MACRO - AND MICROMORPHOLOGY OF THE LEAF, STEM AND ROOT OF AMARANTHUS CHLOROSTACHYS WILLD

Jaha M. Sarg, Ehsan M. Abd Al – Aziz, Salem A. Salem and Rawia A. Zayed Department of Pharmacognosy, Faculty of Pharmacy, University of Zagazig, Egypt

ABSTRACT

A study of the macro - and micromorphology of the leaf, stem and root of Amaranthus chlorostachys willd is presented with the aim of finding the diagnostic characters of these organs by which the plant could be easily identified both in the entire and powdered forms.

INTRODUCTION

Amaranthus chlorostachys willd, Family Amaranthaceae is a herbaceous plant growing wild in Nile-Delta of Egypt ⁽¹⁾. Some plants of Amaranthus species are used in folk medicine as diuretic, antirheumatic, strengthen the liver, control haemorrhage caused by abortion and externally used as an ointment for boils and itches ⁽²⁾.

Phytochemical study of the plant led to the isolation and characterisation of 13 compounds; lupeol , lupeol acetate , α - spinasterol, and two esters from the light petroleum extract. In addition, α - spinasterol glucoside and β -sitosterol glucoside from the chloroformic extract. Quercetin and rutin also have been isolated from the ethyl acetate extract . Column chromatography of the aqueous extract led to the isolation and identification of two - spinasterol saponin glycosides, choline and some other basic nitrogenous substances $^{(3)}$.

Nothing was reported in literature concerning the macro - and micromorphology of the plant except few data given by some floras (1,4).

In this study, the macro - and micromorphology of the leaf, stem and root of the plant are presented with the aim of finding out the diagnostic characters by which the plant could be identified both in the entire and powdered forms.

EXPERIMENTAL

First Material: Fresh samples of Amaranthus chlorostachys Willd, were collected from flowering plants growing wild in a region 20-km west of Zagazig during July 1989 and July 1990. Authenticity of the plant was kindly verified by Prof. Dr. Loutfy Boulos, National Research Center, Cairo, Egypt.

A- Macromorphology:

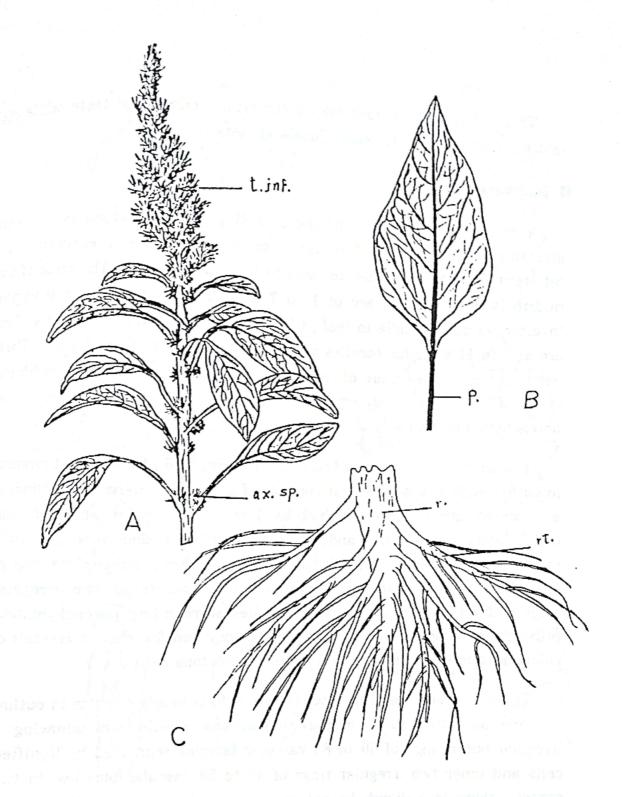
Amaranthus chlorostachys willd (Fig. I,A) is a herbaceous plant with alternate , lanceolate green leaves , showing cylindrical stems which carry numerous , terminal dense spikes and few small axillary ones including intermixed male and female flowers . The plant measures 2 to 3 m in height and flowering from July to september .

