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RESEARCH ARTICLE

Pharmacognostical and Phytochemical Investigation of *Alternanthera Bettzichiana (Regel) Nicols.*

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ABSTRACT

The present study reveals the macroscopical, microscopical and preliminary phytochemical investigation of *Alternanthera bettzichiana* (Regel) Nicols. Very scanty reports are available on microscopical and phytochemical studies, hence, the present study was undertaken to investigate the same. Some of the diagnostic features of leaves and petiole are the presence of arc shaped petiole, amphistomatic, non-glandular trichomes, stomatal complex mostly diacytic and scanty cellular contents. While the preliminary phytochemical studies shows promising presence of an array of phytoconstituents *viz.*, alkaloids, steroids, cardiac and coumarin glycosides, proteins and sugars. The persona from the medical world runs after those plants who have been seated in the range of medicinal plants. But, the least exploited plants can occupy unparallel place in the natural components of the environment by carving their roles

KEYWORDS: *Alternanthera bettzichiana,* Pharmacognostical study, Preliminary phytochemical investigation.

INTRODUCTION

After decades of serious obsession with the modern medicinal systems, peoples have started looking towards the ancient healing systems like Ayurveda, Siddha and Unani. This is due to adverse effects associated with synthetic drugs [1]. Herbal medicine is a triumph of popular therapeutic diversity. India is one of the richest floristic regions of the world and has been a source of plants and their products, since antiquity, human beings utilized them according to needs, particularly as food and medicine. Among the flora, 35000 to 70000 species have been used for medicinal purpose [2]. Almost in all the traditional medicine, the medicinal plants play a vital role and constitute the backbone for the same. In order to make sure the safe use of these medicines, a necessary first step is the establishment of standards of quality, safety and efficacy [3]. Keeping this fact into consideration, the attempts were made to establish basic Pharmacognostical standards of the plant *Alternanthera bettzichiana* (Regel) Nicols. *Amaranthaceae* is a cosmopolitan family consisting of 64 genera and about 800 species, mostly abundant in tropical regions of America, Africa and India [4]. The family represented by herbs and few shrubs, contains most of the allergic species. The genus Alternanthera, a medicinally important member of family *Amaranthaceae* is widely used for the presence of volatile constituent, essential amino acids, flavone glycoside and steroids [5]. *A. bettzichiana*, a horticulture species [6] of many forms, is often found as an escape, reported as a wild edible material [7], for micro propagation studies [8], as adsorbent for removal of Cr (VI) [9], to purify and nourishing blood, antipyretic, as galactagogue and for wound healing. The parallel species of the same genus were also reported for their potential claims in certain viral diseases [10], as an immune modulator [11], protective against cancer [12] and in treatment of diarrhea [13]. The purpose of this study was to screen the microscopical characters and identification of phytochemical constituents of various extracts of *A. bettzichiana*.

MATERIAL AND METHODS:

Fresh samples of the plant *Alternanthera bettzichiana* were collected from the different parts of satpuda region of Chopda Tahsil, Jalgaon district in the month of September 2006. The identity of this plant was verified by Dr.D.A.Patil (Head, Dept. of Botany, S.S.V.P.S. College, Dhule) and authentificated from Botanical survey of India, Pune. A voucher specimen no.JCH 1 has been deposited at the Dept. of Biotechnology, school of life sciences, North Maharashtra University, Jalgaon. Collected samples were thoroughly washed with running water to remove the adhered soil and dried. Free hand sections of single leaflets were taken from the preserved material and observed under microscope. Microscopical drawings were made with the help of camera lucida after clearing the sections. Macroscopic and microscopic characters were studied as described by Metcalf *et al* [14] and Datta *et al* [15].

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MORPHOLOGICAL CHARACTERS*:*

Leaves are dull green in color, with size 1.2-7.6 x 0.5-0.4 cm, elliptic, oblanceolate, rhomboid-ovate, acute or acuminate at apex, attenuate at base; petioles hairy. Heads 1-5 together, globose, sessile, Tepals 5, unequal, Utricle of 1.5 cm long, obvoid.

MICROSCOPY OF LEAF:

LEAF:

The leaves are dorsiventral and amphistomatic. The mid-rib region is shallowly channeled. The cells of upper epidermis are medium sized. They are compactly arranged in one layer. They are barrel to round in shape. The outer wall of epidermal cell is thicker than the inner wall. It is covered by thick cuticle from outside. The lower epidermis and cuticle present posses similar features as the upper ones.

