MACRO-AND MICROMORPHOLOGY OF <u>HIBISCUS SABDARIFFA</u> L.

Part 1: Root, Stem and Leaf.

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ABSTRACT

The macro-and micromorphology of the root, stem and leaf of Hibiscus sabdariffa L. cultivated in Egypt are presented aiming for their identification both in entireand powdered forms.

INTRODUCTION

The genus Hibiscus, includes 200 species which are widely distributed a in the tropics. Some species of the genus have wide medicinal uses. Among the uses of the different Hibiscus species are: the calyx of Hibiscus sabdariffa L. is a good cough sedative and wound dressing, the leaves of Htrionum L. are used as an expectorant, the roots and flowers of H.diversifolius Jacq. are used in pneumonia and the seed of H.cannabinus L. is used as a remedy for eye diseases and dysenteries.

The flowers of <u>Hibiscus</u> <u>sabdariffa</u> L. are used as sedative. emollient, and also cause abundant diuresis accompanied by slight diaphoresis. They also cause activation, neutralization of hepatic secretion, activation of secretion and intestinal contraction and show antiseptic effect. These activities cause rapid digestion, lower arterial blood pressure, and make this drug useful as a substitute for tea and coffe for persons who are sensitive to excitants³.

The anthocyanin content of $\underline{\text{H.sabdariffa}}$ L. was reported by karawya et al 4 .

The genus Hibiscus is a rich source of mucilage, which has a wide-spread application in medicine and industry $^{5-11}$. The majority of the Malvaceae mucilages have shown significant hypoglycemic activity on i.p. injection to normal mice 12 .

Reviewing the current literature, little was reported dealing with morphological characters of <u>H.sabdariffa</u> L. and general histological characters of genus <u>Hibiscus</u>. So, it was found of interest to carry out such study.

EXPERIMENTAL

Material:

The fresh, roots, stems and leaves of <u>H.sabdariffa</u> L. were collected at different stages of growth from growing plants in the Experimental Stations, Faculty of Pharmacy, Assiut University. The identity of the plant was verified by Prof. Dr. A.Fayed, Prof. of plant taxonomy, Dept of Botany, Faculty of Science, Assiut University. The separated parts were preserved in alcohol 70% containing 5% glycerin. The powder was obtained from airdried plants.

MACROMORPHOLOGY

Habitat:

Hibiscus sabdariffa L. (Fig. 1) is an annual, erect clyindrical herb, 1.5-2.5 m. in height. The plant shows monopodial branching with cauline leaves and well developed tap-root system. The flowers arise in the axils of the upper leaves.

1- The Root (Fig. 1A):

It is tap-root bearing several lateral rootlets, yellowishbrown in colour and penetrating the soil vertically. Externally it is longitudinally wrinkled, it has no odour, having characteristic mucilaginous taste, the fracture is fibrous and the central part is solid.

II- The Stem (Fig. 1,B):

It is erect, cylindrical and glabrous. It measures 1.5 to 2.5 m. in height and 1.5 to 3.0 cm in diameter. Externally, it is purple in colour with no odour and having a mucilaginous taste. It bears long internodes varing in length from about 2 cm. at the base to about 20 cm. toward the apical portion. From each node arises a branch from the axil of a long petiolate leaf. It is monopodially branched and the leaves are arranged alternatively.

III- The Leaf (Fig. 1,B)

It is cauline, simple, altrnate and palmately divided into 3 to 5 oblong-lanceolate. It has a serrate margin with acut apex and symmetric base. The upper leaves are stipulate varying in lenght from 5 to 7.5 cm and 3 to 4 cm in width. It has a green colour, often blotched with purple, odourless and having a mucilaginous taste. The stipules are linear, acute. The lower leaves are entire. Petiole varying from 4.0 to 6.5 cm in length reddish purple in colour. The midrib and big veins are prominent on both surfaces. The leaves show pinnate reticulate vennation.

MICROMORPHOLOGY

l- The Root:

A transverse section in the root (Fig. 2,A) appears nearly rounded in outline. It shows a superficial layer of irregular reddish brown cork, followed by parenchymatous cortex interrupted with several groups of fibres. The pericycle is formed from

nearly complete ring of pericyclic fibres interrupted with parenchymatous cells, followed by phloem. The cambium is formed of several well distinct layers, followed by a wide region of radiating xylem. Medullary rays are biseraite traversing the phloem and xylem.

The cork (Fig. 2,B,C) is formed of narrow zone of several layers (4-5) of tangentially elongated radially arranged, polygonal tabular cells with thin non-lignified walls. In surface view, the cells appear polygonal, sometimes isodiametric, they measure about 88-122-158 u in length, 50-78-105 u in width and 10-14-18 u in height.

The Cortex (Fig. 2,B):

The secondary cortex (Phelloderm) is formed of (2-3 layers) of thin walled polygonal parenchymatous cells. The primary cortex is formed of 5-6 layers of polygonal or nearly rounded parenchymatous cells with thin cellulosic walls, narrow inter cellular spaces. Some cells contain large cluster crystals of calcium oxalate, measuring from 45-52-58 u in diameter.

The pericycle (Fig.2,B,C) is formed of successive groups of fibres alternating with parenchymatous zones. The fibres are lignified with wide lumena and acuminate or tapering apices, measure from 3500-3566-3632 u in length and 13-38-63 u in width.

The phloem (Fig. 2,B) consists of intermittent phloem tissue, sieve tubes, companion cells, and phloem parenchyma, but no phloem fibres.

The cambium (Fig. 2,B) is represented by a complete ring of 4-7-rows of tangentially elongated, radially arranged, thin-walled meristematic cells.