The leaves (Fig . Lb)) are simple , petiolate , exstipulate , alternate "phyllotaxis 2/5", green and hairy with pinnately reticulate venation .The lamina is oval - lanceolate , with entire margin, symmetric base , acute to subacute apex , green surface and long petiole .It measures 3 to 10 cm L , and 1.5 to 6 cm B.

The petiole (Fig. I,B) is concave on upperside with two lateral ridges, convex on lower one , green , hairy and measures 2 to 7 cm L, and 0.5 to 1.5mm D_{\odot}

The stem (Fig. I,A) is erect , herbaceous, almost cylindrical and monopodially branched. It is solid with hairy pink to green surface. The internodes are 5 to 12 cm L and $\,0.5$ to $\,1.5$ cm D, the fresh stem is flexible while the dry breaks with a short fracture in the outer part , fibrous in the inner .

The root (Fig . I,C) is a fusiform tap root , bearing many lateral roots and rootlets .It measures 12 to 35 cm L and 0.5 to 3 cm D. Externally, it is fibrous fracture and a yellowish - white interior.



(Fig. 1) A- Sketch of Flowering top of Amaranthus chlorostachys Willd.

C- The root.

B- The leaf.

ax.sp., axilary spike; P., petiole; r., root; rt., rootlet.

t.Inf., terminal inflorescence.

(A x0.8, B x0.8, C x0.4)

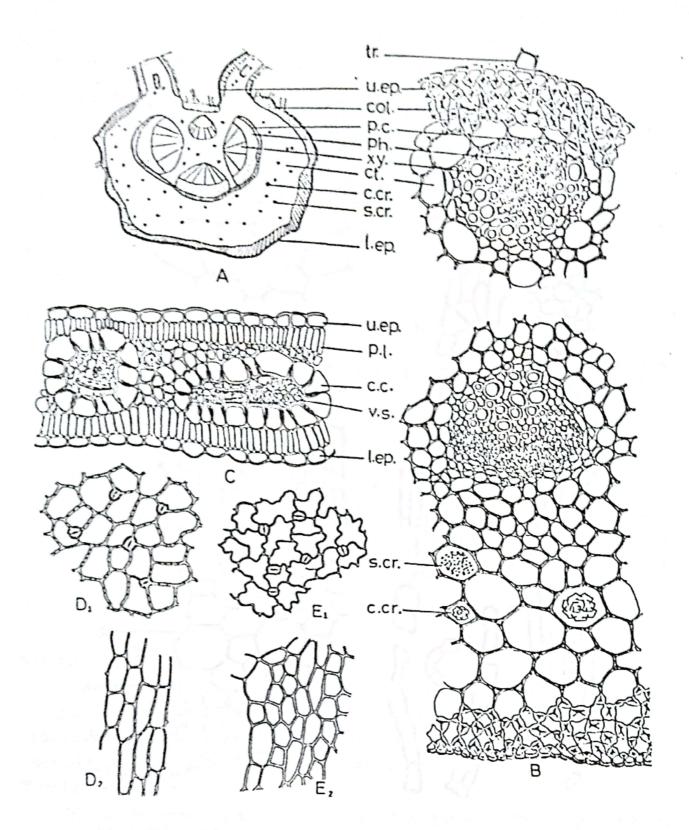
The leaf and stem have no characteristic odour and taste while the root has a faint characteristic odour and taste .

B- Micromorphology:

The transverse section of the leaf (Fig. 2,B) shows an isobilateral structure with one row of palisade cells beneath both epidermises, interrupted by collenchymatous cells in the midrib region. The stele of the midrib is formed of an arc of 1 to 7 vascular bundles with one upper inverted vascular bundle in leaf, while that of the petiole, consists of an arc of 4 to 11 vascular bundles with vascular bundle in each ridge. Each vascular bundle has an arc of collenchymatous pericycle below the phloem region. Cluster and sandy crystals of calcium oxalate are present in the parenchymatous cells.

