

## PHARMACOGNOSTICAL AND PHYTOCHEMICAL INVESTIGATION OF LEAVES OF A WEED TRIDAX PROCUMBENS LINN.

MANISHA SUTAR, KOMAL MALVANKAR AND SONIA SINGH

Alard College of Pharmacy, Pune-57, Department of Pharmacognosy. Email: manishasutar01@gmail.com

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### ABSTRACT

*Tridax procumbens* Linn. (Compositae) is a weed found throughout India. The plant is native of tropical America and naturalized in tropical Africa, Asia, and Australia with increase in use of herbal plants for therapeutic purpose, there is a need for the pharmacognostical identification of the herbs which are widely grown. The *Tridax procumbens* is a weed and pest plant selected for the study is collected from Marunji village of Pune District. This weed is having several therapeutic applications in Ayurveda and reviewed for its ethnopharmacology. To mark its official standards it is necessary to study the plant in crude and powdered form which helps to indentify the different species of tridax. Pharmacognostical study of the leaf shows the lanceolate and ovate shape, wide with irregular serrate margin and pubescent on both side. Microscopically, the midrib and laminar region showed a distinct epidermis. The upper epidermis was single layer, with a small, rounded shape, covering multicellular trichome and the lower epidermis had single layer, elongated cells, arranged closely. Physiochemical evaluation and fluorescence study was carried out. Different extracts were investigated for the presence of preliminary phytochemicals like glycosides, alkaloids, carbohydrates, flavonoids and fatty acid. The present study might be useful to provide the information in regard to its identification parameters and it was thought worthwhile to explore the endangered weed on the basis of its quality control parameter.

**Keywords:** Leaf of tridax procumbense, Macroscopy, Microscopy, Phytochemical study and fluorescence analysis

### INTRODUCTION

The compositae (Asteraceae) is an advanced and botanically highly specialized family of herbaceous plants, herbs, shrubs, or less commonly trees and are arguably the largest family of flowering plants, comprising about 1400 species out of which 674 species are found in India. Some of them are tropical trees and shrubs while few are members of herbs [1]. *Tridax procumbens* is a species of flowering plant in the daisy family (Compositae), a common weed in west Africa, subregion and other tropical zones of world and known as coat button in English, jayanti veda in Sanskrit, Ghamra in Hindi and Dagadi pala in Marathi. It is best known as widespread weed and pest plant can be found in fields, meadows, croplands, disturbed areas, lawns, and roadsides [2]. It is a semi prostrate, annual, creeper herb. Stem is ascending 30-50cm height, branched, sparsely hairy, rooting at nodes. Leaves are simple, opposite, exstipulate, lanceolate to ovate. 3-7 cm long irregularly toothed margin, base wedge shaped, shortly petioled, hairy on both surfaces. Flowers are tubular, yellow with hairs, inflorescence capitulum. *Tridax* has two types of flower: ray florets and disc florets with basal palcentation. Flowering-Fruiting throughout the year. Fruit is a hard achene covered with stiff hairs and having a feathery, plume like white pappus at one end [3]. It shows the presence of fumaric acid, luteolin and glucoluteolin and  $\beta$ -sitosterol-3-O- $\beta$ -D-xylopyranoside [4], lipid constituents [5], saturated and unsaturated Fatty acid [6] and procumbenetin a flavonoid [7]. Alcoholic extract of the aerial parts also exhibited hair growth promoting activity in albino rats [8]. The extract of *Tridax procumbens* also posses anti-diabetic effect [9, 10], haemostatic activity [11], anti-inflammatory activity on Wistar Albino rats [12, 13], antimicrobial activity on *S. aureus*, *P. mirabilis* and *E. coli*, cardiovascular effects [15] and immunomodulatory effects [16]. The leaf juice possesses antiseptic, insecticidal and parasiticidal properties and is used also to check haemorrhage from cuts, bruises and wounds. Its leaves also use for bronchial catarrh, dysentery, diarrhoea [17] and it helps to prevent falling of hairs and promotes the growth of hairs [18], used as insect repellent [19] and aqueous and acetone extract of flower gives anticancer activity [20]. In the West Africa sub-region and tropical zone of the world, Traditional medical practitioners and the native peoples of these areas use the leaves of the plant as a remedy against conjunctivitis and in vitro radical scavenging [21].

