CHAPTER - XIII

Pharmacognosy of Withania somnifera Dunal

Withania somnifera Dunal (Solanaceae) is known as 'asvaghandha, asan or asguned'. It is a reputed medicinal plant belonging to indigenous systems of medicine. All the parts of the plant, but especially the roots are used in medicine. The roots are official in Indian Pharmacopoeia (1955).

Roots have been the subject of several chemical and pharmacological investigations. Trebut (1886), found that the plant had hypnotic and sedative properties due to the presence of an alkaloid somniferine. Later on, Power and Salvay (1911) found traces of an essential oil, resin, phytosterol, palmic acid, cerotic acid, oleic acid, linolic acid, withanol etc. from the root. Majmudar and Guha (1933), have shown that the roots contain more than one basic constituents. Later on, Majmudar (1955) succeeded in isolating no less than eight alkaloids.

Dutta and Mukerji (1950), have given some pharmacognostic description of the roots, while Newwald and Loges (1956) mention the distincuishing pharmacognostic characters of Withania roots and Rauwolfia serpentina roots.

Kurup (1956) found a pale yellow crystalline solid from alcoholic extract of the leaves, which in dilution of 1: 600,000 completely inhibited the growth of S. aureus.

Description of the plant (Plate XXIII, 1):

The plant is a perennial, branched, erect It has a tap root which is more than 5 cm. under shrub. thick. Aerial part is covered by wooly hairs. Stem and a number of branches emerge from the top of the root. Lower leaves are alternate but the upper ones are opposite. Flowers are yellowish-green and in axillary groups of 1-3-6 (Fig. 1B). Calyx is hairy on its outer side, gamosepalous and possesses five long and linear teeth. Calyx-base is deltoid. Corolla is gamopetalous and possesses five lanceolate and acute Stamens are five in one whorl, epipetalous and alternating with the corolla lobes. Ovary is two celled and the two carpels are obliquely placed. Ovules are many on an axile placenta. Stigma is bifid. Fruit is a berry, red when ripe and enclosed in a glabrous slightly five angled inflated calyx having five connivent calyx teeth above (Fig. 1A).

$\underline{\mathtt{M}} \ \underline{\mathtt{O}} \ \underline{\mathtt{R}} \ \underline{\mathtt{P}} \cdot \underline{\mathtt{H}} \ \underline{\mathtt{O}} \ \underline{\mathtt{L}} \ \underline{\mathtt{O}} \ \underline{\mathtt{G}} \ \underline{\mathtt{Y}}$

Leaf (Plate XXIII):

A leaf has a small petiole which is grooved

at the tip. Both the surfaces are dull green and are usually clothed with hairs. It measures 5-10 cm. $\times 2.5-5$ cm. and is ovate with acute apex, oblique base, wavy margin and reticulate venation.

Stem:

Stem or a branch is variously thickened according to its age and go vertically upwards. A node is prominent only at the side from where petiole arises.

Internodes measure 1.5-3 cm. in length. It is cylindrical green and shows longitudinal wrinkles. Young branch possesses many whitish hairs. Transversely cut surface shows yellow-grey cork, whitish-yellow cortex and central hollow pith. Fracture is short and uneven.

Root (plate XXIII):

Young root is straight, unbranched, tuberous and conical. Direction of growth is straight downwards. It is 5-12 mm. in width just below the crown. The thickness varies with age and bears fibre-like secondary roots. Outer surface is buff to grey-yellow and wrinkles are longitudinal. Crown consists of 2-6 remains of a stem bases (Figs. 1A & 1B). Smoothened transverse surface shows small outer bark and in the centre, soft solid mass with scattered pores. Taste is mucilagenous bitter while odour is strong.



PLATE - XXIII

(Figs. 1 - 7: Withania somnifera Dunal)

Fig. 1 - 1A - Entire root from a young plant. x 1

1B - Market sample under the name
'asvaghandha'. x 1.

Fig. 2 - Entire root from a old plant. x 1.

Fig. 3 - 3A - A twig with leaves and fruits. x 1.

3B - A nodal portion of the twig showing a fascicle of flowers. x 1.

Fig. 4 - T.s. lamina. x 325.

Fig. 5 - Glandular and covering trichomes of leaf and stem. x 290.

Fig. 6 - Upper epidermis with stomata and cuticular striations. x 330.

