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Pande J
Phytochemical, Pharmacological and Microbiological laboratory, Department of Biosciences (UGC-CAS), Saurashtra University, Rajkot, Gujarat, India

Padalia H
Phytochemical, Pharmacological and Microbiological laboratory, Department of Biosciences (UGC-CAS), Saurashtra University, Rajkot, Gujarat, India

Donga S
Phytochemical, Pharmacological and Microbiological laboratory, Department of Biosciences (UGC-CAS), Saurashtra University, Rajkot, Gujarat, India

Chanda S
Phytochemical, Pharmacological and Microbiological laboratory, Department of Biosciences (UGC-CAS), Saurashtra University, Rajkot, Gujarat, India

Correspondence
Pande J
Phytochemical, Pharmacological and Microbiological laboratory, Department of Biosciences (UGC-CAS), Saurashtra University, Rajkot, Gujarat, India

Pharmacognostic, physicochemical and phytochemical studies of *Andrographis echioides* Nees. and *Tridax procumbens* L. leaf and stem

Pande J, Padalia H, Donga S and Chanda S

Abstract
Pharmacognostic studies are very important since the parameters estimated are the identity of a particular plant and they are very useful to authenticate the plant under study and prevent it from adulteration and substitution. In the present work, pharmacognostic, physicochemical and phytochemical analysis of *Andrographis echioides* Nees. and *Tridax procumbens* L. leaf and stem has been attempted. The macroscopic, microscopic and powder study laid down the characteristic features of leaf and stem of the said plants. The crude powder of leaf and stem of both the plants was evaluated for the presence or absence of various phytochemicals like alkaloids, flavonoids, cardiac glycosides, tannins, saponins, triterpenes, etc. The parameters evaluated in physicochemical analysis were ash values (total ash, water soluble ash, acid insoluble ash, sulphated ash), extractive values in different solvents and fluorescence analysis. The microscopic studies showed presence of palisade tissue, parenchymatous tissue, xylem, phloem, pointed multicellular trichomes and anomocytic stomata. In both the plants, leaf and stem possessed maximum amount of flavonoids and methanol had maximum extractive value. Fluorescence analysis imparted characteristic colours to the leaf and stem powder under different chemical reagents when observed under visible and UV light. The different salient diagnostic features established in this study will help for proper identification and standardization of the drug in crude form.

Keywords: *Andrographis echioides*, *Tridax procumbens*, macroscopic, microscopic, phytochemical analysis, physicochemical analysis, leaf, stem

Introduction
Medicinal plants or plant-derived products play a valuable and significant role in the treatment of many diseases that occur in humans. There are numerous plants but, it is not easy to know the accurate number of medicinal plants available on earth till date [1]. As per WHO reports, around 35,000-70,000 plants were used across the world to treat various diseases. In the last few decades, pharmaceutical companies have focused on research and development of newly occurring plant-derived drugs [2]. Considering the ever increasing demand of plant drugs it is essential to maintain the quality, reproducibility and efficacy of herbal drugs. Pharmacognostical standardization is an efficient tool to establish quality control parameters of plants. It helps to assure the authentication of plants and prevention of adulteration [3, 4]. Standardization and quality control of plants are also essential for the worldwide approval of herbal products in modern system of medicines. Hence, each country has adopted a set of guidelines and quality control of the herbal medicine [5].

Andrographis echioides (Nees.) belong to the family Acanthaceae. It's vernacular name is Kalukariyatun and common name is false water willow. The plant is a herbaceous in nature, straight, medium-sized annual herb. It is widely distributed in the tropical India and Sri Lanka. The plant is used as a folk medicine like ulcer and hair tonic. The plant shows different chemical constituents like 2',6'-dihydroxyacetophenone 2'-O-β-D-glucopyranoside, echioidinin 5-O-β-D-glucopyranoside, echioidinin, pinostrobin, andrographidine C, dihydroechioidinin, tectochrysin 5-glucoside, methyl salicylate glucoside, 5,7,8-trimethoxyflavone, skullcapflavone I 2'-methyl ether, androechin, skullcapflavone I 2'-O-β-D-glucopyranoside, tectochrysin, 5,7,2'-trimethoxyflavone, echioidin, skullcapflavone, 5,7-dimethoxyflavone, andrographidine [6]. The plant shows different biological activities like anti-ulcer [7], antioxidant [8], anti- inflammatory, analgesic and antipyretic [9], antibacterial [10], anthelmintic [11], etc.

Tridax procumbens (Linn.) belong to the family Compositae (Asteraceae). It's vernacular name is Pardesi bhangro and common name is coat buttons and tridax daisy. The plant is wild,

herbaceous in nature, prostrate, annual herb. It is widely distributed all over India. The plant is used as a folk medicine like bronchial cough, dysentery, diarrhoea, wound healing, etc. The plant shows different chemical constituents like methyl, 14 oxoacagaecunoate, methyl 14-oxononacosanoate, 3-methyl-non dodecylbenzene, heptacosanyl cyclohexane carboxylate, 12-hydroxytetracosa-15-one, β-amyrone, fucosterol and sitosterol, arachidic, behenic, lauric, linoleic, linolenic, myristic, palmitic, stearic acids, etc [12]. The plant shows different biological activities like anticoagulant [13], antidysentery [14], anti-malaria [15], anti-inflammatory [16], antioxidant [17], antimicrobial [18], antidiabetic [19], anticancer [20] and antifungal [21].

Considering the above, in the present study, pharmacognostic, physicochemical and phytochemical analysis of leaf and stem of *Andrographis echioides* and *Tridex procumbens* was done.

Materials and methods

Plant collection

The *Andrographis echioides* Nees. and *Tridex procumbens* Linn. was collected in August, 2016 from Rajkot, Gujarat, India. The plant was compared with voucher specimen of *A. echioides* (Voucher specimen number PSN 573) and *T. procumbens* (Voucher specimen number PSN 414) deposited at Department of Biosciences, Saurashtra University, Rajkot, Gujarat, India. The leaf and stem were separated, washed thoroughly with tap water, shade dried and homogenized to fine powder and stored in closed container for further studies. For physicochemical studies, 5 g of dried powder of leaf and stem was extracted by using solvents of different polarities. The solvent was evaporated to dryness and dried crude extract was weighed and extractive values were determined. Macroscopic and microscopical characters were studied as described in quality control methods [22]. Photographs at different magnifications were taken by using digital camera.

Pharmacognostic studies

Macroscopic studies

Macroscopic studies were carried out by using organoleptic evaluation method. The shape, size, colour, odour, taste, base, texture, margin, apex of leaves and stem of plants were observed and noted down [22].

Microscopic studies

Microscopic studies were carried out by preparing thin sections of leaf and stem. The thin sections were further washed with water, stained with congo red, malachite green and mounted in glycerine for observation and confirm its lignifications (10x, 40x). The powder microscopic studies were also carried out and the specific diagnostic characteristic features were recorded [23].

Powder Microscopy

The powder microscopy of leaf and stem of both the plants was studied using standard procedures [23]. The images of different fragments of tissues was captured and diagnostic characteristic features were recorded.

Qualitative phytochemical analysis

The detection of alkaloids, flavonoids, tannins, phlobatanins, saponins, steroids, cardiac glycosides, triterpenes and anthocyanins were carried out following the procedure of Harborne, (1998) [24]. The details of the procedure followed is as described earlier [25].

Physicochemical analysis

The physicochemical parameters like loss on drying, total ash, acid-insoluble ash, water-soluble ash, sulphated ash and extractive values were determined as per WHO guidelines [26]. in dried powder of leaf and stem of both the plants. The details of the procedure followed is as described earlier [25].