The transverse section of the stem (Fig. 5, B, 6, B) is almost circular in outline with 4 to 8 ridges. It consists of an outer epidermis surrounding a parenchymatous cortex with 4 to 6 rows of angular subepidermal collenchyma in the ridges and well differentiated endodermis. The stele exhibits anomalous secondary thickening (5); shows a complete ring of irregular 13 to 44 vascular bundles and inner scattered two irregular rings including 23 to 52 vascular bundles surrounding parenchymatous pith with a hollow core in the old stem. Sandy and few cluster crystals of calcium oxalate are present in the parenchymatous cells.

Transverse section of the root (Fig. 8 & 9) is nearly circular in outline showing an outer cork, a parenchymatous phelloderm showing 3 irregular outer rings of 60 to 85 vascular bundles separated by lignified cells and inner two irregular rings of 40 to 58 vascular bundles. In the center , there is a diarch lry xylem surrounded by a narrow secondary vascular tissue of an outer phloem and inner xylem , showing two wide parenchymatous primary medullary rays .The young root (Fig. 8,B) shows a piliferous layer followed by a parenchymatous phelloderm, an endodermis and a parenchymatous pericycle enclosing diarch radial vascular bundles.



(Fig. 2) A- Diagram of T.S. in B- Detailed T.S. of the midrib.

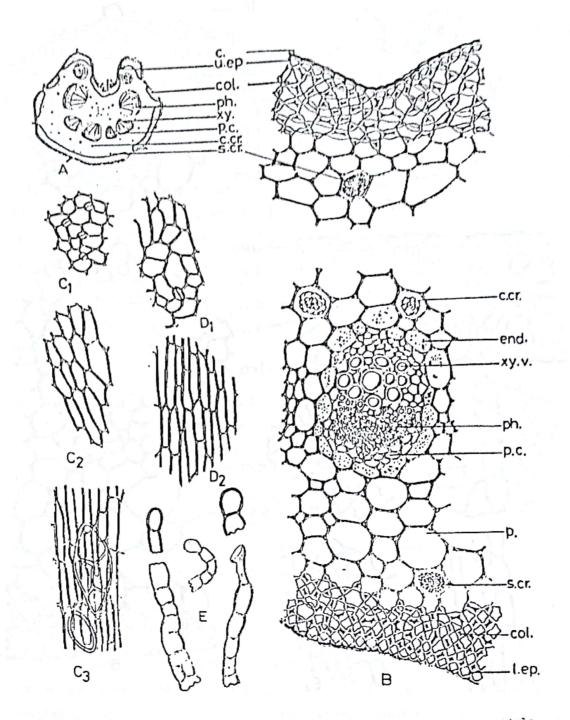
C- Detailed T.S. of the lamina. D₁- Upper epideris of lamina.

D₂-Upper epidermis midrib. E₁- Lower epidermis of lamina.

E₂-Lower epidermis of midrib.

C.c., Club-shaped-cells; C.c r., Cluster-crystal of calcium oxalate; Col., Collenchyma; Cr., Cortex; L. ep., Lower epidermis; P.C., pericyclic collenchyma; ph., phloem; P.L., Poricade-layer; S.cr., Sandy crystals of calcium oxalate; U.ep., Upper epidermis; V.S., Vascular strand; xylem.

(All x324; except Ax 56, B&C x208)



(Fig. 3) B- Diagram of T.S in the petiole.

C- Upper epidermis of the petiole

C: over the collenchyma of the rid KB. C: over the central collenchyma.

D- Lower epidermal cells of the petiole

D: over the collenchyma

D: over the collenchyma

E- Trienomes.

C., Cuticle; C.cr., Cluster crystals of calcium exalate; Col.,

Collenchyma; end., endodermis; L. ep., Lower epidermis, P., Parenchyma; pericyclic collenchyma; ph., Phloem; S.cr., Sendy crystal of calcium exalate; U.ep., Upper epidermis, xy., xylem.

(All x 172 except A x44)

The Epidermis

The epidermises of the lamina (Fig. 2,D $_1$ & E $_1$) consist of polygonal with wavy beaded anticlinal walls and covered with thin smooth cuticle. The epidermal cells of the petiole (Fig. 3,C & D), the neural regions , (Fig. 2,D $_2$ & E $_2$) and the stem (Fig. 5,C) are polygonal , axially elongated with thick outer periclinal walls, straight beaded anticlinal walls and covered with thin smooth cuticle. The dimensions of the epidermal cells are show in table (1) .