Fig. 7 - Lower epidermis with stomata and cuticular striations. x 330.

x-x-x-x-x-x

Cr₁ and Cr₂ - calcium oxalate crystal; Ep₁ - upper epidermis;

Ep₂ - lower epidermis; G.Tr. - glandular trichome; Fl. - flower;

Ft. - fruit; Pal. - palisade; Spg. - spongy; S.B. - stem pase;

S.Rt. - secondary root; Tr. - trichome.

Old root gives out a number of secondary and tertiary roots (Fig. 2). Main root and its branches are woody with an external brown-black cork. Externally, it shows a number of short longitudinal striations. Both the secondary and tertiary roots are fibrous.

HISTOLOBY

Leaf (Plate XXIII & XXIV)

beneath the upper epidermis only (Plate XXIII,4). Epidermal cells are tabular with straight anticlinal walls. Cells of the upper epidermis are protected by a thicker layer of cuticle than those of the lower one. Some of the epidermal cells on both sides, bear warty covering trichomes. These may be unicellular or multicellular and uniseriate or branched (Plate XXIII, 5). Some of these candelabra trichomes are multicellular upto 15 cells. Glandular trichomes having one celled to multicelled heads are also present and are located on the upper epidermis (Plate XXIII, 5).

Cells of the upper epidermis in surface view show less wavy walls and prominent cuticular striations than those of the lower one (Plate XXIII, 6) and 7). Stomata, which are more on the lower epidermis are of ranunculaceous and crufiferous types. Palisade

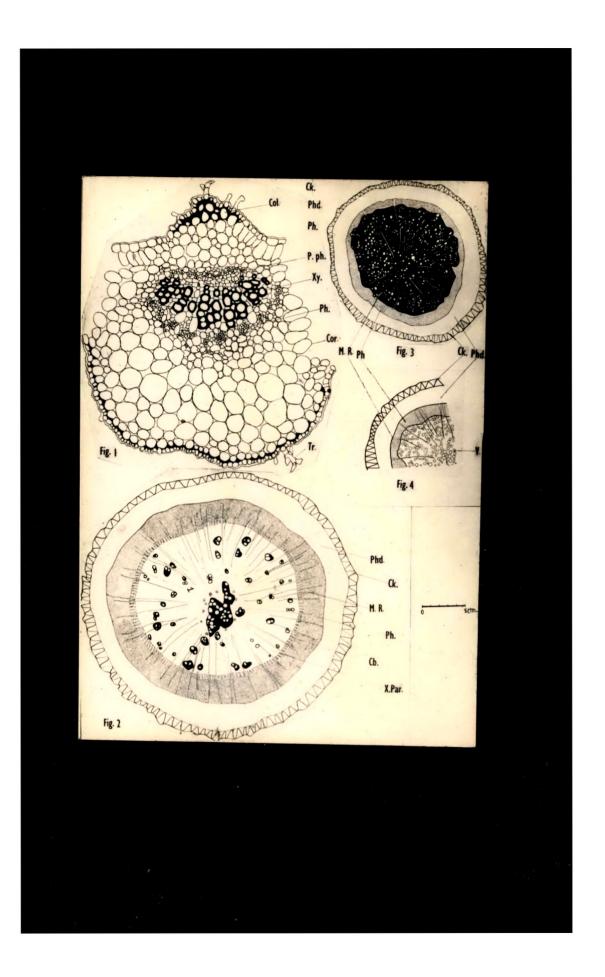


PLATE - XXIV

(Figs. 1 - 4: Withania somnifera Dunal)

Fig. 1 - T.s. midrib. x 290.

Fig. 2 - T.s. root market sample (diagrammatic). x 75.

Fig. 3 - T.s. old root (diagrammatic). x 28.5.

Fig. 4 - T.s. secondary root, old stem. x 4.

X-X-X-X-X-X-X

Cb. - cambium; Ck. - cork; Col. - collenchyma;

Cor. - cortex; M.R. - medullary ray; Phd. - phelloderm;

Ph. - phloem; P.Ph. - perimedullary phloem;

V - vessel; Xy. - xylem; X.Par. - xylem parenchyma;

consists of one layer of closely packed columnar cells studded with chloroplasts. Spongy tissue consists of 3-4 layers of big rounded cells with large intercellular spaces; isolated cells contain calcium oxalate prisms or clusters (Plate XXIII, 4).