### MATERIAL AND METHOD

#### Plant material collection and Authentication

The fresh leaves of *Tridax procumbens* L. were collected in the month of October from Marunji village, District -Pune (MS), India. The plant of *Tridax procumbens* was authenticated by Prof. T Srinivasu, Department of botany, Institute of Science, Mumbai. Collected fresh leaves were washed and sun dried and used for the further investigations.

**Reagents:** All the reagents were of analytical grade.

#### Methods

The organoleptic and morphological characters of the leaves were studied [22] under dissecting microscope. Transverse sections were taken and studies [23], for the different histological analysis along with the surface preparation. Where as the dried powder material was studied for microscopical characters [24] like stomata, trichome, lignified cells, palisade cells, parenchymatous cells, calcium oxalate crystals and xylem vessels. Powder drug was also studied with routine chemical reagents to observe the fluorescent effect under UV (Short wave length and long wavelength). Physicochemical parameters were determined [25] like ash value, extractive value, loss on drying etc. Phytochemical constituents were identified by performing the preliminary phytochemical investigation [26] for different extracts. The extracts were prepared by using water, ethanol, methanol, chloroform and ethyl acetate. The results for all the parameters were illustrated and photographs of the microscopical characters were taken.

### RESULT AND DISCUSSION

#### Leaf morphology

Leaves were simple, opposite, serrate or dentate, acute, fleshy and pubescent, 3 to 7 cm long, 1 to 4 cm wide, with irregularly toothed margins; base wedge-shaped, shortly-petiole, more hairy on lower surface than upper surface, shown in Fig 01 (a) and (b).

#### Leaf Microscopy

#### Surface preparation

leaf shows the presence of anisocytic type of the stomata covered with thin walled epidermal cells. Leaf surface also shows the

presence of simple, multicellular (3-6 celled) covering trichome shown in Fig 02(a) and (b).

Single layered epidermis covered with the cuticle and interrupted by simple and 3-5 celled multicellular covering trichomes and anisocytic type of the stomata at the lower epidermis.

#### Transverse Section of Leaf

The leaf is dorsiventral, epidermis single layered on both the surfaces and covered with thick cuticle. T.S passing through the midrib region shows slight depression on ventral side and slightly protuberated on dorsal size. Upper epidermis shows single layered multicellular covering trichome. Lower epidermis is single layered, elongated cell and closely arranged. Spongy parenchyma is multi-layered, loosely arranged, round shaped and shows pink colored xylem vessels. Spongy parenchyma shows the presence of calcium oxalate crystals. Transverse section passing through the laminar region shows bilayered palisade cells just below the upper epidermis. Upper epidermis followed by 5-7 celled mesophyll parenchyma mostly devoid of inter cellular spaces. Palisade cells are bilayered, tubular shaped and compactly arranged. Vascular bundles is concentric in shape, Meristeel consists of single centrally located collateral vascular bundle surrounded by some parenchymatous cells filled with content, shown in Fig 03(a) and (b).

#### Powder analysis

It is dark green, fine, odorless powder with slight bitter taste. The powder microscopy reveals the presence of multicellular covering trichome, anisocytic type of the stomata, calcium oxalate crystals, xylem vessels and phloem fibers. Powder shows the spiral thickenings vascular bundles, elongated palisade cells and thin walled parenchyma cells, shown in Fig 04.

#### Physico-chemical Parameters

##### 1. Description of leaf powder

Leaves were dried in sunlight and powder was prepared which shows color. Same powder was subjected to various physicochemical reactions.

##### 2. Ash Value

Percentage of Total ash, Acid-insoluble ash, Water-soluble ash values of the powdered drugs were performed as per Pharmacopoeial standard procedure and result were reported in Table 01.

##### 3. Extractive values

Water soluble and Alcohol soluble extractive values were determined according the Pharmacopoeial standard procedure and result were reported in Table 01.

##### 4. Moisture content determination

Moisture content determination was carried out by loss on drying method according to the pharmacopoeial standard procedure and result were reported in Table 01.

#### Fluorescence Analysis of leaf powder [27, 28]

The powder was subject to fluorescence analysis with different acids and reagents. The behavior of powdered drugs with different acid and chemical reagent were observed under UV light and visible light as per the standard procedure and results were reported in Table 02.

