repairs harm from wounds. Their square measures 3 main sorts of wound healing, betting on treatment, and wound kind. This square measure is known as primary, secondary and tertiary wound healing.

Primary Wound Healing: It refers to once doctors shut a wound victimisation staples, stitches, glues or alternative styles of wound-closing processes. Closing a wound during this means reduces the tissue lost and permits the body to specialize in closing and healing a smaller-area wound instead of the larger initial wound. As an example, a doctor would possibly sew up an oversized cut instead of permit the body to heal over the whole cut.

Secondary Wound Healing: It happens once a wound that can't be sewn causes an oversized quantity of tissue loss. Doctors can leave the wound to heal naturally in these cases. This might be a lot of common for wounds that have a rounded edge, uneven cowl surfaces, or square measure on surfaces of the body wherever movement makes stitches or alternative closure strategies not possible. It depends upon the body's own healing mechanism. This method takes longer, which can

result to accrued wound size, the danger of infection and contamination, and alternative factors like the employment of bound medications.

Tertiary Wound Healing: It happens once there is a requirement to delay the wound closing method. This might be necessary if a doctor fears that they may trap infectious germs during a wound by closing it. In these cases, they will permit the wounds to empty or stay up for the consequences of alternative therapies to require place before closing the wounds ⁶.

Classification of Wounds: Wounds are classified as open and closed wounds on the underlying reason behind wound creation and acute and chronic wounds on the idea of physiology of wound healing.

1. Open Wounds: In this case, blood escapes the body and haemorrhage is clearly visible. It's any classified as; incised wound, laceration or tear wound, abrasions or superficial wounds, puncture wounds, penetration wounds, and gunfire wounds ⁷.

2. Closed Wounds: In clos3d wounds, blood escapes the vascular system however remains within the body. It includes contusion or bruises, haematomas or blood growth, crush injury *etc.*

3. Acute Wounds: It may be a tissue injury that ordinarily precedes through an orderly and timely reparative method that leads to sustained restoration of anatomic and purposeful integrity. It classified measure sometimes caused by cuts or surgical incisions and complete the wound healing method among the expected time frame ⁸.

4. Chronic Wounds: Chronic wounds classified wounds that have didn't progress through the traditional stages of healing and so entered a state of pathologic inflammation; chronic wounds either need a chronic time to heal or recure often times native infection, hypoxia, trauma, foreign bodies and general issues like DM, deficiency disease, immunological disorder or medications for most frequent causes of chronic wounds ⁹. The response to injury, either surgically or traumatically elicited, is immediate, and also the broken tissue or wound then passes through 3 phases to have an effect on a final repair:

- The inflammatory section
- The proliferative section
- The remodelling section

The inflammatory section prepares the world for healing and immobilizes the wound by inflicting it to swell and become painful in order that movement becomes restricted. The fibro plastic section rebuilds the structure; then, the remodelling section provides the ultimate kind.

The Inflammatory Phase: The inflammatory section starts straight away once the injury that typically lasts between 24 and 48 h will persist for up to two weeks in some cases. They launch the haemostatic mechanism to right away stop blood loss from the wound site.

The clinically recognizable cardinal sign of inflammation, rubor, calor, tumor, heartbreak, and fuction-laesa seem because of the consequence. This section is characterized by constriction and protoplasm aggregation to induce clotting and afterward vasodilation and body process to supply inflammation at the wound site ¹⁰.

The Proliferative Phase: The proliferative phase primarily involves the generation of the repair materials and the majority of the striated muscle injuries ¹¹.

Remodelling Phase: This is a necessary part of tissue repair and is usually unnoticed; the ultimate outcome of those mixed events is that the broken tissue will be repaired with the scar ¹¹.