The xylem (Fig. 2,B,C) is a comparatively wide cylinder of lignified elements. The vessels are mainly solitary or in small groups having bordered-pitted walls measure from 43-128-170 u in diameter, which are accompanied with tracheids, wood fibres and wood parenchyma. The tracheids having lignified walls and showing simple pits, measuring 180-465-750 u in length and 18-43-68 u in width. Numerous wood fibres have slightly irregular walls, wide lumena and acute or accuminate apices measure from 725-750-775 u in length and 13-26-38 u in width. The wood parenchyma cells are rectangular to subrectangular in shape with pitted lignified walls.

The medullary rays (Fig. 2,B) are bi or tri-seriate. The cells are radially elongated with slightly thick lignified and pitted walls. Starch granules are scattered in the cells of the medullary rays.

THE POWDER

Powdered root (Fig. 2,C) is pale brownish-yellow in colour, odourless and having a mucilaginous tast. It is characterized microscopically by the following:

- 1- Fragments of polygonal, thick-walled, non-lignified cork cells.
- 2- Numerous cluster crystals of calcium oxalate.
- 3- Fragments of tracheids and wood parenchyma with lignified pitted walls.
- 4- Fragments of xylem vessels with lignifed pitted walls.
- 5- Fragments of pericyclic fibres with thick-lignified walls.
- 6- Fragments of wood fibres with lignified, thin walled, wide lumena and acute apices.

II- The Stem:

A transverse section in the young stem (Fig. 3A) is more or less rounded in outline. It is formed of an epidermis carrying covering and glandular trichomes, the cortex is formed of 2-3 rows of chlorenchymatous cells followed by 3-4 rows of collenchymatous cells and 4-5 rows of parenchyma. The pericycle shows intermittent groups of lignified fibres, the central stele enclosing a wide parenchymatous pith. Starch granules, cluster crystals of calcium oxalate and mucilage cavities are observed in the parenchymatous tissues of the cortex and pith. Cluster crystals of calcium oxalate are also observed in the chlorenchymatous cells.

In the old stem (Fig. 3B) the pericycle is developed into several tangentially arranged batches of lignified fibres compressing the cortex into a narrow zone of parenchymatous cells, and the xylem increase redially compressing the pith region. The mucilage cavities are not observed.

The epidermis (FIg. 3C,4) is formed of one row of cells which are polygonal tabular isodiametric with straight, and thin-cell-ulosic anticlinal walls measure from 58-71-85 u in length, 28-43-58 u in width and 23-33-43 u in diameter. They are covered with smooth cuticle, carrying oval anisocytic stomata and both covering and glandular trichmes. The glandular trichmes are formed of unicellular stalk. multicellular (8-12 cells), biseriate, globular head measure from 80-99-118 u in length and 60-68-75 u in width. The non-glandular trichomes occur either as single or stellate of 2-4 unicellular hairs with thin smooth cellulosic walls, wide lumena and acute apices, measure from 230-490-750 u in length and 20-29-38 u in width.

The cortex(Fig. 3C) is comparatively narrow and formed of 3-4 rows of thick-walled parenchymatous cells with intercellular spaces. Cluster crystals of calcium oxalate are scattered in the cortical tissue, measure from 45-53-60 u in diameter.

The endodermis (Fig. 3C) is formed of thin-walled parenchymatous cells containing numerous starch granules.

The pericycle (Fig. 3C,4) is formed of successive groups of fibres which are tangentially arranged. They are spindle shaped with slightly thick lignified walls, wide lumena and acute apices measure from 1320-1375-1430 u in length, 28-30-33 u in width The parenchymatous cells between the groups of fibres contian cluster crystals of calcium oxalate resembling those of the cortex.

The phloem (Fig. 3C) is formed of phloem tissue consisting of sieve tubes, companion cells and phloem parenchyma.

The cambium (Fig. 3C) is represented by 2-4 rows of tangentially elongated, radially arranged, thin-walled meristematic cells.

The xylem (Fig. 3C,4) consists of lignified elements. The vessels (Fig. 4) are solitary or in small groups of 2-3 vessels showing spiral, pitted, reticulate and scalariform thickening measuring from 23-55-88 u in diameter. The vessels are accompanied with tracheid, wood fibres and wood parenchyma. The fibres are abundant, spindle-shaped having thin-lignified walls, lumena and blunt or tapering apices measuring from 450-663-375 u in width. The wood parenchyma consists of elongated cells with pitted lignified walls. The medullary rays are 3-6 cells wide. The cells are radially elongated lignified and pitted, measuring from 166-173-180 u in length and 33-44-55 u in width.

The pith (Fig. 3C) is formed of large, more or less rounded wide parenchymatous cells and wide intercellular spaces, some

cells are filled with very minute starch granules. Cluster crystals of calcium oxalate resembling those of the cortex are observed.

THE POWDER

Powdered stem (Fig. 4) is greenish-yellow in colour, odour-less and having a mucilaginous taste. Microscopically it shows the following:

- 1- Fragments of polygonal epidermal cells with straight anticlinal walls and smooth cuticle, carrying anisocytic stomata.
- 2- Numerous glandular and covering trichomes.
- 3- Numerous cluster crystals of calcium oxalate.
- 4- Fragments of both pericyclic and wood fibres, the latter show more wide lumena.
- 5- Fragments of tracheids, wood parenchyma and medullary ray cells with pitted lignified walls.
- 6- Fragments of xylem vessels with spiral, pitted, reticulate and scalariform thickening.

III- The Leaf:

A transverse section in the lamina through the midrib (Fig. 5A) appears biconvex in outline, in the midrib region. It shows an upper and lower epidermises enclosing in between a dorsiventral mesophyll which is replaced in the midrib region by the vascular strand and the cortical tissue.

The upper epidermis (Fig. 5B,D) is formed of polygonal tabular cells, with straight thin-cellulosic anticlinal walls and covered with smooth cuticle. The cells of the lower epidermis (Fig. 5B,E)is more or less similar to the upper one. Oval anisocytic stomata are distributed on both surfaces, but they are more on the lower one. Glandular trichomes are present on both surfaces resembling those of the stem.