#### Extraction of powdered leaves with different solvents

Extracts were prepared with various solvents like ethanol, methanol, ethyl acetate, chloroform and aqueous extract was prepared. Percentage yield of extract obtained were reported in Table 03.

#### Preliminary Phytochemical investigation

The ethanolic, methanolic, ethyl acetate, chloroform and aqueous extracts were investigated for the presence of preliminary phytoconstituents and result were reported in Table 04.

#### DISCUSSION

In the last two decades of the century the scientists are sincerely trying to evaluate many plant drugs used in traditional system of medicine. The pharmacognostical and phytochemical data is one of the major criteria for identification of plant drug those supplied in crude as well as powdered form. It also helps to differentiate the closely related species or varieties with similar pharmacology and chemical constituents. The leaves are greenish with a smooth and thick texture and possess number of trichomes on both leaf surfaces. The microscopy shows (Fig 3 a and b) the presence of vascular bundle, collenchymas, spongy parenchyma and Palisade cells. Palisade cells are elongated, bilayered, tubular shaped and compactly arranged. Surface of the leaf show presence of simple multicellular covering trichome and anisocytic stomata (Fig.2 a and b), while the powdered drugs shows the presence of trichomes, stomata, xylem vessels and phloem fibers, palisade cells, calcium oxalate crystals and epidermal cells (Fig 4).

The physicochemical constants such as total ash 12%, water soluble ash (4.05%), acid insoluble ash (2.35%), water Extractive (2.8%), alcohol Extractive (2.4%) and loss on drying (4.66%) were determined (Table 1). The soluble extractive value (Table 3) with different solvents like ethanol (6%) Methanol (7.2%), Chloroform (5%), Ethyl acetate (6.8%) and Aqueous extract (26%) and investigated for the types of chemical constitues present.

The behavioral changes of powder when treated with different acid and reagents were studied as fluorescence analysis. It shows different colors under daylight and UV (short and long wavelength) light. Various qualitative chemical tests were carried out for the different extract which indicates the presence of alkaloid, glycosides, phenols and flavonoids. Thus the proposed work was aimed to design a perfect protocol for the identification of different species and result found to be significant and encouraging towards the goal for standardization. Further it provides a good platform to study its medicinal value in the treatment of diseases and for well being of the human health.