Stomata: Are anomocytic, present on both surfaces of the lamina , being more on the lower , few are present on the surface of petiole and the stem (Fig . 2, D & E, 3 ,C & D, 5 C_1 & C_2). They are oval or rounded in outline ,measuring 12 to 25 μ L and 10 to 20 μB .

Trichomes : (Fig. 4 ,A & 7,B) .Both covering and glandular trichomes are present on surfaces of leaf, petiole , and stem .They are multicellular uniseriate formed of 8 to 19 cells with occasionally collapsed ones, occasional trichomes are biseriate or multiseriate with branched base and few collapsed cells. The glandular trichomes have unicellular heads measuring 16 to 36 μ D.The cells have thin cellulosic walls and are covered with thin smooth cuticle . Trichomes measure 120 to 636 μ L and 10 to 33 μ D .

Cortex:

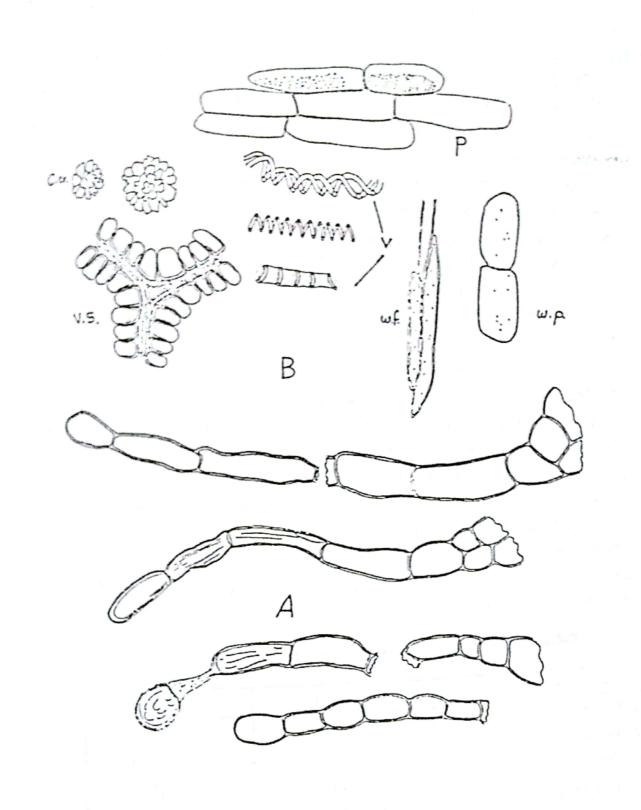
The cortex of the midrib, the petiole (Fig. 2,B & 3,B) and the stem (Fig.5 B & 6 ,B) is parenchymatous with 4 to 6 rows of angular collenchyma in the ridges region. Sandy and cluster crystals of calcium oxalate are present in the parenchymatous cells.

The Endodermis :

The endodermis is well differentiated as tangentially elongated cells contain starch granules in leaf and petiole, with pink pigments in stem and young root.

Table (1): Dimensions of the epidermal cells of leaf and stem (in micron)

Epidermis	"L" Length	"B" Breadth
Upper epidermis of the lamina.	20 - 42	16 - 28
Lower epidermis of the lamina.	24 - 42	10 - 28
Upper neural epidermis	32 - 56	5 - 14
Lower neural epidermis upper epidermis of petiole.	9 - 41	7 - 20
Over parenchyma	17 - 40	11 - 29
Over central collenchyma.	46 -115	5 - 17
Over collenchyma of wings.	28 - 97	9 – 29
Lower epidermis of petiole over collen-chyma.	64 -112	8 - 22
Lower epidermis of petiole over parenchyma.	20 - 64	14 - 42
Epidermis of stem over parenchyma.	20 – 56	20 - 28
Over collenchyma	28 -108	5 - 20



(All x440)

A- Trichomes. B- Isolated element from leaf.

C.cr., Cluster crystals of calcium oxalate; P., parenchymatous cells contain sandy crystals of calcium oxalate; V., Xylem vessel, V.C., venn-contain sandy crystals of calcium oxalate; V., Xylem vessel, V.C., venn-ation with clubshaped cells; W.f., Wood fiber; W.P., Wood parenchyma.