The mesophyll (Fig. 5,C) is dorsiventral, being differentiated into one row of cylindrical columnar palisade cells and spongy tissue. The spongy tissue is formed of 6-7 rows of chlorenchymatous cells which are thin walled, rounded or slightly irregular in shape showing fairly wide intercellular spaces. Cluster crystals of calcium oxalate are scattered in the spongy tissue.

The cortical tissue (Fig. 5B) is formed of an upper and lower subepidermal masses of collenchyma, each of 3-5 rows of polygonal collenchymatous cells with thick cellulosic walls showing no intercellular spaces. The rest of cortical tissue is formed of almost rounded, large thin-walled parenchymatous cells, some of them containing cluster crystals of calcium oxalate and few starch granules.

The vascular tissue (Fig. 5B) is represented in the midrib by a large strand with the xylem to the adaxial side and the phloem to the abaxial side.

The xylem (Fig. 5B,7) is formed of lignified vessels and thin-walled wood parenchyma. The vessels possess spiral and scalariform thickenings. Medullary rays are formed of 2-3 rows of radially elongated thin-walled cellulosic cells. The phloem is formed of small, thin-walled shining cellulosic cells, hardly differentiated into sieve tubes, companion cells and phloem parenchyma. The phloem tissue is interrupted by one row of thin-walled parenchymatous cells. The cambium is hardly distinguishable, being represented by a little cambiform tissue between the xylem and phloem.

The Petiole:

A transverse section in the petiole (Fig. 6A) is more or less rounded in outline, showing a hairy epidermis followed by the cortex. The pericycle is formed of numerous groups of lignidied

fibres interrupted by parenchymatous cells. The vascular tissue is formed of several vascular strands in a continuous ring enclosing a wide parenchymatous pith.

The epidermis (Fig. 6B,C) is formed of polygonal, tabular isodiametric cells with straight, thin cellulosic anticlinal walls. They are covered with smooth cuticle, carrying anisocytic stomata and both covering and glandular trichomes resembling those of the stem.

The cortex (Fig. 6B) is formed of 1-2 rows of chlorenchy-matous cells followed by 4-5 rows of collenchymatous cells with no intercellular spaces and 4-5 rows of parenchymatous cells with distinct intercellular spaces. Cluster crystals of calcium oxalate are observed in the chlorenchymatous and cells of the cortex.

The pericycle (Fig. 6B,7) is formed of a continuous ring of fibres separated by parenchymatous cells. The fibres are strap-shaped with thick lignified walls, narrow lumena and acute apices.

The phloem (Fig. 6B) is formed of thin-walled, shining soft cellulosic elements.

The cambium (Fig. 6B) is represented by 2-3 rows of tangentially elongated thin-walled meristematic cells.

The xylem (Fig. 6B,7) is formed of lignified vessels with spiral and scalariform thickenings. Medullary rays are uniseriate, occasionally biseriate, formed of radially elongated, thick-walled lignified cells.

Wood fibres (Fig. 6B,7) are similar to those of the pericycle but with thinner walls and wide lumena.

The pith (Fig. 6B) is formed of wide parenchymatous cells with thick pitted cellulosic walls and wide intercellular spaces. It shows cluster crystals of calcium oxalate as well as numerous scattered starch granules.

POWDERED LEAF AND PETIOLE

Powdered leaf (Fig.7) is green in colour, odourless and having a mucilaginous taste. It is characterized microscopically by the following:

- 1- Fragments of mesophyll parenchyma and palisade cells.
- 2- Fragments of upper and lower epidermal cells as well as epidermal cells of the petiole.
- 3- Numerous glandular and covring trichomes.
- 4- Numerous cluster crystals of calcium oxalate which are either free or in the cells.
- 5- Fragments of lignified vessels, with spiral and scalariform thickenings.
- 6- Fragments of wood fibres of the petiole which are thinner in walls and wider in lumena than those of the pericyclic fibres.