Palisade is discontinuous over the meristele in the midrib (Plate XXIV, 1). Meristele consists of a radiating xylem towards the adaxial side and phloem towards the abaxial side. Perimedullary phloem is also present. Rest of the tissue of the midrib is parenchymatous except for the first 2-3 layers just below the upper epidermis which are collenchymatous. Calcium oxalate crystals are absent in the midrib.

Leaf constants: Stomatal index for the lower epidermis is 9-14 and that for the lower epidermis is 15-21. Palisade ratio is 3-6, rarely 7. Vein islet number is 10-16.

Stem (Plates XXV & XXVI):

Apical portion of the stem in transection shows a circular outline. Epidermis has developed a thin layer of cuticle and bears many multicellular uniseriate or branched trichomes similar to those of the leaf. Cells of the hypodermis, upto 2-3 layers show little thickening at the cell corners. Rest of the cortex is parenchymatous (Plate XXVI, 1).

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Withania somnifera Dunal

Epidermis still persists in case of the stem base though secondary growth has already set in. Cells of the epidermis show a layer of cuticle and bear trichomes (Plate XXV, 1). Periderm consists of 2-3 layers of cork cells, a layer of phellogen and 1-2 layers of phelloderm. Collenchyma forms the next zone of 3-6 layers of cells. Rest of the cortex is parenchymatous and 4-6 cells thick. A few scattered pericyclic fibres which may occur in small groups lie outside on the periphery of the secondary vascular tissues (Plate XXV, 1). Primary phloem consists of crushed groups of cells. Cambium consists of 2-4 layers of characteristic cells. Xylem is a comparatively bigger zone and consists of vessels, tracheids, fibres and xylem parenchyma. The vascular tissues are traversed by a number of uniseriate to multiseriate medullary rays. Permedullary phloem is present below the primary xylem groups on the periphery of pith (Plate XXV,1). The central pith is solid and parenchymatous. In the commercial drug which also contain stem bases, a similar arrangement of tissues is obserbed as seen above.

Root (Plates XXIV, XXV & XXVI)

Young root from the seedling shows diarch condition of the stele (Plate XXVI, 2). Soon cambium originates in the usual manner. Initially, after giving rise to a few vessels and tracheids, cambium mostly gives



PLATE - XXV

(Figs.1 - 4: Withania somnifera Dunal)

Fig. 1 - T.s. old stem. x 320.

Fig. 2 - T.s. young root. x 320.

Fig. 3 - T.s. old root. x 320.

Fig. 4 - Root maceration. x 320.

x-x-x-x-x-x-x

Cb. - cambium; Ck. - cork;

Col. - collenchyma; Cr. - sandy crystals

M.R. - medullary ray; Par. - parenchyma;

Ph. - phloem; Phd. - phelloderm;

P.fb. - pericyclic fibres; Tr. - trichome

or tracheid; V - vessel; Xy. - xylem.

out xylem parenchyma on its inner side. Thus, in a young root, one finds a small lignified xylem core in the centre while the rest of the xylem consists of xylem parenchyma traversed by a few small groups of vessels and tracheids only.

In an 1-2 months old root, 2-6 layers of cork cells protect the root externally. Cells of the cork are isodiametric, suberised and arranged radially. Phellogen is obscure. Phellodern is a broad zone consisting of 8-9 rows of tangentially alongated and somewhat radially arranged and compact cells. Phloem consists of the usual elements but phloem parenchyma are far more in number as compared with the rest of the elements. Cambium consists of 4-8 layers of tangentially elongated cells and it mostly gives out storage parenchyma on both sides. Secondary xylem is a wide zone consisting mostly of xylem parenchyma; a few scattered groups of vessels associated with tracheids and occasionally fibres as well, are embedded in it (Plates XXIV,2; XXV, 2; XXVI, 3,4 & 5). Xylem parenchyma are thin-walled and are arranged without leaving intercellular In the centre, primary xylem along with some secondary xylem together form a central woody core (Plates XXIV, 2: XXVI, 3, 4 & 5). Around the woody core, small groups of interxylary phloem are observed (Plate XXIV,2). Vessels occur singly or in groups of 2-4. They are lignified

and mostly show bordered pits with slit-like openings (Plate XXV, 4). Spiral and annular vessels are not altogether ruled out. Tracheids show oblique bordered pits and times one accompanied by fibres. Secondary medullary rays are having radially elongated cells and traverse the secondary vascular tissues.