Fluorescence analysis

Fluorescence study of leaf and stem of both the plants powder was performed as per Chase and Pratt, [27]. A small quantity of the powder was placed on a grease free clean microscopic slide and 1-2 drops of freshly prepared reagent solutions were added, mixed by gentle tilting of the slide and waited for a few minutes. Then the slide was placed inside the UV chamber and observed in visible light, short (254 nm) and long (365nm) ultra violet radiations. The colours observed by application of different reagents in different radiations were recorded.

Results

Organoleptic and macroscopic characteristics of *Andrographis echioides*

Organoleptic and macroscopic characteristics of *A. echioides* leaf and stem are given in Table 1 and Fig. 1.

Leaves

The leaf was simple, opposite, decussate, lanceolate, entire margin, acuminate apex, reticulate venation, odour was characteristic and taste was bitter. The average leaf size was 7-8 cm in length and 2-3 cm width (Fig.1 and Table 1).

Stem

The stem were green, woody, erect, square or quadrangular, up to 40 cm height, bearing numerous branches and 2-4 mm thickness, odour was characteristics and taste was bitter. Outer surface was rough and hard (Table 1).

Microscopic characteristics

Leaf

The transverse section of *A. echioides* leaf is shown in Fig. 2. The leaf lamina was dorsiventral in nature. The upper epidermis and lower epidermis was single layered and covered with a single layer of cuticle (Fig. 2a). The palisade tissue was single layered on upper surface and covered with multicellular pointed trichomes. The trichomes were covering type which were multicelled (5-6 celled) and more in number on dorsal side. The basal cells of the trichomes were swollen and tip pointed (Fig. 2b). Starch granules were found in ground tissue. The mesophyll layer was small, only 3-5 celled. The T.S. passing through the mid rib region showed vascular bundles present towards the ventral surface. Some parenchymatous cells were surrounded by centrally located collateral vascular bundles (Fig. 2c). The anomocytic stomata were present in lower epidermis. The stomata were surrounded by small subsidiary wavy cells, whereas the guard cells were comparatively larger in size and each stoma was surrounded by 4–5 subsidiary cells (Fig. 2d).

Stem

The transverse section of *A. echioides* stem is shown in Fig. 3. The epidermis was single layered with cubical cells (Fig. 3a). The epidermis was surrounded by cuticle and many trichomes were present (Fig. 3b). The stem consisted of four bulges at four corners (Fig. 3 c). The cortex consisted of 4-6 layers,

vascular bundles were surrounded by polygonal lignified parenchyma and chollenchyma cells (Fig. 3d). The vascular bundles consistsed of secondary phloem with sieve tubes, companion cells and phloem paranchyma and secondary xylem consisted of lignified trachea, tracheids, few vessels and xylem fibre. Fibers were pitted elongated and moderately thickened (Fig. 3e). Pith was very large and consisted of thick walled sclerenchymatous cells (Fig. 3f).

Powder microscopy of leaf

The crude powder of *A. echioides* leaf was dark green in colour, fine with characteristic odour and slight bitter in taste. The powder microscopy characteristics are shown in Fig.4. The specific characteristics determined from the powder study under microscopic investigation showed single layered lower epidermis, multicellular trichomes, anomocytic stomata, pitted vessels, etc.

Powder microscopy of stem

The crude powder of *A. echioides* stem was green in colour with characteristic odour. The powder characteristics are shown in Fig. 5. The specific characteristics determined from the powder study under microscopic investigation showed different types of xylem like spiral and bordered pitted vessels, annular vessels, sclerenchymatous tissue and uniseriate trichomes, etc.

Organoleptic and macroscopic characteristics of *Tridax procumbens*

Organoleptic and macroscopic characteristics of *T. procumbens* leaf and stem are given in Table 2 and Fig. 6.

Leaves

The leaf was simple, phyllotaxy was opposite decussate, shape was lanceolate to ovate, margin was irregularly toothed, apex was acute, leaf base was symmetrical, appearance rough and scabrous, reticulate venation, odour was characteristic and taste was acrid. The average leaf size was 7 - 9.5 cm in length and 3.5 - 4 cm in width (Fig. 6).

Stem

The stem was green, woody, erect, and cylindrical, up to 40 cm in height bearing numerous branches and 3-6 mm thickness, odour was characteristic and taste was acrid. Outer surface was rough and soft (Table 2).

Microscopic characteristics

Petiole

The transverse section of *T. procumbens* is shown in Fig. 7. The petiole was kidney shaped towards the distal end (petiole) and crescent shaped towards the laminal side. The single layered upper and lower epidermis was covered with unicellular and multicellular, 2-3 celled trichomes (Fig. 7a). The hypodermis was 1-2 celled with collenchymatous tissue. The ground tissue was parenchymatous, vascular bundles were 'arc' shaped and three in number, the size of the vascular bundles varied from centre to leaf margin i.e. large too small. They were centripetally arranged i.e. xylem surrounded by the phloem (Fig. 7b).

Leaf

The transverse section of *T. procumbens* leaf is shown in Fig.7. The leaf lamina was dorsiventral in nature. The upper epidermis and lower epidermis were single layered. The palisade tissue was single layered on both the surfaces and was covered with thick cuticle, the mesophyll was small, 3-5 layered. The T.S. passing through the mid rib region showed vascular bundles towards the ventral surface. The trichomes were simple, multicellular (2-3 celled) with covering type and were more in number on dorsal side (Fig. 7c). The prismatic crystals of calcium oxalate were found in ground tissue. Centrally located conjoint collateral vascular bundles were surrounded by parenchymatous cells filled with dark content (Fig. 7d). The basal cells of the trichomes were swollen and tip pointed. The anomocytic stomata were present in lower epidermis (Fig. 7e). The stomata were surrounded by small subsidiary wavy cells, whereas the guard cells were comparatively larger in size and each stoma was surrounded by 4–5 subsidiary cells and hence the type of stomata is anomocytic or ranunculaceous (Fig. 7f).

Stem

The transverse section of *T. procumbens* stem is shown in Fig. 8. The epidermis was single layered, thick walled, narrow and small, and was surrounded by trichomes (Fig. 8a). 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 (Fig. 8b). The vascular bundles were conjoint, collateral, close and arranged in a ring form. The xylem consisted of protoxylem, xylem vessels, tracheids, and xylem fibre (Fig. 8c). The pith was very large, with thick and same size of hexagonal parenchymatous cells (Fig. 8d). The longitudinal section of stem middle layer consisted of 4-5 layers of rectangular parenchymatous cells (Fig. 8e).

Powder microscopy of leaf

The crude powder of *T. procumbens* leaf was dark green in colour, fine, odourless and slight bitter in taste. The powder microscopy characteristics are shown in Fig. 9. The specific characteristics determined from the powder study under microscopic investigation showed bordered pitted vessels, unicellular trichomes, rosette crystal calcium oxalate, anomocytic stomata, spongy parenchymatous cells, etc.

Powder microscopy of stem

The crude powder of *T. procumbens* stem was green in colour, odour was characteristic and taste was bitter. The powder characteristics are shown in Fig. 10. The specific characteristics determined from the powder study under microscopic investigation showed scalariform vessels, annular vessels, bordered pitted vessels, phloem tissue, spiral vessels, square crystals of calcium oxalate, unicellular trichomes, multicellular trichomes and glandular trichomes, etc.