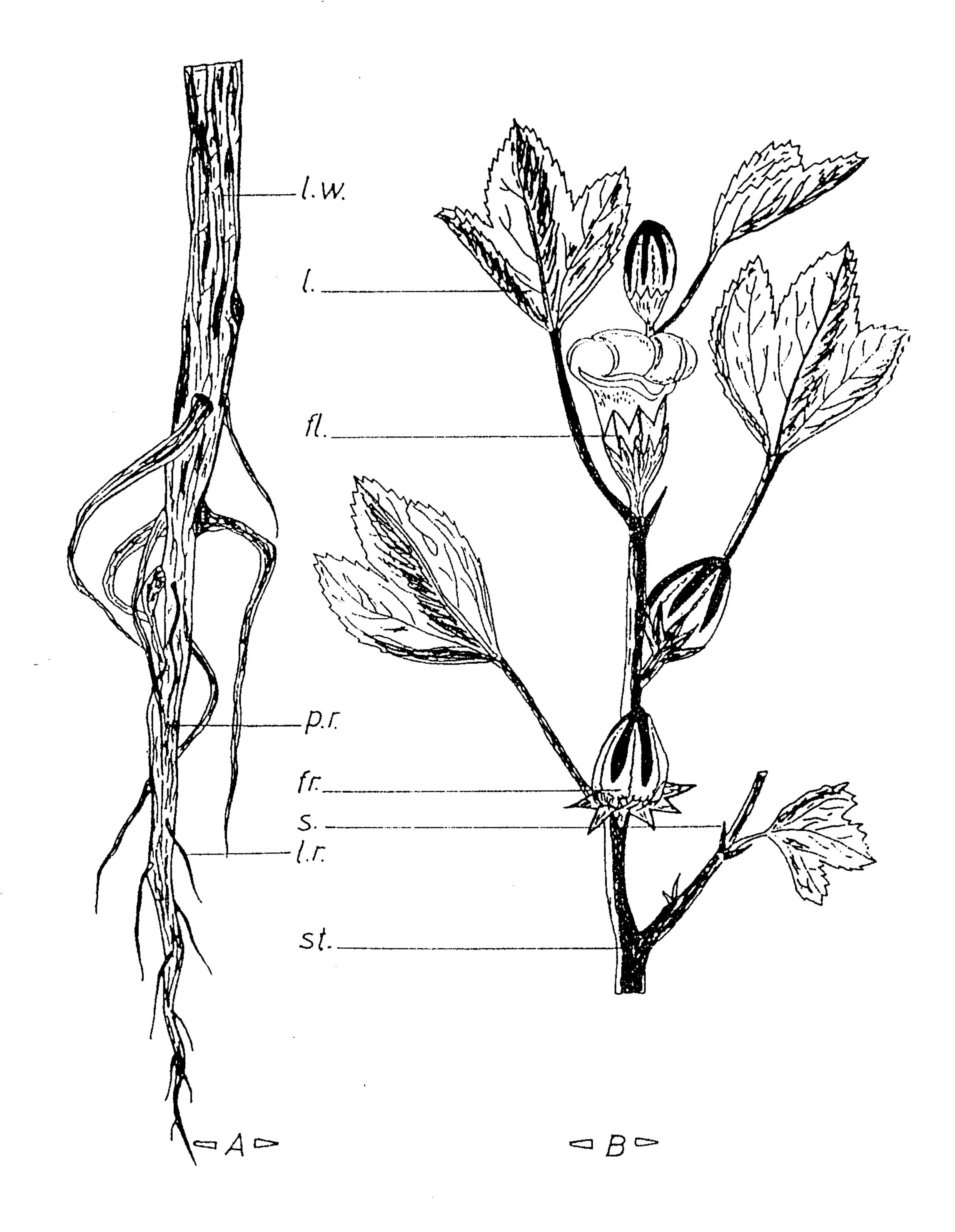


Fig. 1: Sketch of Hibiscus sabdariffa L.

A- The root (x 0.7)

B- Aerial part of the plant (x 0.5)

fl., flower; fr., fruit; l., leaf; l.r., lateral rootlets; l.w., longitudinal wrinkles; p.r., primary root; s., stipule; st., stem.

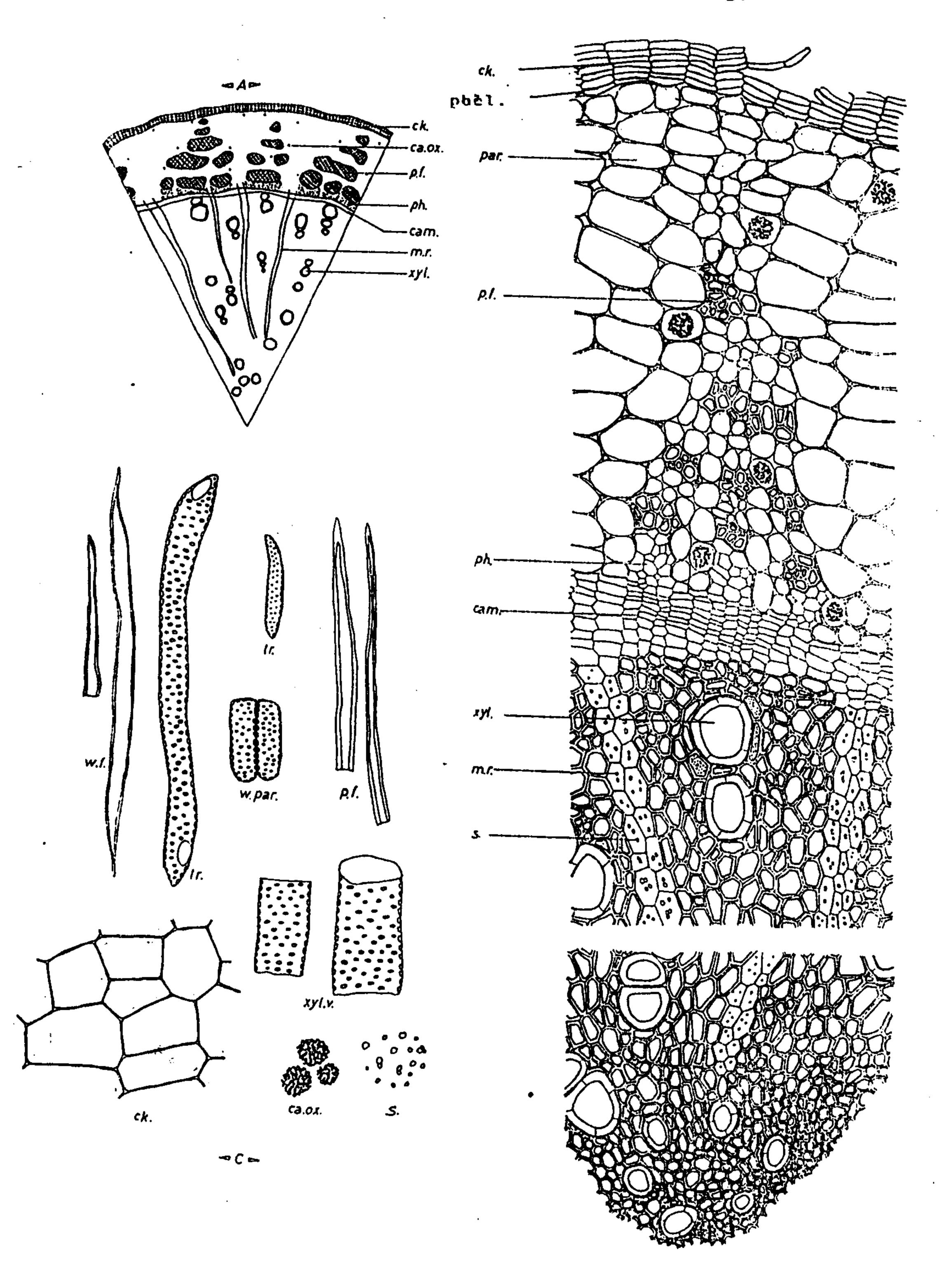


Fig. 2: The root of Hibiscus sabdariffa L.