Pericycle:

The pericycle of the vascular bundles of the midrib, petiole (Fig. 2, B & 3, B) and the stem (Fig. 5, B & 6, B) are formed of moderately thick walled collenchymatous cells.

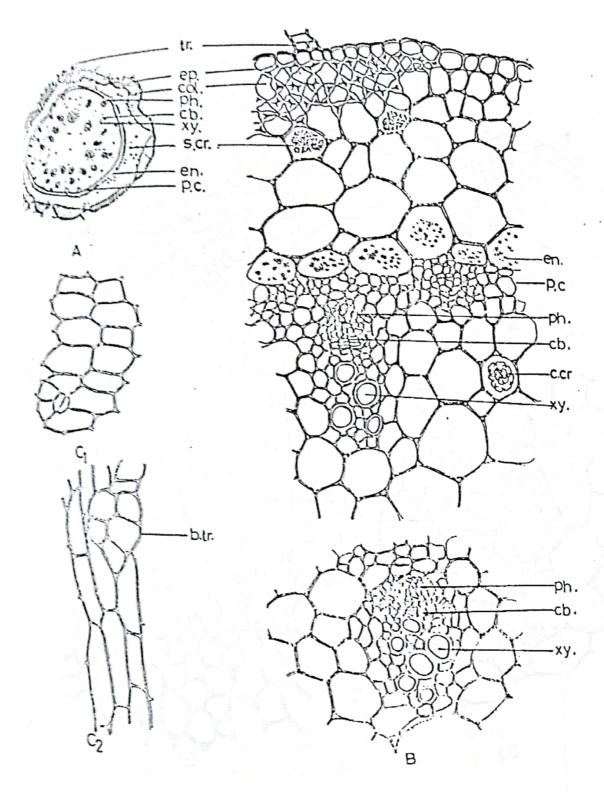
Vascular tissue:

The vascular bundles of the leaf (Fig. 2,B) young stem , (Fig. 5 ,B) and young root (Fig. 8, B) consist of cellulosic phloem of thin - walled elements, xylem formed of lignified spiral, annular, pitted and reticulate vessels 8 to 53 μ D , and thin walled cellulosic parenchyma . In old stem (Fig. 6,B) and old root (Fig. 9) , the phloem is formed of moderatelly thick - walled cellulosic elements , the cambium is indistinct, and the xylem is wholly lignified. It consists of wood fibres, vessels, few tracheids, tracheidal vessels and wood parenchyma, wood fibres (Fig. 7,A) have thick lignified pitted walls, wide lumen and acute to blunt or forked apices.

Vessels (Fig. 7, A) are annular, pitted or reticulate. Tracheids and tracheidal vessels (Fig. 7, A) are few with numerous bordered pits. Wood parenchyma (Fig. 7, A) is diffused, formed of polygonal, axially elongated cells with moderatly thick pitted, lignified walls. The medullary rays (Fig. 7, A) are uniseriate being cellulosic in the phloem and lignified in the xylem. In the root, the outer rings of vascular bundles are embedded in a layer of thick wall, lignified pitted cells. The dimensions of vascular tissue are shown in table (2).

Table (2): Dimensions of Vascular Tissue in Microns

The element	" L" Length	" D" Diameter
1. Wood fibre	188-352	8 - 37
2. Vessel		35 - 69
Tracheidal vessel	85- 205	11 - 25
4. Tracheids	80- 240	8 - 24
5. Wood parenchyma	56 - 200	8 - 41
6. Medullary rays	93- 187	22 - 46