Physicochemical analysis

The physical constant investigation of the drugs is an important parameter in detecting adulteration or improper handling of drugs. The physicochemical parameters of *A. echioides and T. procumbens* leaf and stem is given in Table 3. The moisture content of dry powder of *A. echioides* leaf and stem was 8.00% and 8.5% while that of *T. procumbens* was 10.50% and 10.75% respectively. The moisture content is quite and low and itt would discourage the growth of bacteria, fungi or yeast. The total ash in leaf was 21.91% in *A. echioides* and 20.45% in *T. procumbens*, while water soluble ash and acid insoluble ash was 5.5% and 3.6% in *A. echioides*

and 6.50% and 7.16% in *T. procumbens* respectively. The total ash in stem was 13.33% in *A. echioides* and 14.75% in *T. procumbens*, while water soluble ash and acid insoluble ash was 0.21% and 0.16% in *A. echioides* and 7.50% and 6.00% in *T. procumbens* respectively, the sulphated ash of leaf and stem was 29% and 16.16% in *A. echioides* and 13.78% and 16.83% in *T. procumbens* respectively. The extractive value of leaf and stem is given in Table 3. In both the plants and both the parts, maximum extractive value was in methanol and minimum in petroleum ether extracts. The methanol soluble extractive values of in *A. echioides* leaf and stem were more than that of *T. procumbens* leaf and stem; the water soluble extractive values of leaf and stem extracts of *A. echioides* were also more than that of *T. procumbens* leaf and stem (Table 3).

Qualitative phytochemical analysis

The qualitative phytochemical screening of the crude powder of *A. echioides* leaf and stem and *T. procumbens* leaf and stem are given in Table 4. In both leaf and stem of both the plants flavonoids were present in maximum amount followed by triterpenes. Other phytoconstituent were present in trace amount; phlobatanins and anthocyanins were absent (Table 4).

Fluorescence analysis

Fluorescence study of leaf and stem of *A. echioides* and *T. procumbens* was attempted. The crude powder was treated with various reagents and they revealed characteristic fluorescence at 366 nm and 254 nm wavelengths (Tables 5 - 8). The different colours observed in *A. echioides* leaf and stem were green, dark green, light green, yellowish green, brown, yellowish brown and black while in *T. procumbens* leaf and stem, the colours observed were green, dark green, light green, brownish green, dark brown, blackish green, black, and yellowish black.

Table 1: Organoleptic features of *Andrographis echioides* (Nees.)

	Observation	**Observation**
Part	**Leaves**	**Stem**
Arrangement	Opposite	-
Size	7-8 cm long, 2-3 cm wide	2 to 4 mm thickness,40 cm height
Shape	Lanceolate to ovate	Square or quadrangular
Colour	Green	Green
Odour	Characteristics	Characteristics
Taste	Bitter	Bitter
Appearance	Scabrous	Rough and hard
Margin	Entire	-
Apex	Acuminate	-
Base	Symmetrical	-
Petiole	Short	-
Texture	Short	-
Veination	Reticulate veination	-
Outer surface	-	Light green colour. Rough surface

Table 2: Organoleptic features of *Tridax procumbens* L.

	Observation	**Observation**
Part	Leaves	Stem
Arrangement	Opposite	-
Size	7-9.5 cm long, 3-4.5 cm wide	3-6 mm thickness,40 cm long
Shape	Lanceolate to ovate	Cylindrical
Colour	Green	Green
Odour	Characteristic	Characteristic
Taste	Acrid	Acrid
Appearance	Rough & Scabrous	Rough & Scabrous
Margin	Irregularly toothed	-
Apex	Acute	-
Base	Symmetrical	-
Petiole	Short	-
Texture	Short	-
Veination	Reticulate veination	-
Outer surface	-	Light green colour. Rough surface

Table 3: Physiochemical parameter of *A. echioides* and *T. procumbens* leaf and stem

Sr No	**Parameters**	**% value(w/w) Leaf of *A. echioides***	**% value(w/w) Stem of *A. echioides***	**% value(w/w) Leaf of *T. procumbens***	**% value(w/w) Stem of *T. procumbens***
1	Loss on drying	8.00	8.5	10.50	10.75
2	Total ash	21.91	13.33	20.45	14.75
3	Water soluble ash	5.5	0.21	6.50	7.50
4	Acid insoluble ash	3.6	0.16	7.16	6.00
5	Sulphated ash	29	16.16	13.78	16.83
6	Petroleum ether soluble extractive value	1.06	0.89	0.78	0.42
7	Toluene soluble extractive value	2.28	1.53	0.41	0.66
8	Ethyl acetate soluble extractive value	2.24	2.38	0.50	0.54
9	Methanol soluble extractive value	15.60	11.82	7.33	6.81
10	Water soluble extractive value	23.19	19.43	17.20	10.48

Table 4: Qualitative phytochemical analysis of *A. echioides* and *T. procumbens* leaf and stem

Sr No.	Phytochemicals	Leaf of *A. echioides*	Stem of *A. echioides*	Leaf of *T. procumbens*	Stem of *T. procumbens*
1	Alkaloids				
	(1) Mayer's reagent	++	+	++	++
	(2) Dragondroff's reagent	-	-	-	-
	(3) Wagner's reagent	+	+	++	++
2	Flavonoids	+++	+++	+++	+++
3	Tannins	+	+	+	+
4	Phlobatanins	-	-	+	+
5	Saponins	+	+	+	+
6	Steroids	++	++	+	+
7	Cardiac glycosides	+	+	+	+
8	Triterpenes	++	++	++	++
9	Anthocyanins	-	-	-	-

Note – (+++) more amount, (++) moderate amount, (+) less amount, (-) absent

Table 5: Fluorescence analysis of *A. echioides* leaf powder

Sr No.	Treatment	Visible light	Under UV light Short Wave length (254nm)	Under UV light Long Wave length (365nm)
1	1N NaOH (aq)	Brown	Black	Dark green
2	1N NaOH (alco)	Light green	green	Yellow green
3	Ammonia	Green	Black	Dark green
4	Picric acid	Brown	Green black	Black
5	Petroleum ether	Light green	Dark green	Light green
6	50% HCL	Dark brown	Black	Black
7	50%H_2SO_4	Brown	Black	Dark green
8	Ethyl acetate	Yellow green	Dark green	Light green
9	Ethyl alcohol	Yellow green	Dark green	Light green
10	Methanol	Green	Dark green	Light green
11	50% KOH	Dark green	Black	Yellowish brown
12	50%HNO_3	Brown	Black	Dark green
13	Acetic acid	Yellowish green	Green	Light green
14	Iodine in water (1%)	Green	Black	Brown green
15	$FeCl_3$	Back	Black	Black

Table 6: Fluorescence analysis of *A. echioides* stem powder

Sr No.	Treatment	Visible light	Under UV light Short Wave length (254nm)	Under UV light Long Wave length (365nm)
1	1N NaOH (aq)	Dark green	Black	Dark green
2	1N NaOH (alco)	Dark green	Black	Brown
3	Ammonia	Dark green	Black	Dark green
4	Picric acid	Brown	Black	Dark black
5	Petroleum ether	Green	Dark green	Green
6	50% HCL	Black	Black	Black
7	50%H_2SO_4	Dark green	Dark green	Green
8	Ethyl acetate	Dark green	Black	Green
9	Ethyl alcohol	Dark green	Dark green	Green
10	Methanol	Dark green	Black	Brown
11	50% KOH	Dark green	Dark green	Green
12	50%HNO_3	Brown	Black	Black
13	Acetic acid	Blackish green	Green	Light green
14	Iodine in water (1%)	Dark green	Black	Brownish green
15	$FeCl_3$	Black	Black	Black