A- Diagrammatic transverse section. (x20)

B- Detailed sector. (x82)

C- Powdered root. (x82)

cam., cambium; ca. ox., calcium oxalate; ck.,

cork; par., parenchyma; m.r., medullary ray;

p.f., pericyclic fibre; ph., phloem; s., st arch;

tr., tracheid; w.f., wood fibre; w.par., wood

parenchyma; xyl., xylem; xyl.v., xylem vessel.

Macro-and Micromorphology of <u>Hibiscus Sabdariffa</u> L.

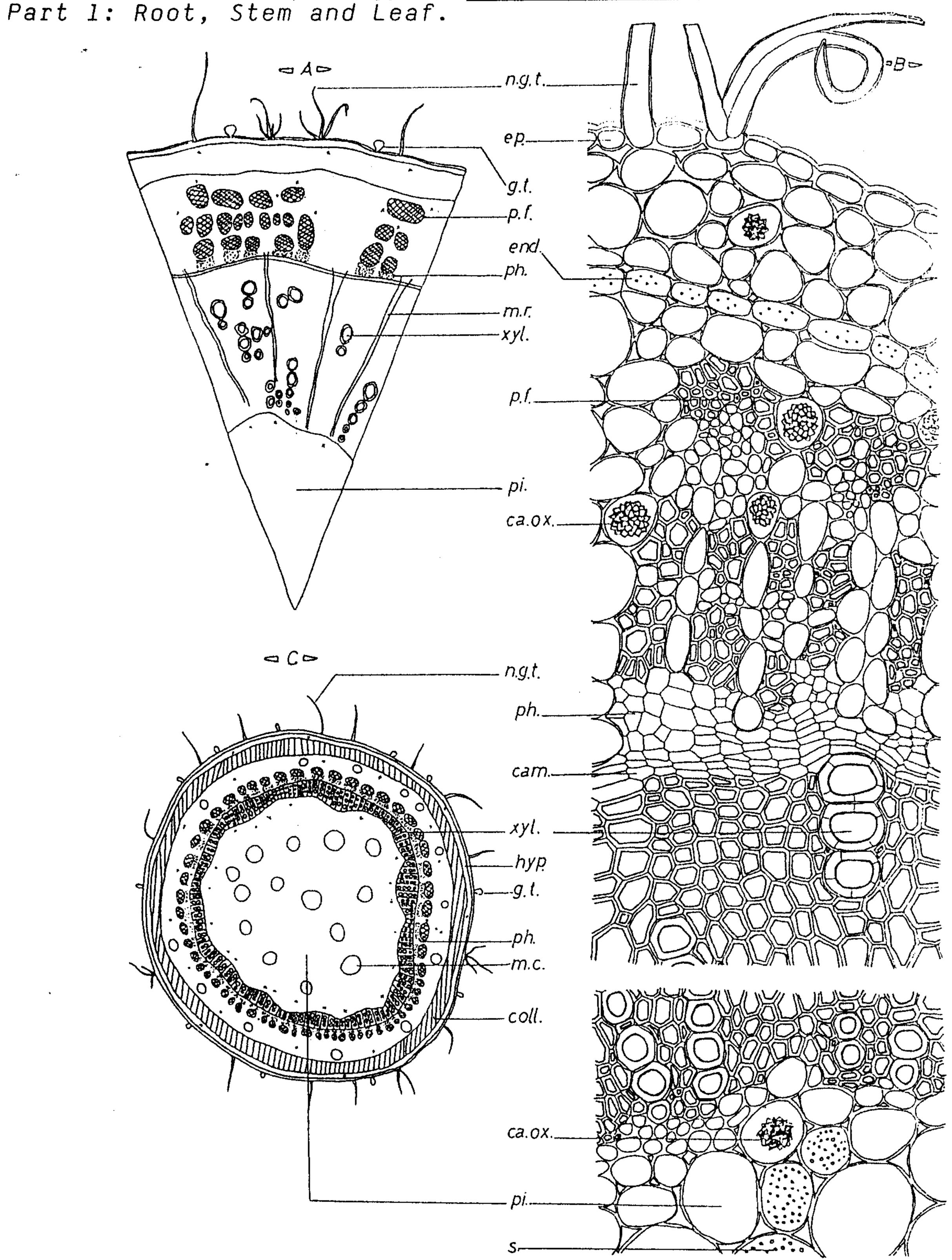


Fig. 3: The stem of Hibiscus sabdariffa L.

A- Diagrammatic transverse section in the old stem (x'29)

B- Detailed sector in the old stem. (x120)

C- Diagrammatic transverse section in the young stem(x 10)

cam., cambium; ca. ox., calcium oxalate; coll.,

collenchyma; end., endodermis; ep., epidermis; g.t.,

glandular trichomes; hyp., hypodermis; m.r., medyl
lary ray; m.c., mucilage cavity; n.g.t., non-gland
ular trichome; p.f., pericyclic fibre; ph., phloem;

pi., pith; s., starch; xyl., xylem.