Transection of the market drug shows practically the same arrangement of tissues as observed in 1-2 month old roots (Plate XXIV, 2).

TABLE - 29

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Measurements of root (young) elements

Elements		Length	* *	Width
Vessel	; !	176-203- <u>250</u> -315-326	μi	27-38- <u>50</u> -61-73 /
Tracheid	1	2 11- 296- <u>323</u> -276-400	M;	16-19- <u>21</u> -23-27 /
Fibre	í	420-507- <u>595</u> -717-770	μi	15-16- <u>18</u> -23-25 /4

Frequency of yessels and tracheid formation increases with age of the root. Thus, even in a young root of 4000 μ , 5200 μ diameter, though scattered in xylem parenchyma, vessels containing areas are bigger and closer to the cambium (Plate XXIV, 2). As the root become

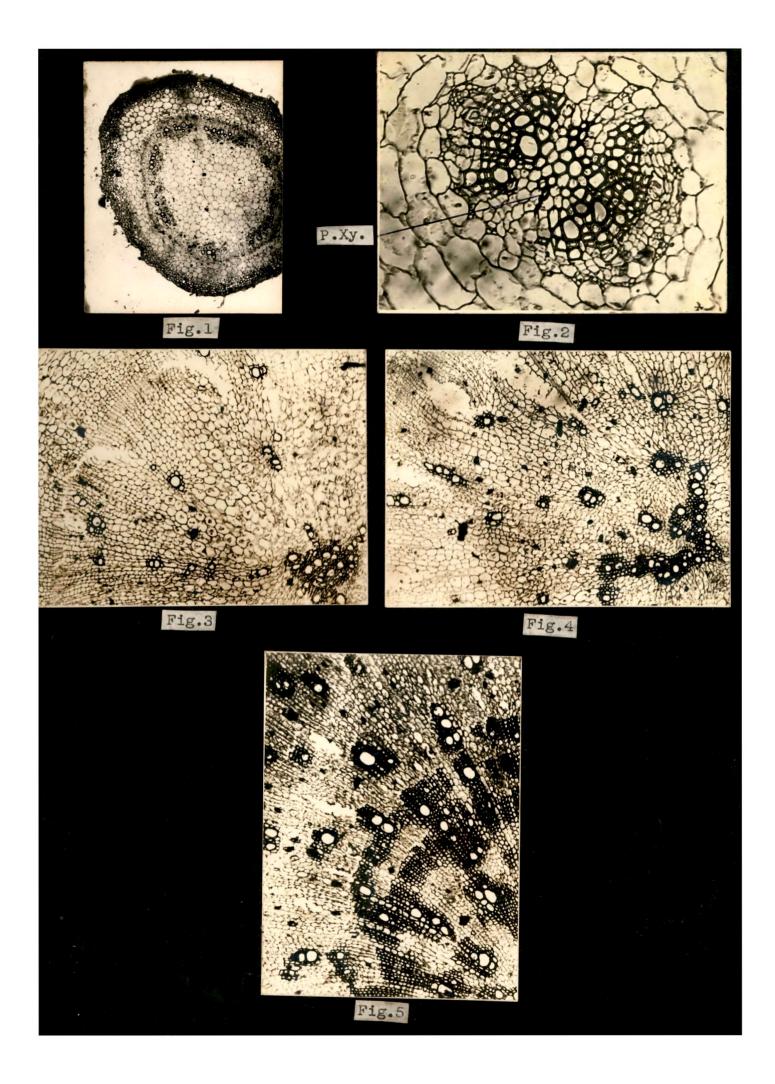


PLATE - XXVI

(Figs. 1 - 5: Photomicrographs of

Withania somnifera Dunal.)

Fig. 1 - T.s. young stem. x 48.

Fig. 2 - T.s. young root showing diarch condition of xylem. x 230.