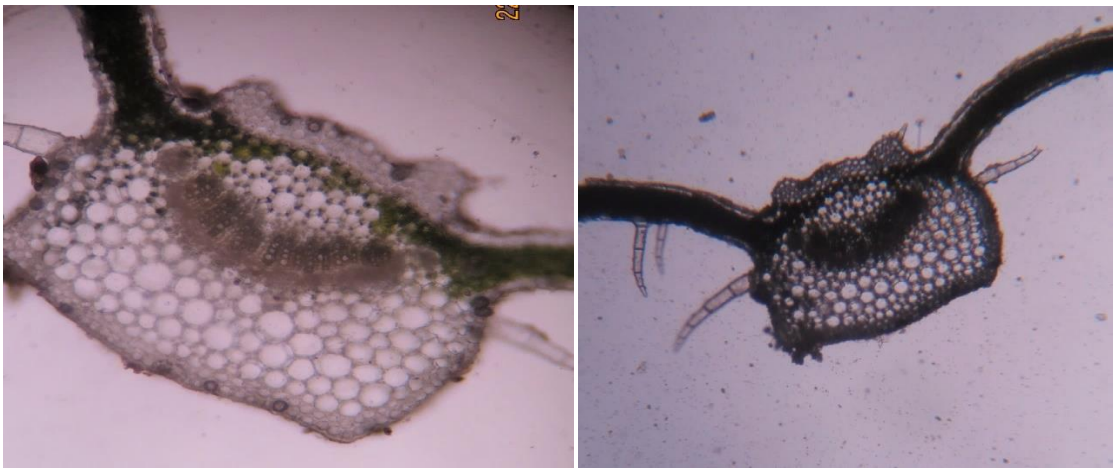
Table 7: Fluorescence analysis of *T. procumbens* leaf powder

Sr No.	Treatment	Visible light	Under UV light Short Wave length (254nm)	Under UV light Long Wave length (365nm)
1	1N NaOH (aq)	Dark green	Black	Dark green
2	1N NaOH (alco)	Dark green	Black	Light green
3	Ammonia	Dark green	Black	Dark green
4	Picric acid	Brownish black	Black	Dark brown
5	Petroleum ether	Dark green	Black	Light green
6	50% HCL	Blackish brown	Black	Black
7	50%H_2SO_4	Blackish brown	Black	Black
8	Ethyl acetate	Dark green	Black	Dark green
9	Ethyl alcohol	Dark green	Black	Dark green
10	Methanol	Dark green	Black	Blackish green
11	50% KOH	Dark green	Black	Brownish green
12	50%HNO_3	Dark brown	Black	Black
13	Acetic acid	Blackish green	Black	Dark green
14	Iodine in water (1%)	Dark green	Black	Dark green
15	$FeCl_3$	Yellowish dark black	Black	Dark black

Table 8: Fluorescence analysis of *T. procumbens* stem powder

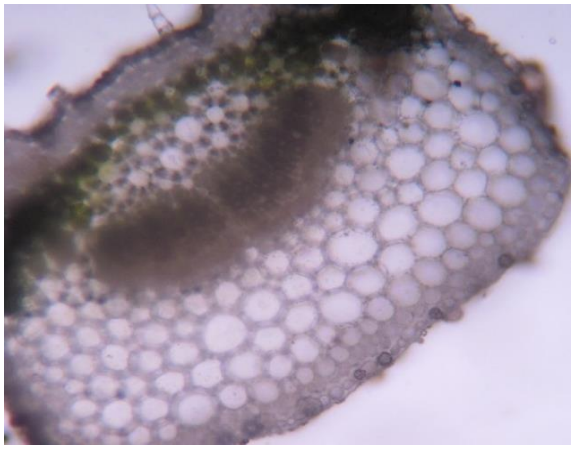
Sr No.	Treatment	Visible light	Under UV light Short Wave length (254nm)	Under UV light Long Wave length (365nm)
1	1N NaOH (aq)	Greenish brown	Black	Dark green
2	1N NaOH (alco)	Green	Black	Light green
3	Ammonia	Green	Black	Dark green
4	Picric acid	Brown	Black	Dark brown
5	Petroleum ether	Green	Black	Light green
6	50% HCL	Brown	Black	Black
7	50%H_2SO_4	Light brown	Black	Black
8	Ethyl acetate	Light green	Black	Light green
9	Ethyl alcohol	Green	Black	Green
10	Methanol	Green	Black	Green
11	50% KOH	Brownish green	Black	Brownish green
12	50%HNO_3	Brown	Black	Black
13	Acetic acid	Blackish green	Green	Light green
14	Iodine in water (1%)	Dark green	Black	Brownish green
15	$FeCl_3$	Black	Black	Black

Fig 1: Macroscopic characteristics of *Andrographis echioides*

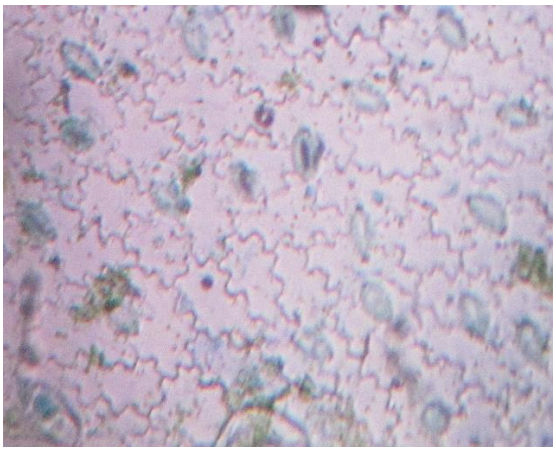


a) T.S of leaf mid rib

b) T.S of entire leaf with epidermis

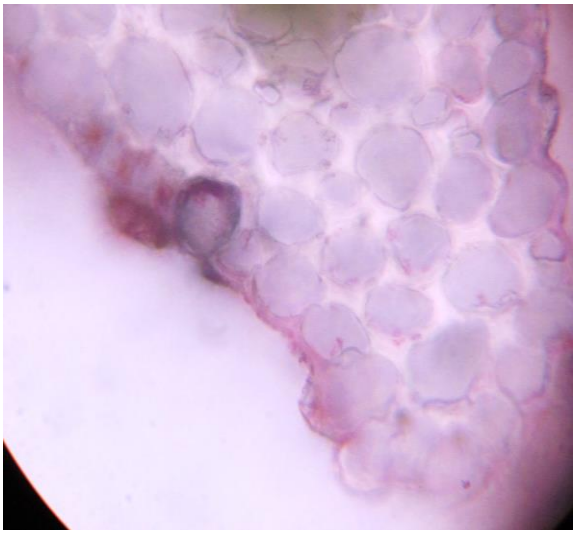


c) Lower epidermis, sclerenchyma and starch granules

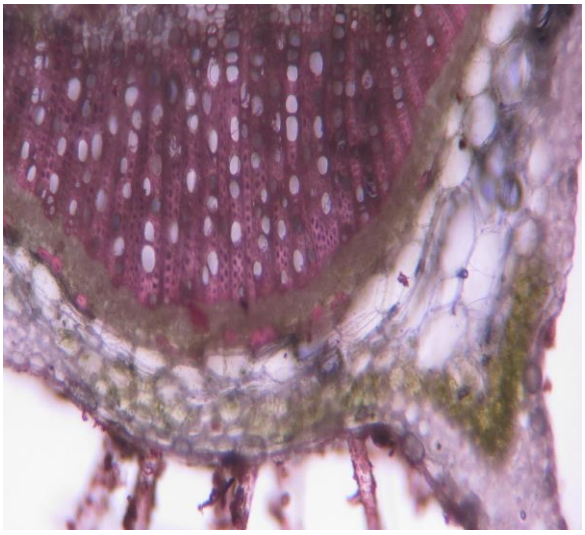


d) Anomocytic stomata

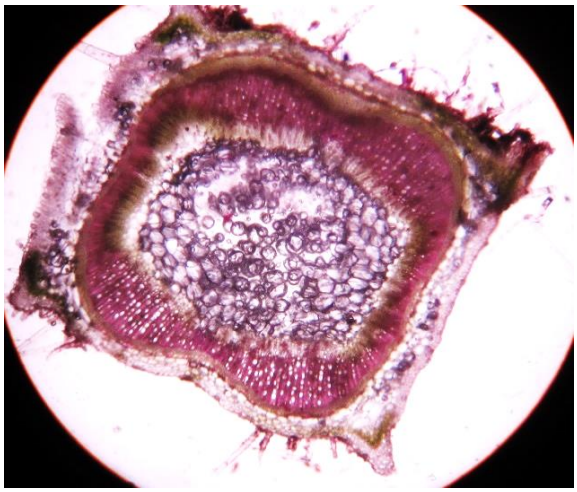
Fig 2: Microscopic characteristics of *A. echioides* leaf