Fig. 01(a): Leaves of *Tridax procumbens*



Fig. 01(b): Upper and lower surface of leaf

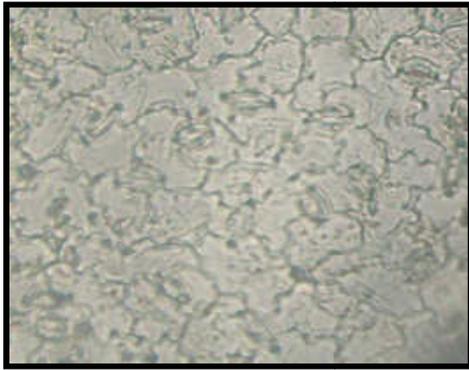


Fig. 02(a): Stomata of leaf



Fig. 01(b): Trichome of leaf

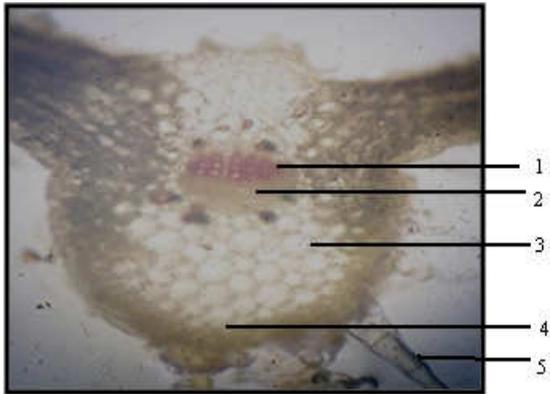


Fig. 03(a): lamina with midrib of leaf

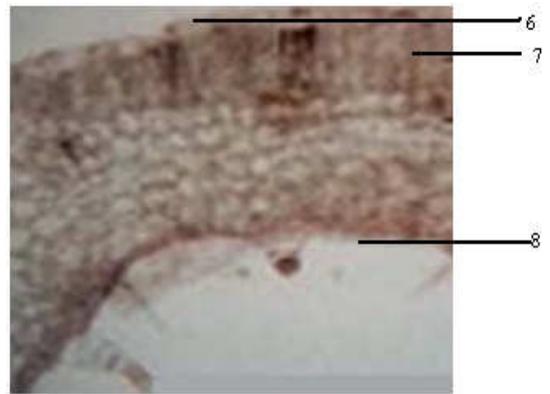
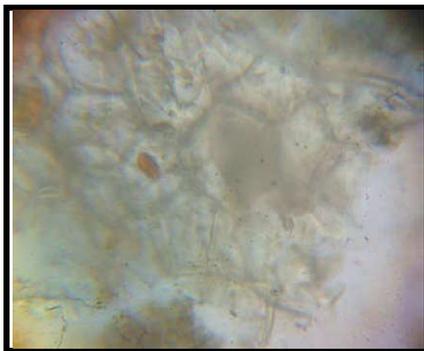


Fig. 03(b): lamina showing palisade cells

- 1. Xylem
- 2. Phloem Cells
- 3. Spongy parenchyma
- 4. Collenchyma cells

- 5. Trichome
- 6. Upper epidermis
- 7. Palisade Cells
- 8. Lower epidermis



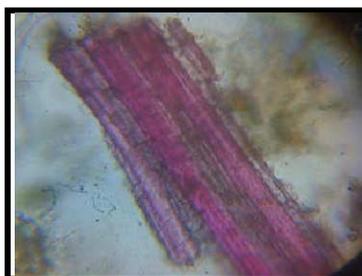
Epidermal Cells



Spiral Vessels



Stomata



Phloem fibers



Palisade cells



Trichomes Calcium



Oxalate crystal

Fig. 04: Powder characters of *Tridax procumbense* leafTable 01: Physico-Chemical Evaluation of *Tridax Procumbense* Leaf

Physicochemical parameter	Result
Description	Green
Total ash	12%
Water soluble ash value	4.05%
Acid insoluble ash value	2.35%
Water Extractive	2.8%
Alcohol Extractive	2.4%
Loss on drying	4.66%

Table 02: Fluorescence analysis of *Tridax procumbense* leaf

Reagent	Color of powdered crude drug		
	Day light	Long wavelength	Short wavelength
Powder as such	Green	Green	Dark green
Powder+ water	Green	Green	Green
Saturated Picric acid	Yellowish green	Dark green	Yellowish-black
Powder + Conc HNO <sub>3</sub>	Radish	Light brown	Reddish brown
Powder+ Conc H <sub>2</sub> SO <sub>4</sub>	Black	Black	Black
Powder+ Conc HCL	Green	Black	Black
Powder + Glacial Acetic Acid	Dark green	Green	Black
Powder + Iodine solution(N/20)	Dark green	Dark green	Brown
Powder+ FeCl <sub>3</sub> (5%)	Yellowish brown	Dark green	Dark brown

Table 04: Preliminary Phytochemical investigation

Phytoconstituents	Chemical Test	Ethanol	Methanol	Chloroform	Ethyl Acetate	Aqueous
		Extract	Extract	Extract	Extract	Extract
Alkaloid	Mayer, s Test	+	-	+	+	-
	Dragendroff's Test	+	-	+	+	-
	Wagner's test	-	-	+	+	-
	Hager's Test	-	-	+	+	-
Carbohydrates	Molisch's Test	+	-	+	+	+
	Benedicts Test	+	+	+	+	+
	Fehling's Test	-	-	+	-	-
Glycosides	Brontrager's Test	+	+	+	+	+
	Modified Borntrager's Test	+	+	+	+	+
	Legal Test	+	-	+	+	-
Phenol	Ferric chloride test	-	-	+	-	-
	Dil HNO <sub>3</sub> test	-	-	-	+	-
Protein	Biuret Test	+	-	-	-	-
	Million's Test	+	-	-	-	-
	Precipitation test	-	+	-	-	-
	5% CuSO <sub>4</sub>	-	+	-	-	-
	5 % Lead Acetate	-	+	-	-	-
Flavoniods	5%mercuric chloride	-	+	+	-	-
	Shinoda test	+	+	-	-	-
	Alkaline reagent test	+	+	-	-	-

Table 03: Extraction of *Tridax procumbens* leaf with different solvents

Extract( based on type of solvent used )	% w/w
Aqueous	26
Chloroform	5
Ethanol	6
Methanol	7.2
Ethyl acetate	6.8

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