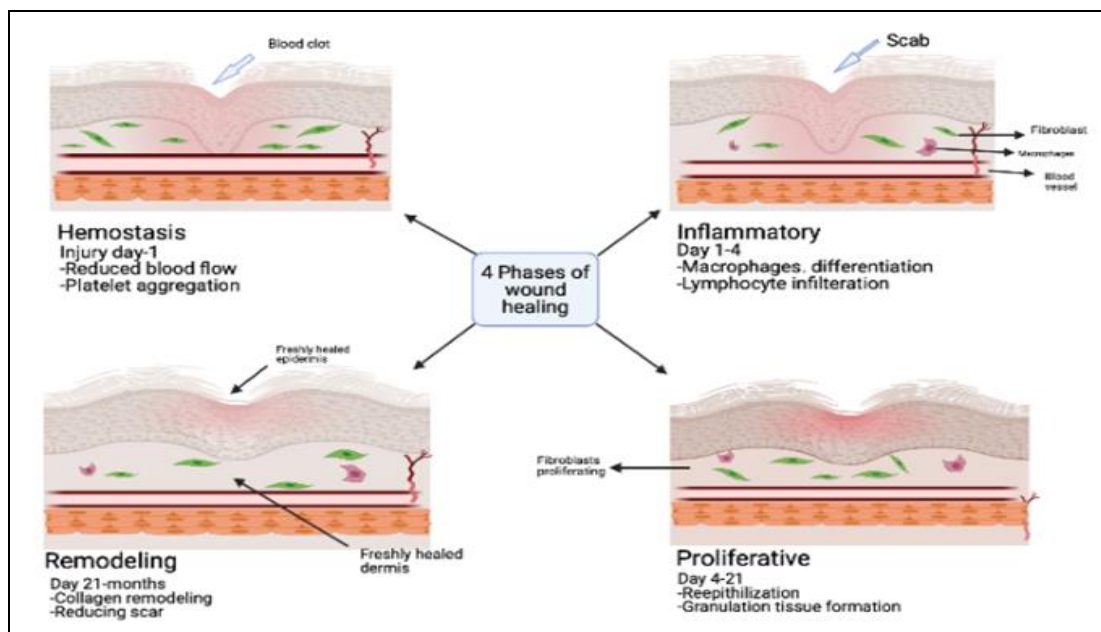


FIG. 1: MECHANISM OF WOUND HEALING

Plant Profile: Tridax flower usually called “Ghamra” and in English popularly known as “Coat buttons” because of appearance of flowers that has been extensively utilized in ayurvedic system drug of medication for numerous ailments and is distributed for “Bhringraj” by a number of the practitioners of writing that is documented medicine for liver disorders ¹².

Botanical name: *Tridax procumbens*.

Common names:

Marathi: kambermoodi, jakhamjudi, tantani.

Hindi: Ghamra

English: coat buttons, Tridax flower

Sanskrit: jayantiveda

Classification:

TABLE 1: TAXONOMIC CLASSIFICATION OF TRIDAX DAISY

Kingdom	Plantae
Sub-kingdom	Tracheobionta
Division	Spermatophyte
Subdivision	Magnoliophyte
Class	Magnoliopsida
Sub-class	Asteridae
Order	Asterales
Family	Araceae
Genus	Tridax
Species	Procumbens

Biological source: it is obtained from fresh leaves juice of *Tridax procumbens*.

Parts used: whole plant (leaf, stem, flower, root etc.)



FIG. 2: WHOLE PLANT OF TRIDAX DAISY

Geographical Source: the plant is native of tropical America and naturalized in tropical Africa, Asia, Australia and India. It is a wild herb distributed throughout India¹².

Description: An annual spreading herb grows up to 20 cm tall. Leaves: straight forward, opposite, serrate or rough, acute, fleshy and pubescent. Flowers: flowers like yellow-centered white or yellow flowers with three-toothed rays. Fruits: Hard achenes covered with stiff hairs and having a feathery white calyx at one end. Seeds: various, little with a tuft of satiny hairs on one aspect for wind spreading. Flowers and fruits seem throughout the year. Coat buttons are found along roadsides, waste grounds, dikes, railroads, riverbanks, meadows and dunes. Its widespread distribution and importance as a weed are due to its spreading stems and abundant seed production¹².

Macroscopy: *Tridax procumbens* could be a little perennial herb having short, bluish blade-like leaves. Corolla is yellow in color. It is a common weed that grows in open places, coarse-textured soils of tropical regions, sunny, dry localities, waste areas. The stem is ascending 30-50 cm in height, branched, rooting at nodes. Leaves are simple, opposite, stipulate, simple to ovate. 3-7 m long on an irregular basis toothed margin, base wedge-formed, shortly and leaf stalk, hairy on both surfaces. Flowers are tubular, yellow with hairs, inflorescence capitulum¹³.

Microscopy: Microscopic studies were carried out by preparing thin sections of leaf, stem, and petiole. The thin sections were collected in a watch glass and bleached with a bleaching agent along with little boiling.

Thin sections were further washed with water, stained with safranin, and mounted in glycerin for observations¹⁴.