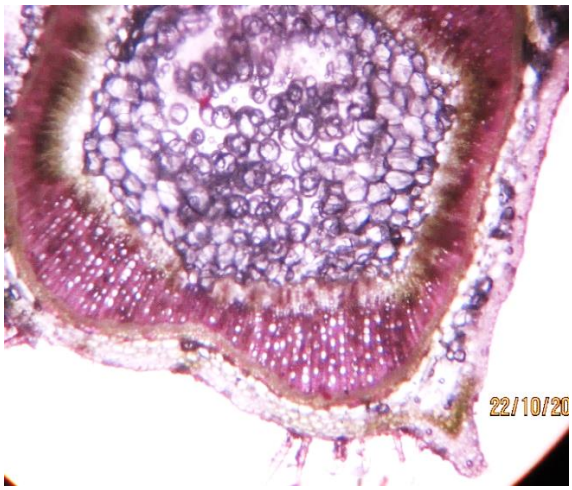
a) Epidermis with cubical cell



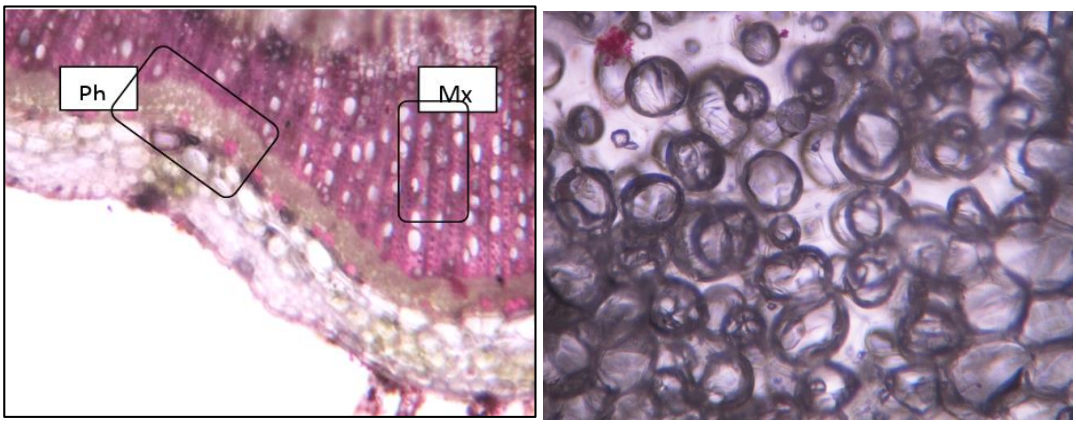
b) Vascular bundle and cortex



c) T.S of stem with trichomes

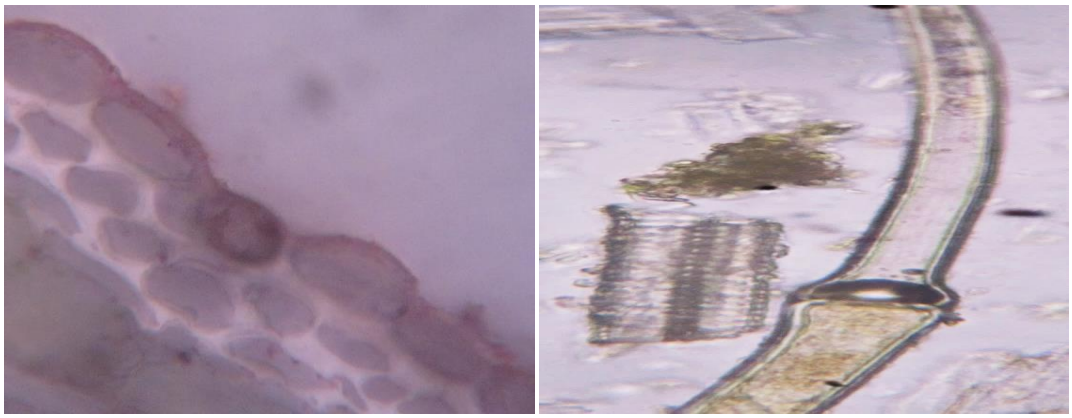


d) T.S of stem with entire pith

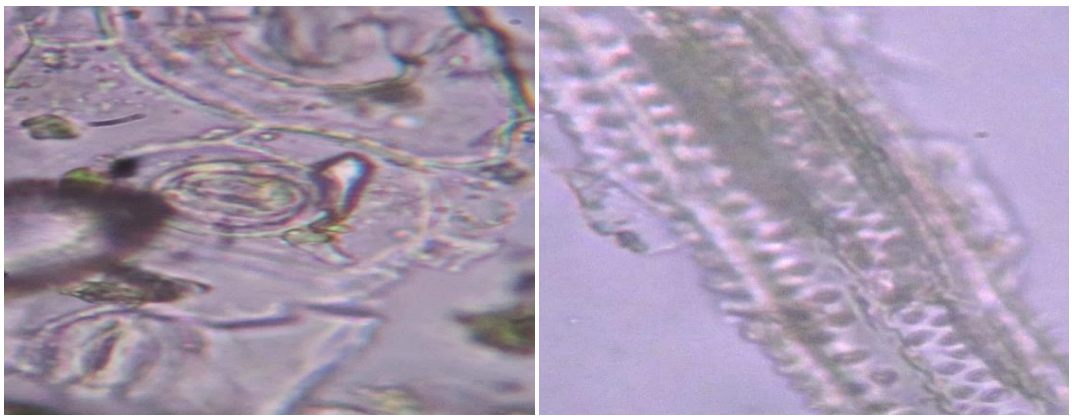


e) Meta xylem and phloem **f)** Pith

Fig 3: Microscopic characteristics of *A. echioides* stem

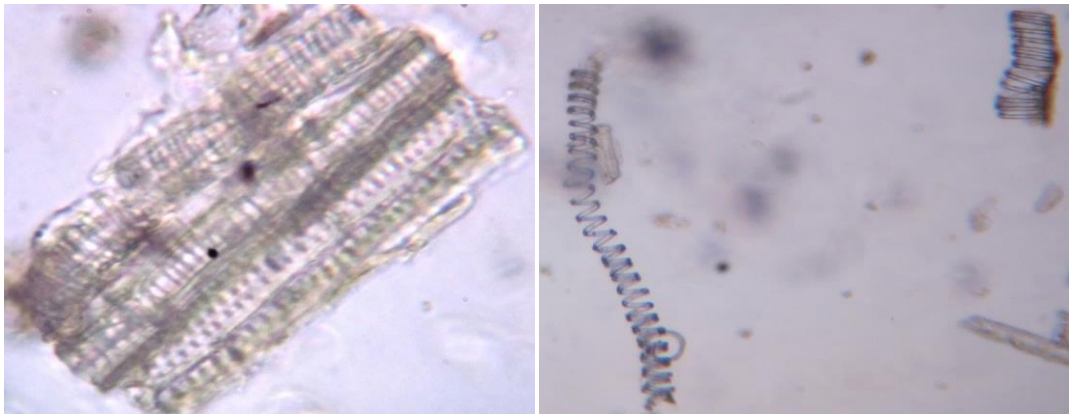


a) Lower single layer epidermis **b)** Multicellular trichome

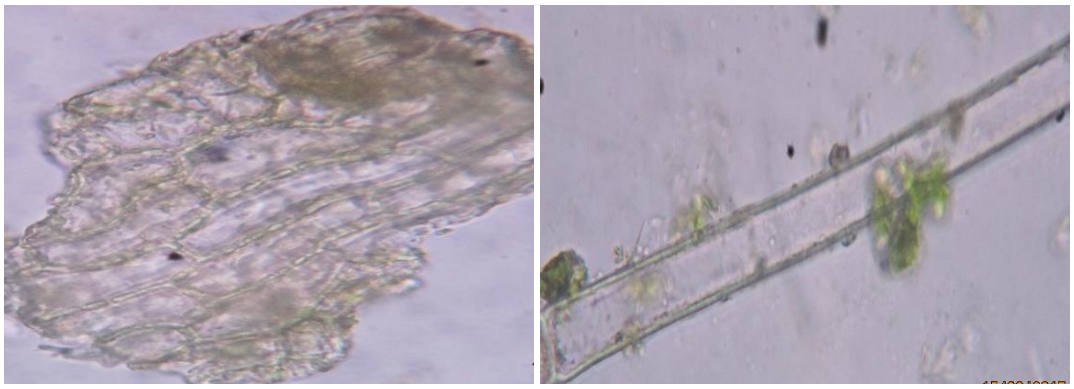


c) Anomocytic Stomata **d)** Pitted vessels