Le Diagram of T.S. in young stem

C1:Epidermal cells between ridges

C2:Epidermal cells over ridges

C3:Epidermal cells over ridges

C4:Epidermal cells over ridges

C5:Epidermal cells over ridges

C6:Epidermal cells over ridges

C6:Epidermal cells over ridges

C7:Epidermal cells over ridges

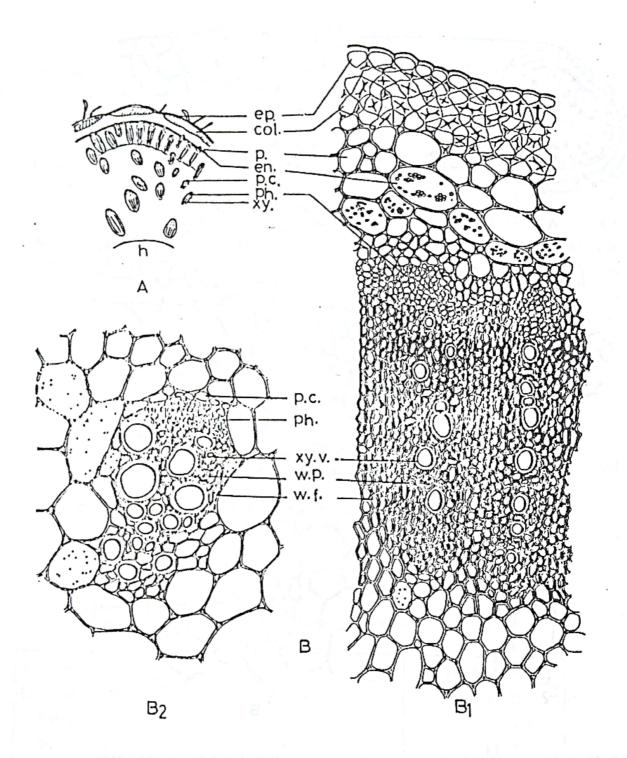
C8:Epidermal cells over ridges

C8:Epidermal cells over ridges

C8:Epidermal cells over ridges

C9:Epidermal cells over r

(Ax 44, B x196, C₁ x208, C₂ x208)



(Fig. 6) A- Diagram of T.S. in old stem.

B- Detailed of T.S. in old stem.

B₁: In the outer part

B₂: In the inner vascular bundle

Col., Collenchyma; en., endodermis., ep., epidermis; P., parenchyma; P.C., Pericyclic collenchyma; Ph., Phloem; w.f., Wood fibre; W.P., Wood parenchyma; Xylem vessels; Xy., Xylem.

(All x212, except A x28)

The pith is parenchymatous in young stem showing a hollow core in the old stem and contain sandy crystals of calcium oxalate.

Peridra: It is present in the root (Fig. 8, B & 9).

The cork: is formed of 4 to 6 rows of polygonal radially arranged cells with suberised walls; they measure 29 to 81μ L, and 23 to 52 μ H.

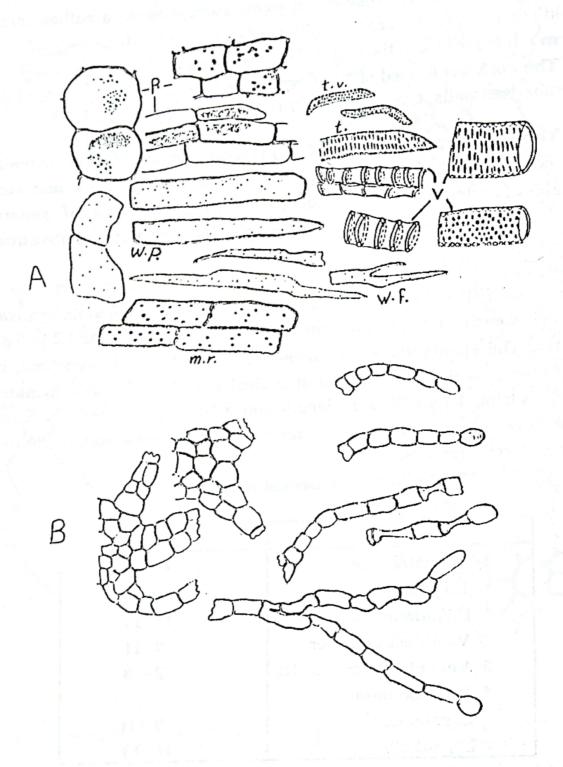
The phelloderm: (Fig. 8,B & 9) is wide, formed of parenchymatous cells containing simple or / and compound starch granules and sandy crystals of calcium oxalate. Successive irregular rings of separate vascular bundles being formed in phelloderm with subsequent lignification in the region of outer 3 rings.