Petiole: The leaf stalk was found to be an excretory organ form towards the distal finish, and crescent formed towards the bedded aspect. Single stratified stratum was coated with cuticle and interrupted by simple, cellular, 3-5 celled trichomes. Hyper dermis was 1-2 celled and collenchymatous. Ground tissue parenchymatous, vascular bundle 5, the size of the vascular bundles varies from center to margin that is large to small. These were centripetal that is xylem surrounded by the phloem.

Root: Dicot type of root is present in *Tridax procumbens*, and it consisted of 2-3 layered cells cork, 8-12 layered cells epidermis, xylem, phloem, medullary rays.

Leaf: Transverse section of leaf showed dorsiventral, stratum single bedded on each the surfaces and lined with thick cuticle. T.S. passing through the middle rib region showed slight depression on the ventral aspect and slightly protuberated on dorsal side. The basal cells of the trichomes were swollen, and trichomes looked like claw. Meristeel consists of single centrally located collateral vascular bundle surrounded by some parenchymatous cells filled with dark content. T.S. passing through the laminar region shows single layered palisade cells just below the epidermis followed by 5-7 celled mesophylls, parenchyma mostly devoid of intracellular spaces.

Stem: The epidermis was single-layered, thick-walled, narrow, and small is surrounded by trichomes. Cork cells consisted of 2-4 layers, vascular bundles were surrounded by polygonal lignified parenchymatous cells, above the cambium, many patches of small group of sieve tissue were embedded in parenchymatous cells.

Phytochemical Constituents: The leaf and alternative components of *T. procumbens* are reported to have flavonoids, alkaloids, carotenoids, hydroxycinnamates, lignans, benzoic acid derivatives, phytosterols, tannins, crude proteins, soluble carbohydrates, and calcium oxide¹⁵. The presence of fumaric acid, β -sitosterol, and pentacyclic triterpenoid oleanolic acid have also been reported¹⁵.

Luteolin, glucoluteolin, quercetin and isoquercetin have been reported in flower extracts¹⁶. Some of the other phytochemicals present abundantly in the *T. procumbens* are 2,6-dihydroxyacetophenone, 2-O- β -D-glucopyranoside, echiodinin, pinostrobin, dihydroechiodinin, tectochrysin-5-glucoside, methyl salicylate glucoside, 5,7,8-trimethoxyflavone, skullcapflavone-2-methyl ether, androechin, tectochrysin, 5,7,2'-trimethoxyflavone, echiodin, skullcapflavone ii, 5,7-dimethoxyflavone and andrographidine.

Flavonoids: A recent study has incontestable the presence of 23 flavonoids in *T. procumbens* [19] with total content around 65g/kg. kaempferol and catechin and its derivatives (-)-epicatechin, (+)-catechin, (-)-eigallocatechin, (+)-gallo catechin, (-)-epigallocatechin-3-gallate (EGCG) and (-)-epicatechin-3-galleate account for concerning 17.59% and 26.3% respectively. The remaining 56.11% represent 16 flavonoids particularly biochanin, apigenin, naringenin, daidzein, quercetin, butein, robinetin, baicalein, nobiletin, genistin, ellagic acid, luteolin, myricetin, baicalin, isorhamnetin and silymarin¹⁷. The flavonoids detected in *T. procumbens* are identified to mediate medicine activities as well as atom scavenging, medicament, antiallergic, antiplatelet aggregation, antimicrobial, antiulcer, antiviral, growth and antihepatotoxicity¹⁸. Two new flavones 8,3'-dihydroxy-3,7,4'-trimethoxy-6-O- β -D-glucopyranosyl flavone and 6, 8, 3'-trihydroxy-3, 7, 4'-trimethoxy flavone were isolated from the entire plant together with 4 known compounds Puerarin, esculetin, oleanolic acid and betulinic acid exhibiting antioxidant activity¹⁹. A new flavonoid Procumbenetin, from the aerial parts of *T. procumbens*, has been characterized as 3,6-dimethoxy-5,7,2',3'4'-pentahydroxy flavone 7-O- β -D-glucopyranoside based on spectroscopic techniques and by chemical means²⁰. Kaempferol is the main flavonoid found in the leaves of *T. procumbens* preclinical studies have shown that kaempferol and its glycosidic derivatives exhibit wide range of medicinal properties such as antioxidant, anti-inflammatory, analgesic, antimicrobial, antifungal, anticancer, cardioprotective, antidiabetic and antiallergic activities. Kaempferol has many beneficial effects on inflammatory diseases by mediating anti-inflammatory or immunomodulatory activities.