Fig 4: Microscopic powder study of *A. echioides* leaf



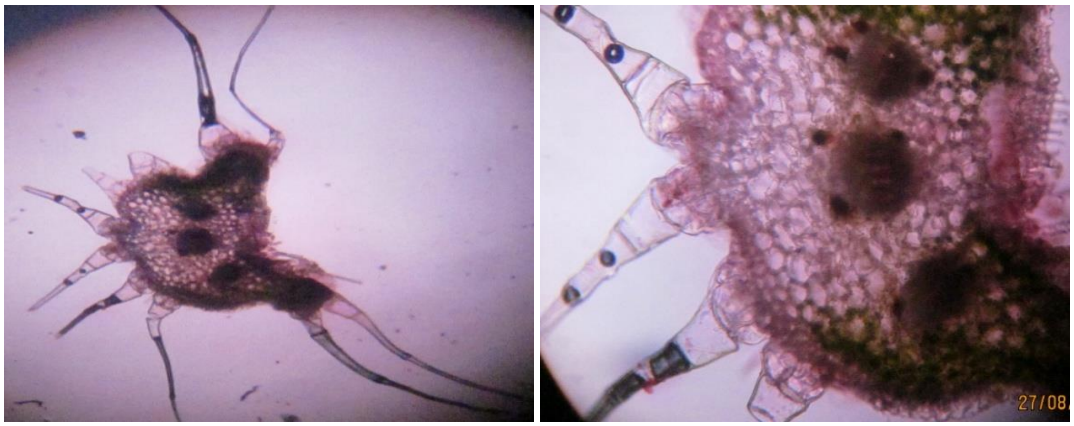
a) Spiral and bordered pitted vessels **b)** Annular and spiral vessels



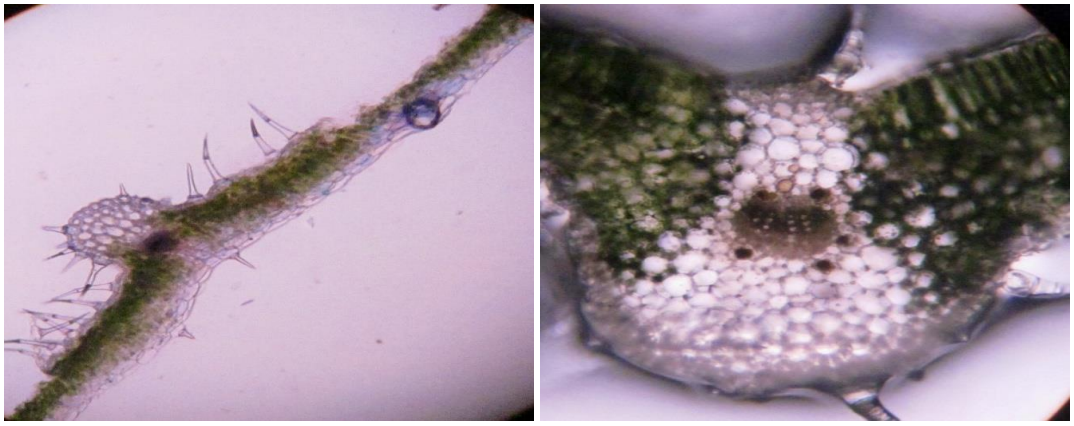
c) Sclerenchymatous fibres **d)** Uniseriate trichome

Fig 5: Microscopic powder study of *A. echioides* stem

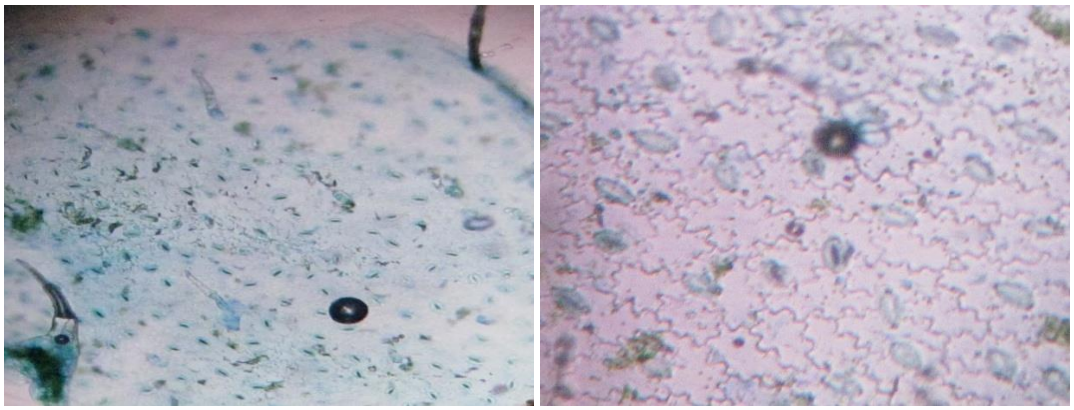
Fig 6: Macroscopic characterstics of *T. procumbens*



a) T.S of petiole with trichomes **b)** T.S of petiole vascular bundles

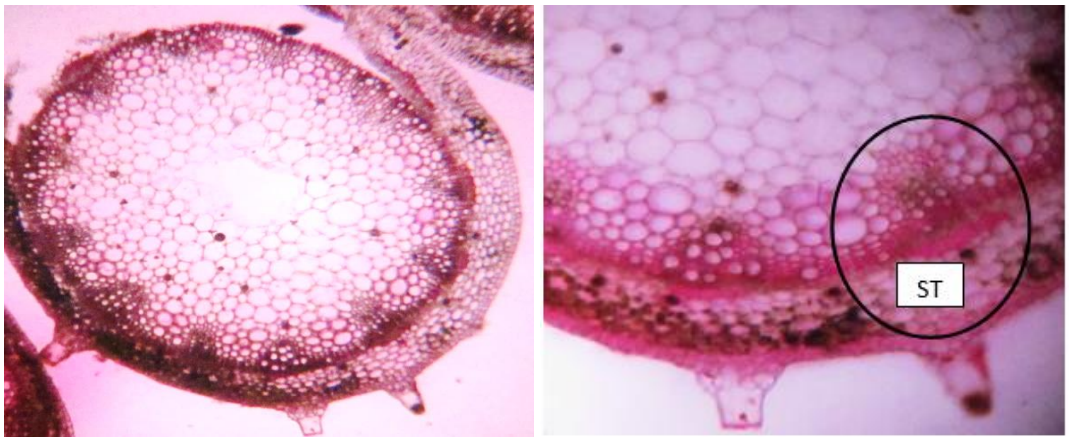


c) T.S of leaf passing through midrib **d)** T.S of leaf passing vascular bundles