The mesophyll is distinctly recognizable in to the upper palisade layer and lower spongy tissue. The cells of the former in one row, compactly arranged and contain abundant chloroplast. The later has loosely arranged, irregular or rounded cells enclosing intercellular spaces in between them. They contain fewer chloroplasts.

The internal structure of midrib region bears some interesting features. The cells on upper and lower sides are mostly considerable medium sized. Few cells are relatively smaller. The lower epidermis in this is followed by single layer chlorenchyma. The vascular tissue extends in the centre. It is represented by three separated vascular bundles. Conjunctive tissue is present surrounding to it. The cells of conjunctive tissue are also medium sized; very few cells are larger. They are thin walled, parenchymatous and polygonal (Fig.1).

PETIOLE:

In T.S., petiole is arc shaped and broadly notched adaxially. The outermost delimiting layer is the epidermis composed of medium sized cells with thick outer walls, which is covered by thick cuticle containing single layered chlorenchyma. The vascular tissue is present in the form of three vascular bundles. The central one is being larger and is embedded in the guard tissue. The cells are parenchymatous and polygonal (Fig.2).

EPIDERMAL CELL COMPLEX:

(A) Leaf adaxial: Stomata mostly diacytic, rarely anomocytic and anisocytic, orientation random, diffuse distribution. Subsidiary cells 2, mostly F-type [16], rarely C-type [16]. Walls undulate, sinuous with U-shaped, sides 2-4. Guard cells are typically chlorophyllous, elliptical with elongated pore. Epidermal cells chlorophyllous, sides mostly 5-6, walls undulate, sinuous U-shaped (Fig.3a).

(B) Leaf abaxial: Stomata mostly diacytic, orientation random, diffuse distribution. Subsidiary cells 2, mostly F-type. Walls undulate, sinuous with U-shaped, sides 2-4. Guard cells are typically chlorophyllous, elliptical with elongated pore. Epidermal cells chlorophyllous, sides mostly 5-7, walls undulate, sinuous U-shaped, thick walled (Fig.3b).

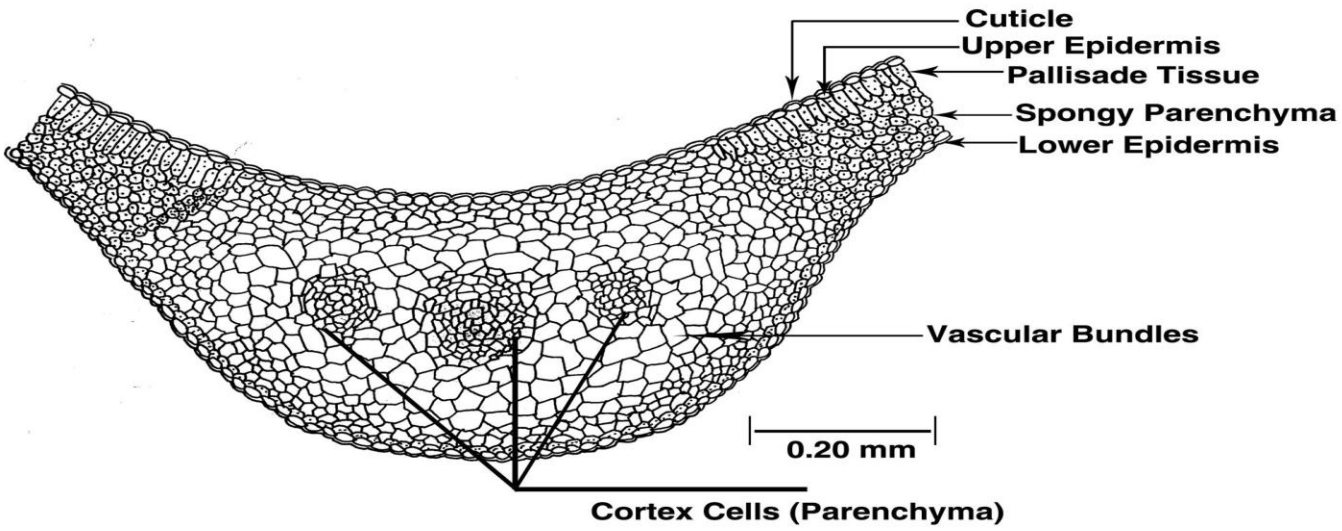
Stomatal frequency of the upper surface is lesser as compared to lower surface (Fig.3b and 3a). Stomatal index of lower epidermis is 12.78 and 10.70 of upper epidermis.