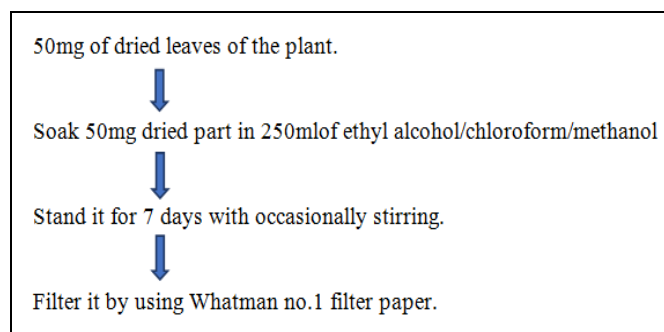
It inhibits various signalling pathways and suppresses matrix degrading enzymes.

Other Phytochemicals: The other bioactive molecule in the leaves of *T. procumbens* includes caffeic acid and ferulic acid, tannins, stigmasterol and lutein^{20, 21}. *In-vitro* studies have shown that caffeic acid and ferulic acid have antioxidant, anti-inflammatory, anticancer, and antimicrobial activities^{22, 23}. Tannic acid and other hydrolysable tannins have multiple health benefits, including reduced risk of cardiovascular disease, anticancer *etc.*^{24, 25}. One of the molecular mechanisms attributed to health benefits is the inhibition of Ca²⁺ activated Cl⁻ channels²⁶. Stigmasterol has pharmacological activities such as anti-hypercholesterolemic, cytotoxicity, antitumor, antioxidant, anti-inflammatory and analgesic²⁷.

Authentication of the Plant Material: The plant material was collected and authenticated in the month of February 2021 from the college campus of KLE college of pharmacy, Belagavi, Karnataka, India. The fresh parts of the plant were used for microscopic studies. The leaves were separated, dried, coarsely powdered, and stored in a closed container for further studies. Macroscopic and microscopical characters were studied as described in the quality control method. Microtome sections were taken, stained, mounted, and observed under binocular and projection microscope.

Plant Extraction: The collected leaves of *Tridax Procumbens* Linn were washed and dried under shade.

Leaf Extraction (Cold Maceration Process):



Physicochemical Properties: Physicochemical parameter includes moisture content, total ash, acid insoluble ash, water-soluble ash, water-soluble extractives and alcohol soluble extractive²⁸.

Phytochemical Screening: For the qualitative phytochemical analysis, acetone and methanolic extracts of *Tridax procumbens* plant leaves were tested using standard protocols^{29,30-33}.

Detection of Steroids: For detection of steroids, 0.5 ml of extract was dissolved in 5 ml of chloroform to this mixture, concentrated sulphuric acid was added from the side of the test tubes upper layer at surface appeared red and acid layer showed yellow with green fluorescence indicating the presence of steroids.

Detection of carbohydrates:

Molisch's Test: 1 ml of extract was treated with few drops of Molisch's reagent and few drops of concentrated H₂SO₄ from the side of the test tube; formation of the violet ring at the junction of two layers indicates the presence of carbohydrate.

Benedict's test: 1 ml of extract was treated with benedict's reagent and boiled for few minutes, and observed for the formation of red precipitate indicating the presence of carbohydrates.

Detection of Proteins:

Xanthoproteic Test: 3 ml of extract was treated with few drops of concentrated nitric acid, resulting in the appearance of yellow color indicating proteins.

Ninhydrin Test: 3 ml of extract was treated with 3 ml of ninhydrin reagent and allowed to boil for a few minutes, resulting in the formation of blue color indicating amino acids.

Detection of Anthocyanins: For detection of anthocyanins 3 ml of extract was treated with few drops of alcoholic FeCl₃ solution appearance of bluish black color indicates the presence of phenols.

Detection of Tannins: For detection of tannins 3ml of the extract was added to 1% of lead acetate, formation of a yellow precipitate indicates the presence of tannins, and in the same way, 3 ml of extract was treated with 3 ml of FeCl₃ appearance of green color indicates the presence of condensed tannins.

Detection of Alkaloids: For detection of alkaloids, the concentrated extract was treated with 2 ml of

diluted HCL and the mixture was gently heated for 20 min allowed to cool and filtered. The filtrate was used for Hager's and Wagner's test. Hager's test- when the filtrate was treated with Hager's reagent appearance of a yellow coloured precipitate indicates the presence of alkaloids. Wagner's test-when the filtrate was treated with Wagner's reagent, formation of reddish-brown precipitates indicates the presence of alkaloids.

Detection of Saponins: For detection of saponins, the plant extract was subjected to a frothing test. 5ml of warm aqueous extract was subjected for vigorous shaking and observed for the formation of stable foam, indicating the presence of saponins.