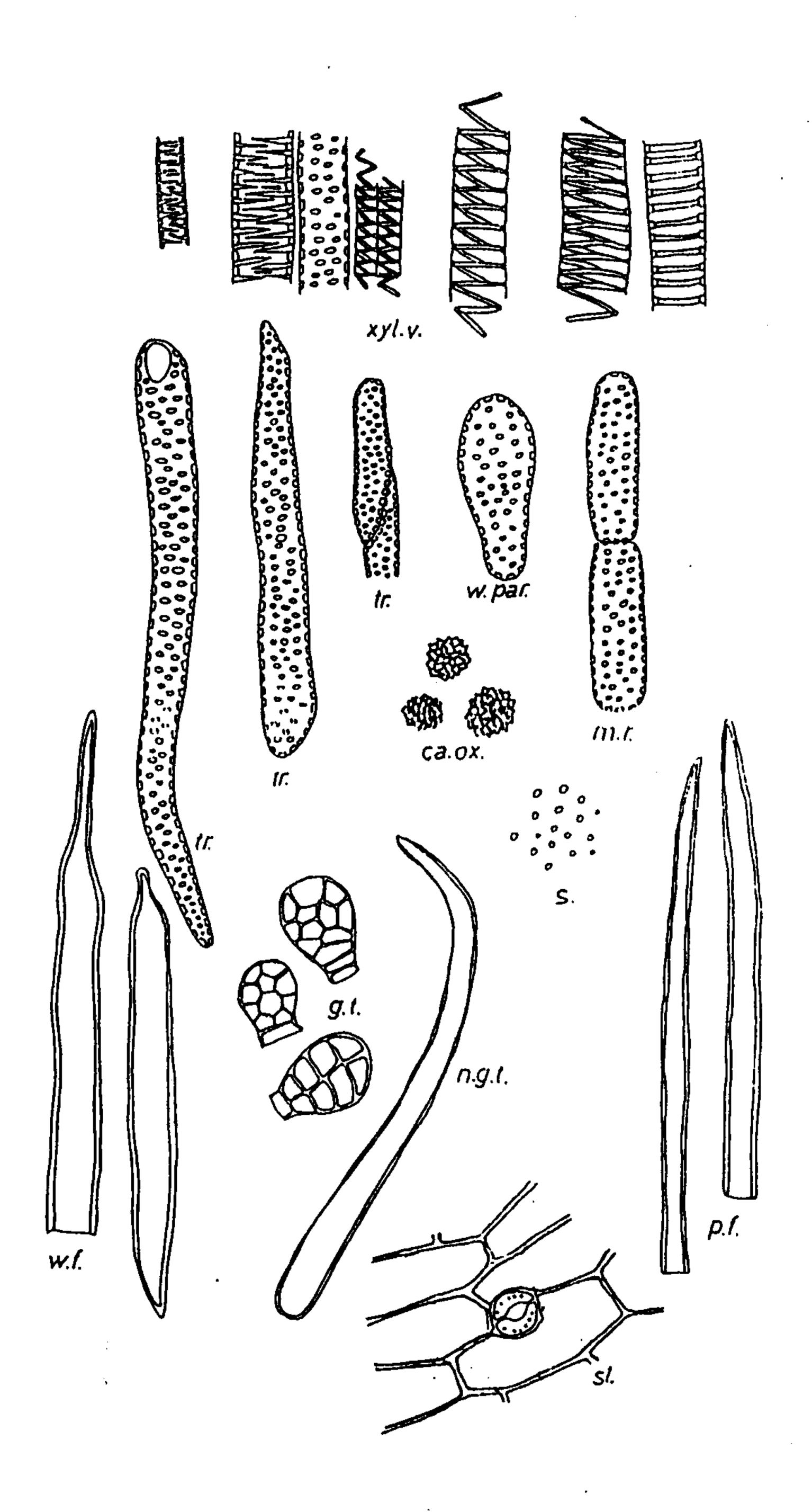


Fig. 4: Powdered stem of Hibiscus sabdariffa L. (x 120)
ca.ox., calcium oxalate; g.t., glandular trichome;
m.r., medullary ray; n.g.t., non-glandular trichome;
p.f., pericyclic fibre; st., stomata; w.f., wood
fibre; w.par., wood parenchyma; xyl.v., xylem vessel.