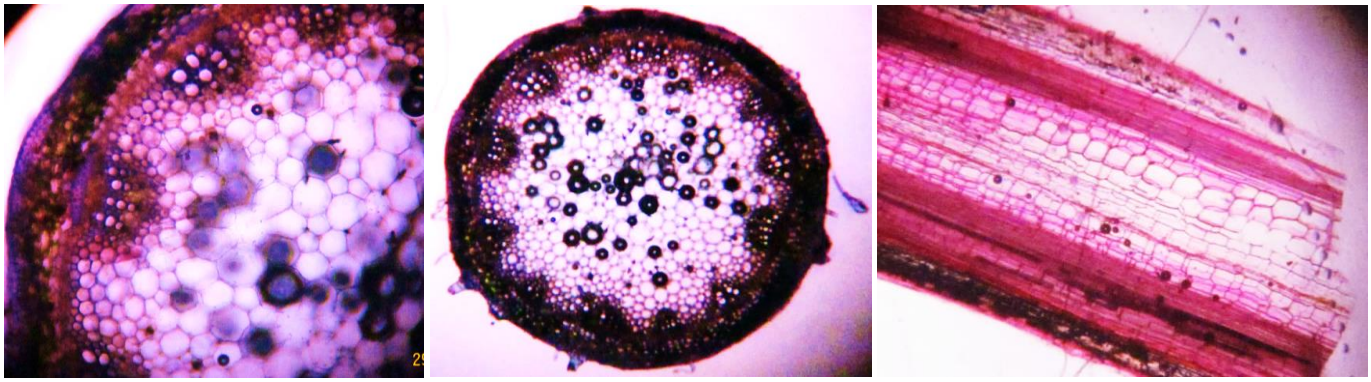


e) Multicellular trichomes and stomata **f)** Anomocytic stomata

Fig 7: Microscopic characterstics of *T. procumbens* leaf

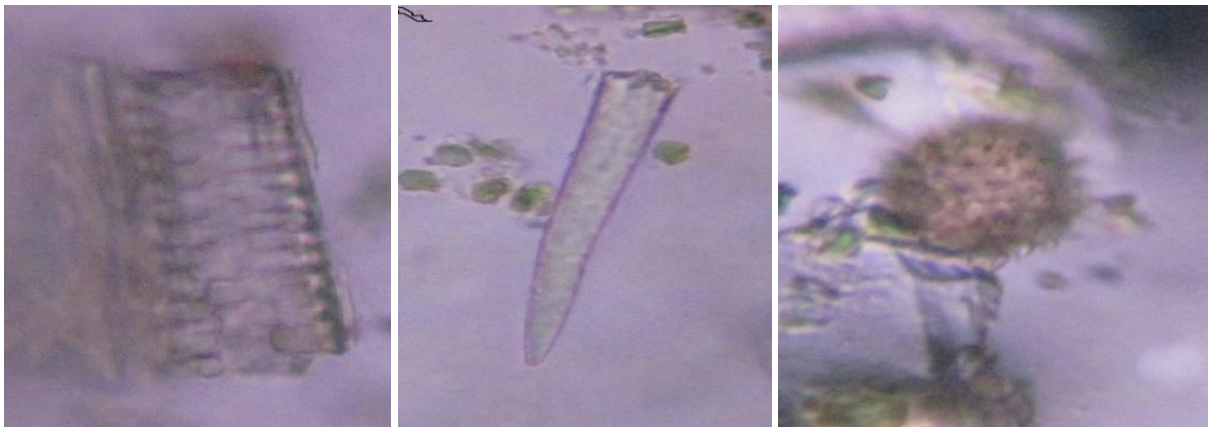


a) T.S of stem with epidermis and trichomes **b)** T.S of stem with cortex, cambium tissue

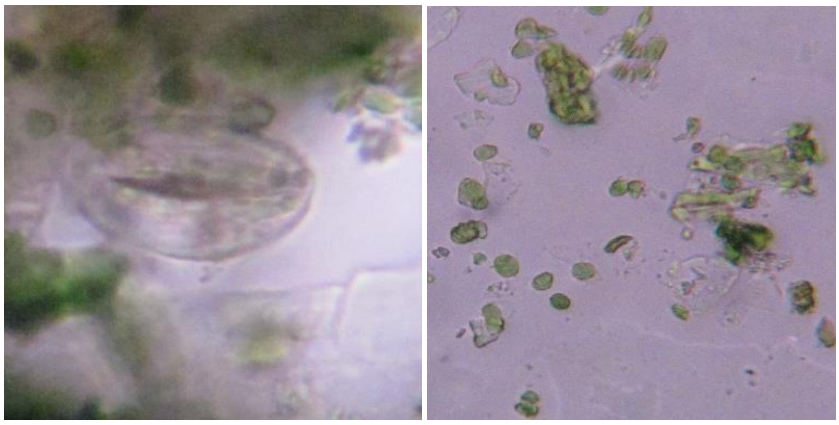


c) T.S.of stem with vascular bundles **d)** T.S of stem with entire pith **e)** L.S of stem with parenchymatous cells

Fig 8: Microscopic characterstics of *T. procumbens* stem

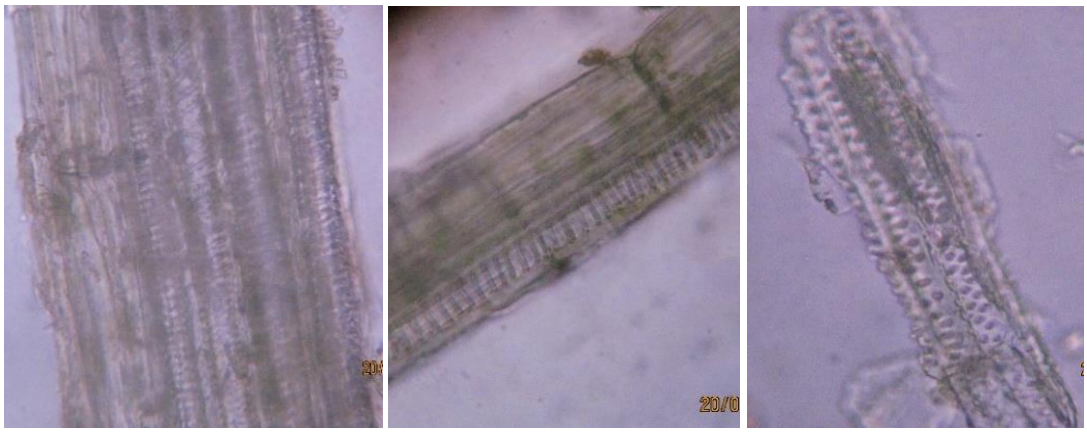


a) Bordered pitted vessels **b)** Unicellular trichome **c)** Rosette calcium crystal