TRICHOMES:

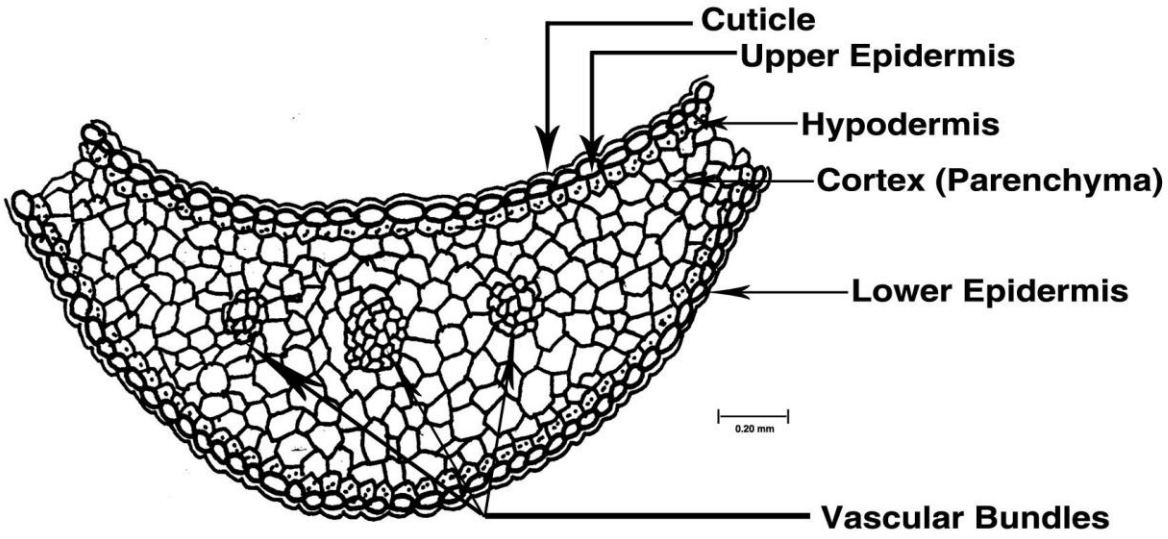
Non-glandular, bicellular, distributed particularly in mid-vein and veinlet region. Fort one celled, circular, thick, long, narrow towards the apex and apex is pointed (Fig.3c).

PHYTOCHEMICAL STUDIES:

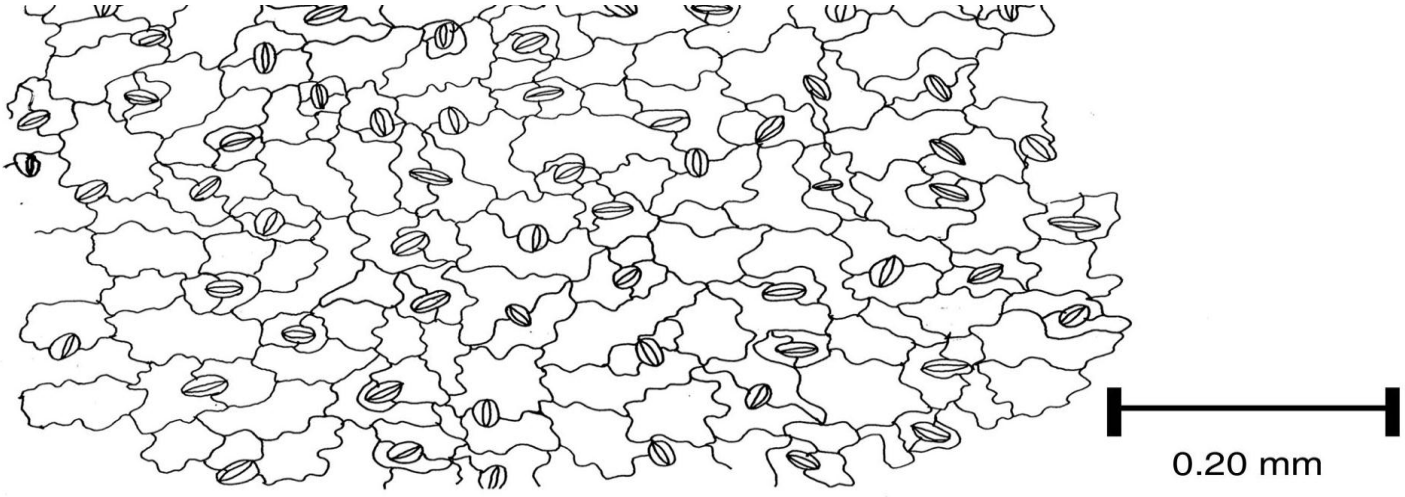
The plant material was collected and dried for several days in shade and powdered with the help of an electric grinder. The powdered material was then subjected for physicochemical evaluation using standard chemical tests. For preliminary phytochemical screening the dried powdered plant material was extracted successively with petroleum ether, chloroform, 90% ethanol in soxhlet extraction apparatus. An aqueous extract was prepared by using cold maceration process. Then extracts were filtered in hot condition and distilled under reduced pressure to get a solid mass. All the extracts were stored in the refrigerator and were subjected to the qualitative phytochemical screening for identification of active constituents using standard methods [17].