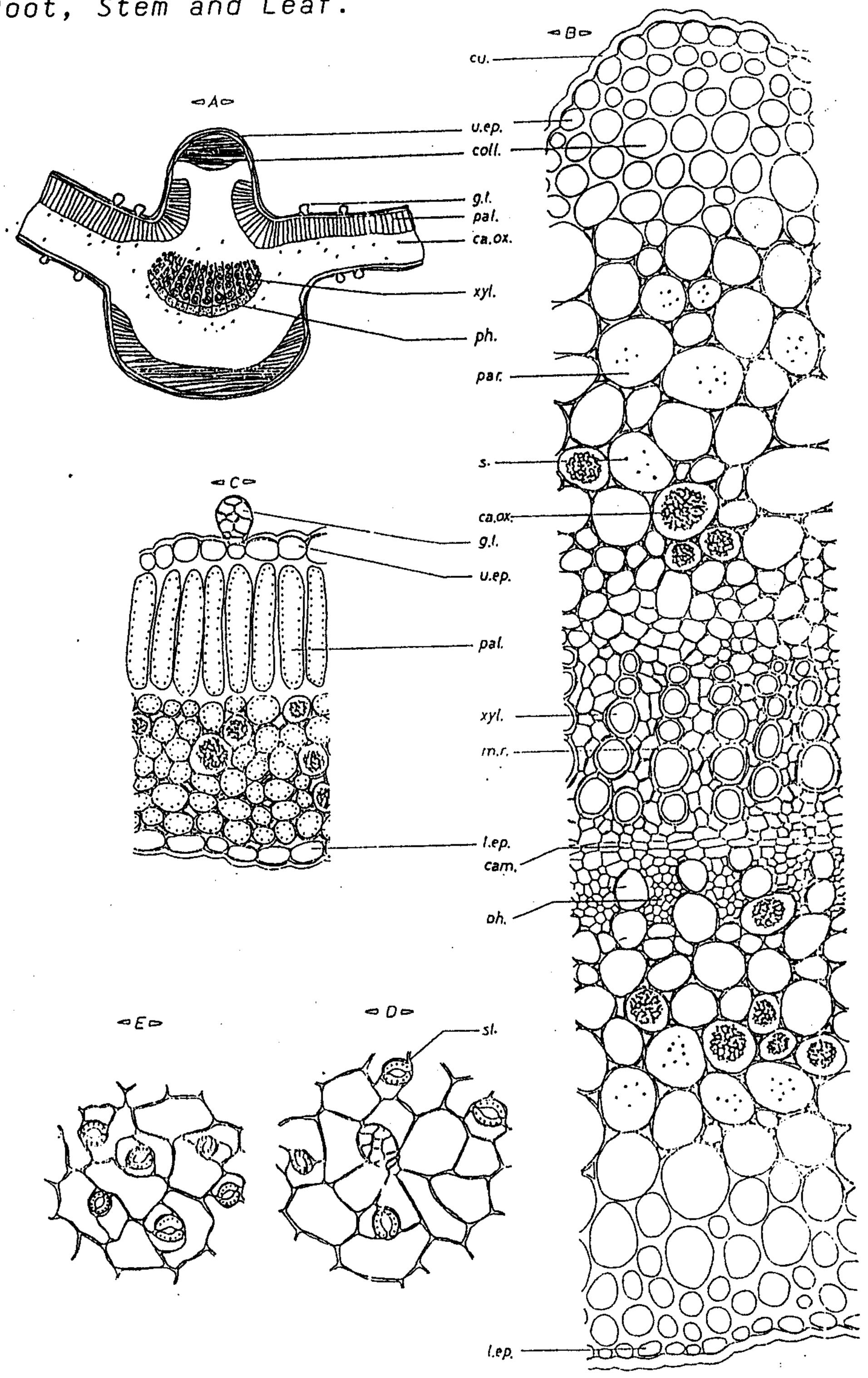


Fig. 5: The leaf of Hibiscus sabdariffa L. A- Diagrammatic transverse section in the leaf. (x 18)B- Detailed sector in the midrib. (x 80)C- Detailed sector in the lamina (x 80)D- Surface preparation of the upper epidermis. (x 80)E- Surface preparation of the lower epidermis. (x 80)cam., cambium; ca.ox., calcium oxalate; coll., callenchyma; cu., cuticle; g.t., glandular trichome; l.ep., lower epidermis; m.r., medullary ray; pal., palisade; par., parenchyma; ph., phloem; s., starch; st., stomata; u.ep., upper epidermis; xyl., xylem.