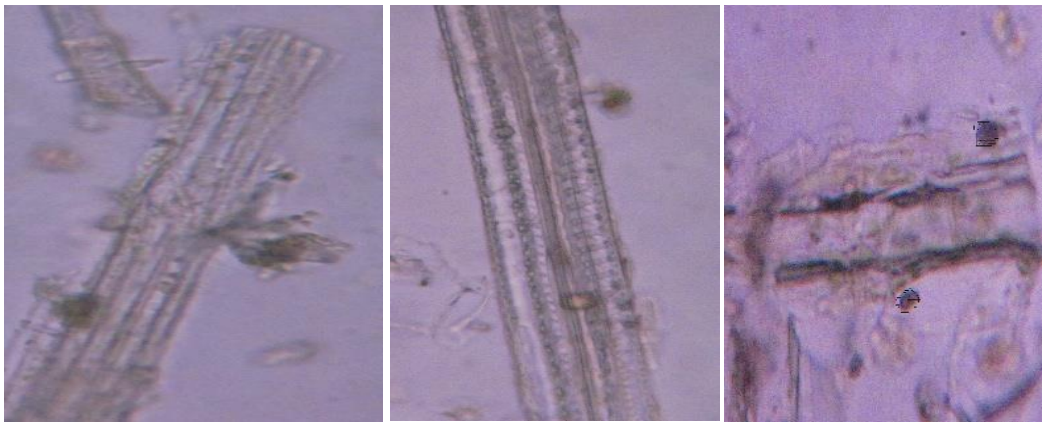


d) Anomocytic stomata **e)** Spongy parenchyma

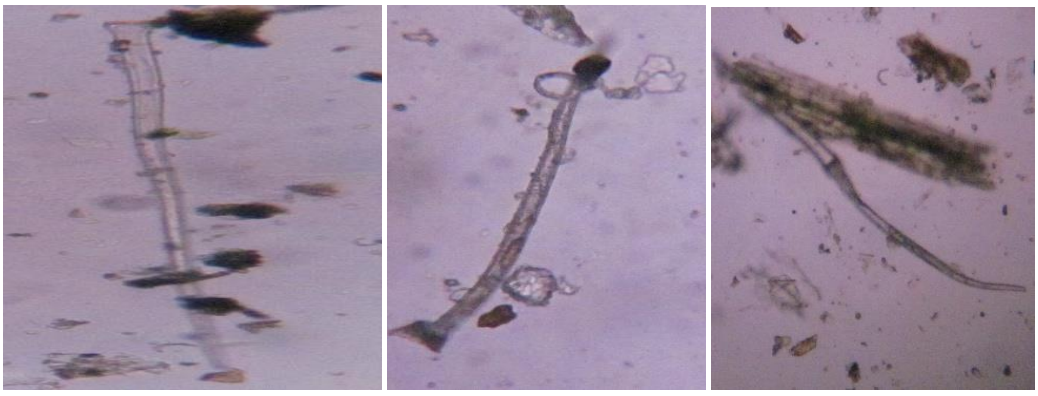
Fig 9: Microscopic powder study of *T. procumbens* leaf



a) Scalariform vessels **b)** Annular vessels **c)** Bordered pitted vessels



d) Phloem tissue **e)** Spiral vessels **f)** Square crystal of calcium



g) Unicellular trichome **h)** Glandular trichome **i)** Multicellular trichome

Fig 10: Microscopic powder study of *T. procumbens* stem

Discussion

Morphological and microscopic characteristics play a vital role in plant systemic study and are used as a tool for the classification of a taxon [28]. The morphological and microscopic features obtained in this study are more or less similar in all the plant samples with slight variation in number of layers of cell in different regions, number of pericyclic fibres, and microscopic measurements of various cells and tissues. Similar studies are reported in our earlier studies [29-31].

The macroscopic features of *A. echioides* leaf showed simple, opposite, decussate, lanceolate, entire margin, acuminate apex and stem showed green, woody, erect, square or quadrangular, upto 40 cm height. The microscopic features of *A. echioides* leaf showed epidermis, multicellular pointed trichomes, starch granules, mesophyll tissue, collateral vascular bundles, anomocytic stomata etc, and stem showed epidermis, four bulges at the four corners, cortex consisted of 4-6 layers, polygonal lignified parenchyma and collenchyma cells, vascular bundles with secondary phloem, sieve tubes, companion cells and phloem parenchyma, secondary xylem with lignified trachea, tracheids, few vessels and xylem fibre.

The macroscopic features of *T. procumbens* leaf showed simple, phyllotaxy opposite decussate, shape lanceolate to ovate, margin irregularly toothed, apex acute, leaf base symmetrical and stem showed green, woody, erect, green, woody, erect, and cylindrical, upto 40 cm in height. The microscopic features of *T. procumbens* leaf showed epidermis, multicellular pointed trichomes, mesophyll tissue, prismatic crystals of calcium oxalate conjoint collateral vascular bundle and anomocytic stomata and the stem showed epidermis, cortex consisted of 2-3 layers, polygonal lignified parenchyma cells, cambium cell, vascular bundles were conjoint, collateral, close, in ring, xylem with protoxylem, xylem vessels, tracheids, and xylem fibre, pith was very large.

Different physicochemical parameters like ash values, extractive values, moisture content etc could help for botanical identification, quality control as well as the therapeutic potential of plant materials. The physicochemical properties help to estimate the amount of impurities like soil and particle present in the drug. It also helps to assess the calculi salts present in the drug sample. Ash values are used to detect the presence of any siliceous contamination and presence of any water soluble salts [32, 33].

Fluorescence is the occurrence of various chemical constituents present in the plant crude powder. Some show fluorescence in the visible range in daylight. The ultraviolet light produces fluorescence in many natural products (e.g. alkaloids like epiberberine, berberine) which do not visibly fluoresce in daylight. Some of the substances may be often converted into fluorescent derivatives by using different chemical reagents though they are not fluorescent, hence we can often assess qualitatively some crude drugs using fluorescence as it is the most important parameter of pharmacognostical evaluation [34].Therefore, fluorescence analysis can be used for the identification of the drug and adulteration can also be determined [27, 35].

Preliminary phytochemical analysis confirms different group of phytoconstituents present in plants like alkaloids, flavonoids, phenols, tannins, saponins, cardiac glycosides, steroids, triterpenes, etc. In both the plants, flavonoids were present in maximum amount in leaf and stem followed by triterpenes. Hence it is suggested that these plants can be used as natural source of antioxidant activity. The therapeutic potential of a medicinal plant is mainly due to the active ingredients present in it. These standardized parameters would certainly help for selection of the right sample of plant material for its phytopharmacological studies.

Conclusion

Herbal medicines are promising choice over modern synthetic drugs because they show minimum or no side effects and are considered to be safe. Generally herbal formulations involve use of fresh or dried plant parts. Correct knowledge of such crude drugs is very important aspect in preparation, safety and efficacy of the herbal product. Pharmacognosy is a simple and reliable tool, by which complete information of the crude drug can be obtained. The major difficulty in the mixture of herbal medicine into modern medical practices is the lack of scientific and clinical data and better understanding of efficacy and safety of herbal products. To ensure the quality and safety of herbal products, standardization is of vital importance. In the present study pharmacognostic, physicochemical and phytochemical analysis of leaf and stem of *A. echioides* and *T. procumbens* was attempted. The parameters evaluated are diagnostic characters of the plants and they can be used as reference material for the preparation of a monograph. They will also be useful in maintaining the authenticity, identity and efficacy of the plants *A. echioides* and *T. procumbens*.

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