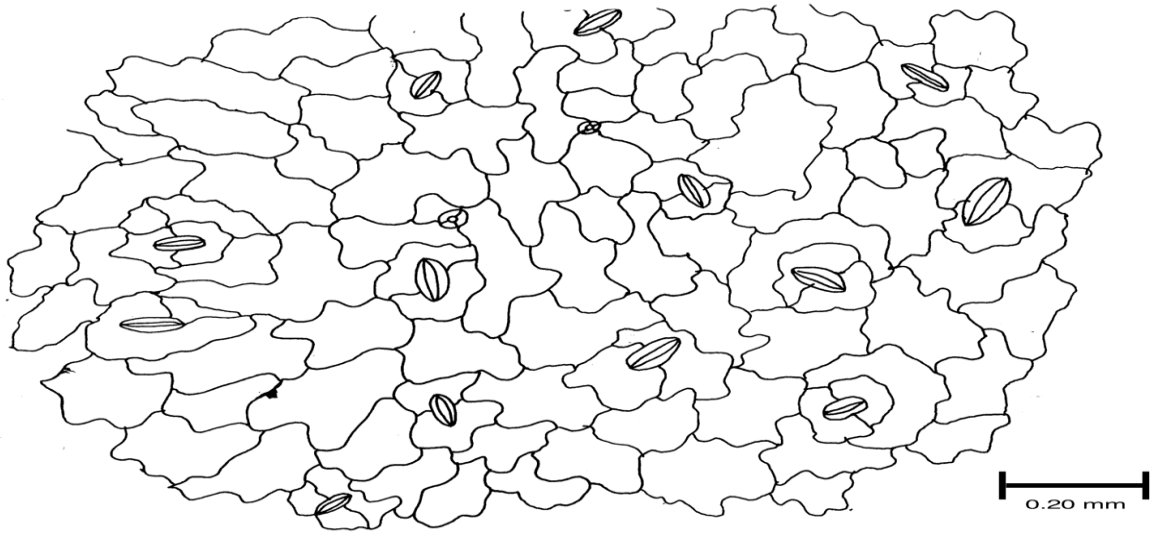
"Figure 1: T.S. of Leaf"



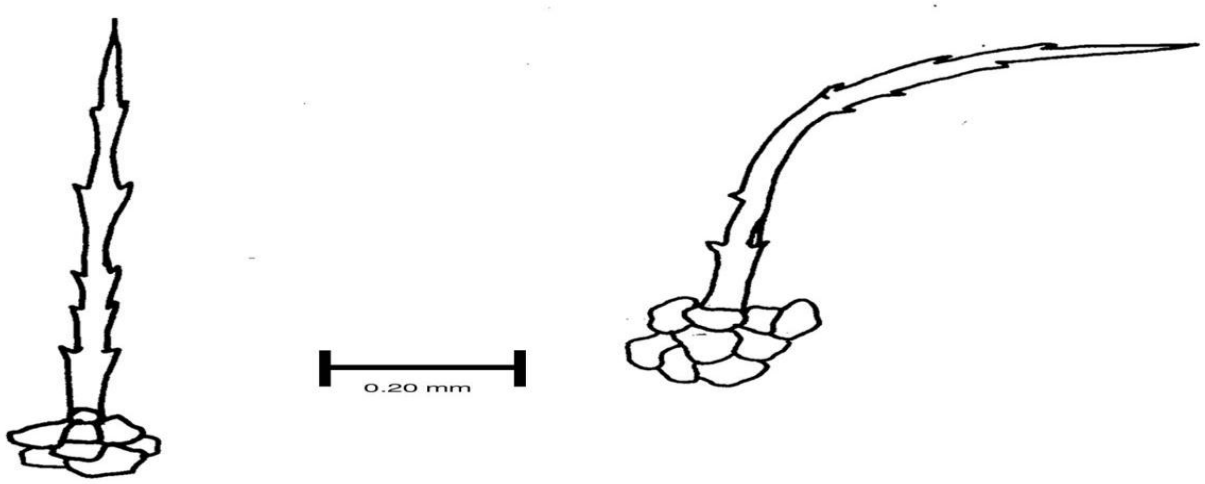
"Figure 2: T.S. of Petiole"