Mesophyll:

The mesophyll (Fig. 2,C) is isobilateral with one row of columnar palisade cells, below each epidermis, and measure 6 to 29 μ in length and 2 to 8 μ in diameter. The spongy tissue is narrow formed of loose chlorenchma, but around the veins there is a sheath of cubical clubshaped parenchymatous cell, measuring 10 to 26 μ in length and 5 to 28 μ in breadth. Some mesophyll cells contain sandy or cluster crystals of calcium oxalate 20 to 56 μ D and starch granules.

Table (3): The numerical values of the leaf

Items	value
1- Stomatal index:	
L.Epidermis .	28 - 30
U.Epidermis	15 - 17
2- Vein - islet number	9-11
3- Veinlet termination No.	2- 3
4- Palisade ratio :	
U. palisade	9 - 11
L. palisade	11 - 13

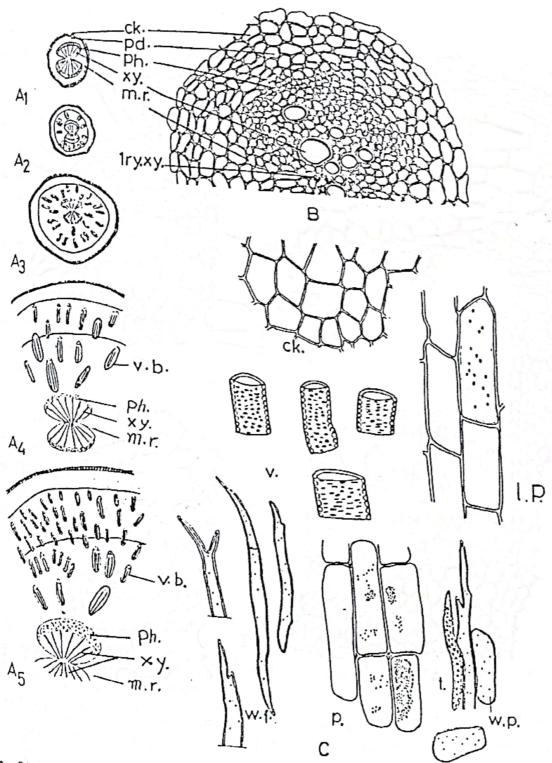


(Fig. 7) A- Isolated elements of stem

M.r., Medullary ray; p., parenchyma contain sandy crystal of calcium oxalate and starch; t., tracheid; t.v., tracheidal vessel;

V., Vessel; W.f., Wood fibre; W.P., Wood parenchyma.

(A x252, Bx 184)



(Fig. 8) A₁ - Diagram of T.S. of young root

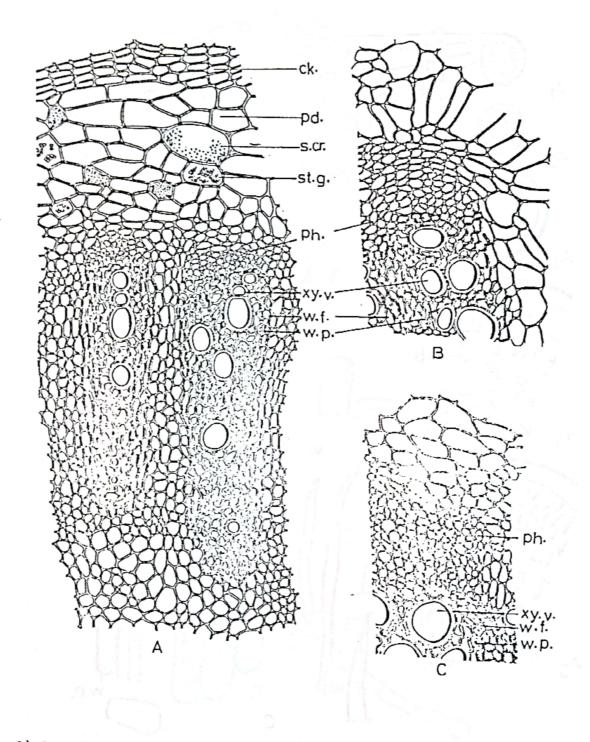
A₁-A₅Steps of anomalous secondry thickening of root

C - Detailed T.S. of young root

Isolated elements of the root

Ck., Cork; L.P., Lignified phelloderm; P., parenchymatous cells contain sandy crystals of calcium oxalate; p d., phelloderm; ph., phloem; V., Vessels; V.b., Vascular bundle; 1 ry.xy., primary xylem; Xy., Xylem.