Figs. 3, 4 & 5 - T.s. of young root from different positions: 3 - near apex;
4 - about the middle; 5 - near base. x 66.

x-x-x-x-x-x

P. Xy. - primary xylem.

progressively old, the soft and tuberous nature of the young root gradually diminishes and is being replaced by hard and woody nature. Thus, while examining roots of various sizes, it has been found that the parenchymatisation of secondary xylem is gradually being replaced by lignification (Plate XXVI 3, 4 & 5). During the process of lignification, the tissues in the centre are first to be modified; gradually the process extends towards the outer side.

In a root, about one year old, xylem consists mostly of lignified elements (Plate XXV;3). On its outermost side, it is protected by a few layers of cork. because most of the cork cells get exfoliated or get crushed. They are isodiametric and suberised. Phellogen of 2-4 diffuse layers may be observed. Phelloderm layers which are increased to about 20 are tangentially elongated, compact and radially arranged (Plate XXV, 3). Phloem which forms the outermost vascular tissue consists of the usual elements. Cambium consists of 4-5 layers of tangentially elongated Central wood is hard and forms a solid central core cells. Plates XXIV, 3; XXV, 3). Secondary xylem and phloem are traversed by uniseriate or more frequently by multiseriate medullary rays. Xylem parenchyma cells are few here as compared to their predominence in young roots.

Secondary roots of both young and old plants are woody. They show the same internal arrangement of

tissues as that found in an old root (Plate XXIV, 4).

Cell contents:

Starch: - Cells of the phelloderm, medullary rays, xylem parenchyma as well as all parenchyma cells of phloem are filled with starch grains in case of root; cells of the cortex, medullary rays, pith as well as parenchyma cells of phloem also contain starch grains. Starch grains are comparatively less in old root, secondary root and stem. In old root and secondary root, starch grains are chiefly located in phelloderm cells. The young roots, where starch is abundant, show bigger grains in the phelloderm cells. Grains are spherical or oval in shape, concentric and measure 4.8-9.5-15-36 μ in diameter. Some of these individual grains are pointed at one end or are flat. few possess a slit in the centre. Compound grains of 2-3 components are of frequent occurrence. Their contact surfaces are flat. Compound grains measure 7.8 - 14.5 - 23 μ in diameter.

<u>Calcium oxalate crystals</u>:

Some of the parenchyma cells especially those of phelloderm and xylem parenchyma of root and cortex and pith of stem contain black granular masses of microspheroid crystals of calcium oxalate (Plate XXV,1,2 & 3)

These dark masses are insoluble in acetic acid but soluble in hydrochloric acid and with 75% sulphuric acid forms acicular crystals. Thus, the dark masses are identified as calcium oxalate crystals.

Alkaloids:

Most of the cells of phelloderm, medullary rays, phloem parenchyma-like all parenchyma cells of the secondary xylem in case of young tuberous roots, are filled with alkaloids which is determined by Wagner's test.

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Withania somnifera Dunal is a perennial undershrub, and young tuberous roots and older roots have machinesical histological distinct and different macroscopic and microscopic characters.

It can be seen that, generally in commerce, young tuberous roots are used and it would be desirable to mention age, season of collection of the drug and percentage of active chemical constituents in the I.P. monograph.

Dutta and Mukerji (1960) and I.P. (1955) mention absence of calcium oxalate crystals. Neuwald

and Loges (1956) only mention the presence of dark granular masses in parenchymatous cells of the root. Thus, the correction as regards presence of calcium oxalate crystals and a detailed revised monograph which naturally includes the stem base should be prepared in view of the above work.

SUMMAKY

Leaf of <u>Withania somnifera</u> is dorsiventral. Epidermis bears characteristic candelabra as well glandular trichomes. Stomata are both of ranumculaceous and cruciferous types. Vascular bundle of the midrib shows perimedullary phloem. Cells of the mesophyll contain calcium oxalate prisms and clusters while the same are absent from the midrib.

Young stem shows many vascular bundles. Old stem shows the presence of secondary growth. Perimedullary phloem and pericyclic fibres are present.

Young root is soft and tuberous and shows extensive development of xylem parenchyma; vessels, tracheids and fibres occur as small scattered groups embedded in parenchyma. With the advance in age, the soft and tuberous nature of young root becomes hard and woody. This is achieved by the modification of parenchymatous tissue into

a lignified one. Old root as well as secondary roots is completely woody.

Stem and root contain abundant starch. Isolated parenchymatous cells of both the stem and the young root contain microspheroidal crystals of calcium oxalate.