"Figure3a: Epidermal Cells (Leaf adaxial)"



"Figure3b: Epidermal Cells (Leaf abaxial)"



"Figure3c: Trichome- Uniseriate"
"Figure 3: Stomatal Complex
3a: Epidermal Cells (Leaf adaxial) 3b: Epidermal Cells (Leaf abaxial) 3c: Trichome- Uniseriate"

"Table 1: Physicochemical characters of *A. bettzichiana*"

Ash Value	Total ash	14 %
	Acid-insoluble ash	1.6 %
	Water insoluble ash	3.0 %
Extractive values	Petroleum ether (40-60^{o})	6.4 %
	Chloroform	25.6 %
	Ethanol	32.0 %
	Water	19.2 %

"Table 2: Preliminary phytochemical Analysis of *A. bettzichiana*"

Chemical Class	Pet. Ether extract	Chloroform extract	Alcoholic extract	Aqueous extract
Test for carbohydrates	-	-	-	+
Test for gums	-	-	-	+
Test For Proteins	-	-	-	+
Test for Steroids	-	**+**	**+**	-
Test for Cardiac glycosides	-	-	**+**	**+**
Test for Coumarin glycosides	-	-	+	+
Test for alkaloids	-	-	+	-

(+) Positive, (-) Negative

RESULT AND DISCUSSION:

In the present study fresh leaves appear with dull green adaxial side and green abaxial side; dried leaves exhibit brown above and pale brown below. Table 1 depicts that the total ash value was higher than that of acid insoluble and water soluble ash value and a decrease in the acid insoluble ash value may be due to presence of siliceous matters. The alcoholic extractive value was higher than the chloroform and aqueous extractive value revealing the presence of larger amounts of alcohol soluble constituents in the plant material. The microscopic characters of leaves of *A. bettzichiana* revealed the presence of cuticularised single epidermal layer. The mesophyll showed presence of single layer of upper palisade layer and loosely arranged spongy tissue with presence of fewer chloroplasts. The midrib region consists three separated vascular bundles is also a specific feature observed in the plant. The presence of diacytic stomata, bicellular and non-glandular Trichomes are the important diagnostic characters. The preliminary phytochemical investigation of the plant has shown presence of various important chemical constituent's *viz.,* alkaloids, cardiac glycosides, steroids and coumarin glycosides (Table 2). Pharmacognosy, one of the basic branch of pharmacy developed nearly two centuries ago to check purity and quality of drugs and other natural products, mostly from Tropical ones, has since changed from alpha Pharmacognosy to identification, detection, isolation, characterization and synthesis of naturally occurring compounds as emphasis changed, over the years, from natural products to synthetic drugs. Indigenous systems are almost exclusively dependant on herbal crude drugs which are highly prone to adulteration and substitution. It is, therefore, of utmost importance that standards are evolved which can help to distinguish the natural drugs from their adulterants and substitutes. Ethno botanical studies can play vital role in this direction [18]. In nutshell, present study is an attempt to establish some preliminary standards for precise and accurate identification of this pharmacologically and phytochemically less explored medicinal plant.

ABBREVATIONS USED:

F- type: Free subsidiary; subsidiary neither abuts on another stomata for any allo subsidiaries.

C- type: Common subsidiary; Coallosubsidiary which abuts on one or more adjacent stomata, but not any other cell.

T.S.: Transverse Section

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