 $(A_1 x36, A_1-A_5 x16, B x224, c x224)$



CONCLUSION

As a result of this study the following diagnostic features were found:

- 1. The plant is an annual herb attaining 3 m . in height, with alternate leaves and cylindrical stem which carry terminal spikes.
- 2. Leaf is green, hairy, petiolate, ovate, lanceolate with entire margins and acute apex .
- 3. Root is fusiform, tap with many lateral rootlets.
- 4- The leaf is iso-bilateral and the midrib with an arc of 1 to 7 collateral v.bs and an inverted v.b.
- 5- The epidermal cells of the leaf and stem are polygonal with straight or wavy occasionally beaded anticlinal walls, covered with thin smooth cuticle and showing anomocytic stomata.
- 6- Numerous non-glandular trichomes are present on epidermis of leaf and stem, bing multicellular uniseriate ends with a long terminal cell covered with thin smooth cuticle. Glandular trichomes have multicellular uniseriate, biseriate and multiseriate branched stalk and unjcellular head.
- 7- Pericycle is collenchymatous.

٠٠

- 8- The vascular tissue of the stem and root exhibit anomalous secondry thickening showing wide irregular rings of v.bs.
- 9- Wood fibres are lignified and pitted, vessels showing spiral, annular, pitted and reticulate thickening.

REFERENCES

- 1- Tackholm ,V; " Students Flora of Egypt ", Cairo University Press, 2 nd Ed., p. 130 - 137 (1974).
- 2- Perry , L.M." Medicinal Plants of East and Southern Asia", Atributed properties ad uses; the MIT press Combridge ,Massachusetts and London, England, pp. 8 - 12 (1980).
- 3- Sarg, T.M; Abd El-Aziz, E.M; Salem, S.A and Zayed R.A; (Phytochemical Invest.) Coopering in Egypt; Investigation of Amaranthus Chlorostachys willd., Growing in Egypt; Zag.J. Pharm.Sci, in press (1992).

- 4- Dassanayaka, M.D., Edt., "Flora of Ceylon", Oxford and IBH Publishing Co., New Delhi, Calcutta, Vol. 1, pp 1-57 (1980).
- 5- Metcalfe, C.R. and Chalk, L. " <u>Anatomy of the Dicotyledons in Relation to Taxonomy with Notes on Economic Uses</u>", Oxford University Press, Amenttous, London, E.C.U. Glasgow, New York, Toronto, Vol. 11, pp. 1062 1072 (1950).

الدراسة العيانية والمجهرية لاوراق وساق وجذر نبات الرُعاف (أمارانثوس كلوروستاكس)

طه مصطفی سرج، احسان محمود عبد العزیز ، سالم عبد المنعم سالم وراویه عبد الهادی زاید

قسم العقاقير – كلية الصيدلة – جامعة الزقازيق – مصر

سبق أن قام الباحثون بدراسة كيميائية لنبات الرُعاف الذي ينمو في مصر وقد أمكن فصل خلات الليبيول والليبيول ومادتين إستر اليفاتي والألفا سبينا ستيرول وألفا سبينا ستيرول وفعد فصل خلات الليبيول والليبيول ومادتين إستر اليفاتي والألفا سبينا وصابونين ومواد نتروجينية جلوكوزيد وبيتا سيتو ستيرول جلوكوزيد والروتين والكورستين وصابونين ومواد نتروجينية قاعدية مثل الكولين .

لذا فقد رؤي أن يتم دراسة الصفات العيانية والمجهرية لهذا النبات حتى يسهل التعرف عليه سواء في حالته الصحيحة أو على هيئة مسحوق .

تتضمن هذه الدراسة الفحص العياني والمجهري الأوراق وساق وجذر هذا النبات .