Detection of Flavonoids: For detection for flavonoids, an alkaline reagent test was performed. The extract was treated with 10% NaOH solution, resulting in the formation of deep yellow color indicating flavonoids.

Pharmacological Action:

Wound Healing Activity: Aqueous extract of *T. procumbens* (leaves) helps heal and helps in the steroid depressed healing in experimental male Wister rats. The increased lysyl oxidase activity induced by the preparation has been checked to show the wound healing activity. The increased nucleic acid level indicates the action at cellular level³⁴. Leaf juice of *Tridax procumbens* was shown to depress wound contraction in experimental animals.

Cardiovascular Effects/Hypotensive Effects: An aqueous extract from the leaf of *T. Procumbens* was studied and obtained positive results on the anesthetized Sprague-Dawley rat³⁵.

Hepatoprotective Activity: The hepatoprotective activity of aerial parts of *Tridax procumbens* was investigated against d-galactosamine / lipopolysaccharide (d-GalN/LPS) induced hepatitis in rats.

Antimicrobial Activity: The whole plant of *Tridax* has been reported for its antimicrobial activity on various species of bacteria. A whole plant is squeezed between the palms of hands to obtain juice. Fresh plants juice is applied twice a day for 3-4 days to cure cuts and wounds.

The extract of the whole plant of *Tridax* showed antimicrobial activity only against *Pseudomonas aeruginosa*. The disk diffusion method was used to test the antibacterial activity³⁷.

Immunomodulatory Activity: Ethanolic extracts of leaves of *Tridax* have immunomodulatory effect on Albino rats dosed with *Pseudomonas aeruginosa* also inhibits proliferation of same³⁸. Also, a significant increase in phagocytic index, leukocyte count, and splenic antibody-secreting cells has been reported to ethanol insoluble fraction of aqueous extract of *Tridax*. Stimulation of humoral immune response was also observed along with elevation in haemagglutination antibody titer. The study also reveals that *Tridax* influences both humoral as well as cell-mediated immune systems³⁹.

Antiparasitic Activity: Parasitic infections caused by protozoa, nematodes, trematodes, and cestodes account for more than 30% of the human population, and the plant extracts and their secondary metabolites may be an excellent strategy to target these infections⁴⁰.

Medicinal Uses: In this modern decade, there are many people suffering from the deprivation of even essential need and the urge to survive has prompted them to explore naturally available resources for therapeutic effects with respect to common ailments, including inflammation. Inflammation is a common reaction of the body to be insult caused by various biological and nonbiological factors present in the environment. The procumbent is valued for its pharmaceutical properties.

Traditional Uses: It is mostly used as an anticoagulant, hair tonic, antifungal, and insect repellent, in bronchial catarrh, diarrhoea, dysentery, and wound healing. Traditional and complementary medicine is increasingly recognized as an integrative approach to health care in many countries (WHO, 2013). The use of plants for medicinal purposes may date back to the Middle Paleolithic age, approximately 60,000 years ago.

T. procumbens is used to treat anemia, colds, inflammation, and hepatopathies in Central. In Guatemala worldwide. *T. procumbens* is used as an antibacterial, antifungal and antiviral. The entire plant is used for the treatment of protozoal

infections with malaria, leishmaniasis, and dysentery. The leaf juice is used to treat wounds and stop bleeding. This species is also used to treat gastrointestinal and respiratory infections, high blood pressure, and diabetes. Aqueous extracts of *T. daisy* have strong anti-plasmodial activity against chloroquine-resistant⁴¹.

CONCLUSION: Wounds are physical injuries that result in an opening or break of the skin. Proper healing of wounds is essential for restoring disrupted anatomical continuity and disrupted functional response of the several cell types to injury. Cutaneous wound repair is accompanied by an ordered and definable sequence of biological events starting with wound closure and progressing to the repair and remodelling of damaged tissue. This review tries to focus on the benefits and why it is needed to continue research plants known to be used in traditional medicine that could lead to discovering and creating new conventional medicines. *Tridax daisy* has a long history of traditional use. Still, isolation and evaluation of each phytochemical have not been properly related to its pharmacological properties and could show difficult reproducibility after isolation and evaluation. Different extracts have been used for the isolation of metabolites and for treating different ailments. The studies on plant *Tridax daisy* also desire novel therapeutic agents from the various types of compounds with diverse pharmacologic properties isolated from them. Therefore, there is huge room for research in the direction of more pharmacological activities of plants and to elucidate the mechanism of action of same in future.

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