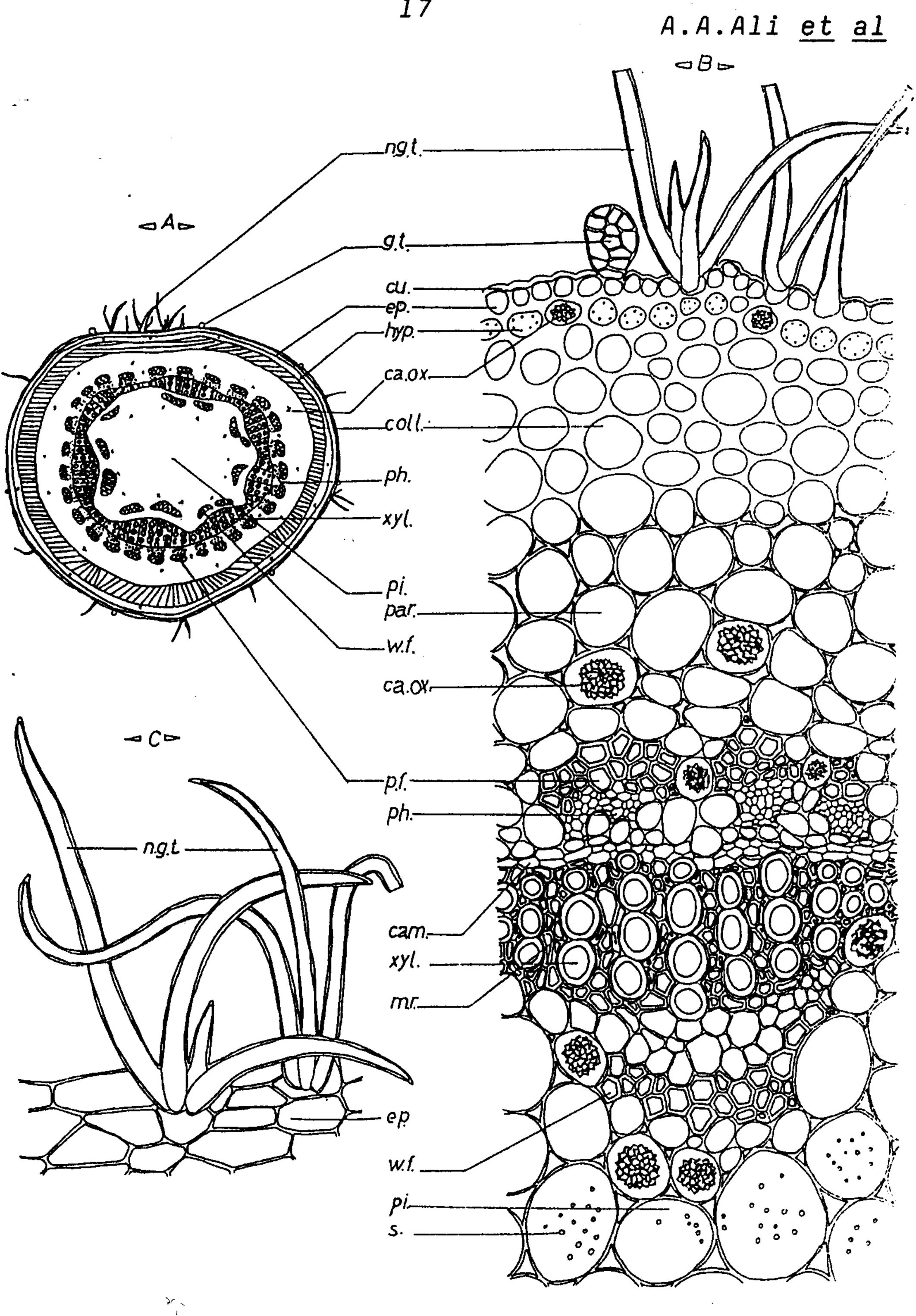


Fig. 6: The petiole of the leaf of Hibiscus sabdariffa L. A- Diagrammatic transverse section. (x 15)B- Detailed sector. (x116) C- Surface preparation. (x116) cam., cambium; ca.ox., calcium oxalate; coll., collenchyma; cu., cuticle; ep., epidermis; g.t., glandular trichome; hyp., hydrodermis; m.r., medullary ray; n.g.t., non-glandular trichome; par., parenchyma; p.f., pericyclic fibre; ph., phloem; pi., pith; s., starch; w.f., wood fibre; xyl., xylem.

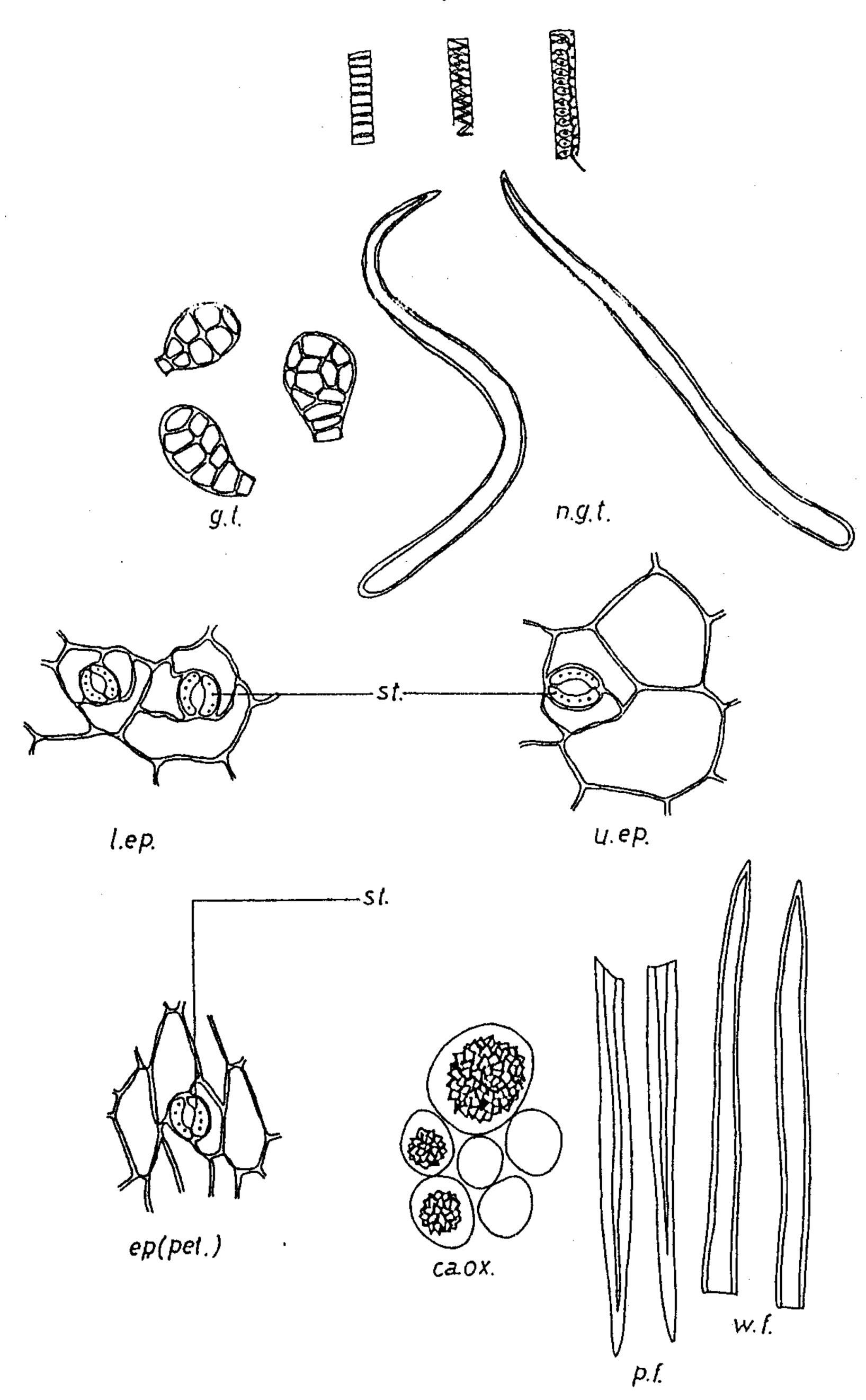


Fig. 7: Powdered leaf and petiole of Hibiscus sabdariffa L. (x 116)

ca.ox., calcium oxalate; ep. (pet.) epidermis of the petiole; g.t., glandular trichome; l.ep., lower epidermis; n.g.t., non-glandular trichome; p.f., pericyclic fibre; st., stomata; w.f., wood fibre; u.ep., upper epidermis; xyl.v., xylem vessel.

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