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An Anatomical Study of Cicer Arietinum

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Cicer

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AN ANATOMICAL STUDY

OF

CICER ARIETINUM

A Thesis

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History of the problem. Cicer arietinum, the only economically important member of the genus Cicer, is found cultivated today in many parts of the world. The regions of its chief importance are Egypt, Southern Europe, Asia, Africa, Central America, Chile and Mexico. The seed is not extensively grown in the United States, however, its use here in industry is considerable. In 1933 the United States imported fourteen million pounds and in 1934 eleven million pounds.

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The very early history of the chick pea, Cicer arietinum, is obscure. The earliest certain evidence of its antiquity is found in the writings of Theophrastus and Dioscorides. The scarcity of archeological evidence for its age is seen in the fact that no remains of the chick pea have been found among the ancient Egyptians and it is absent from the lake

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dwellings of Switzerland, Savoy and Italy. The chick pea is believed to have been unknown to the Hebrews as there is no mention of it in the Bible.[2] DeCandolle states it was thought that the "ketsech" mentioned by Isaiah in the Old Testament was perhaps the chick pea, "but this name is generally attributed, though without certainty, to Nigella sativa or Vicia sativa. As the Arabs have a totally different name for the chick pea, 'omnos' or 'homos', which recurs in the Kabyl language as 'hammez', it is not likely that the 'ketsech' of the Jews was the same plant." DeCandolle, in conclusion, says "these details lead me to believe that the species was unknown to the ancient Egyptians and the Hebrews."[3]

No records are present of early cultivation of the plant in China, Japan, Australia or the United States.[4] Ames assumes that Cicer arietinum was cultivated south of the Caucasus before 1500 B.C. He states that the plant must be very old because it has not been found in any of its wild forms.[5]

The chick pea is a member of the Leguminosae family. It is further classified in the subfamily Papilionatae, tribe Vicieae, genus Cicer and species arietinum.

The Greeks called the chick pea "erebinthos" and the Romans knew it

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2 Oakes Ames, Economic Annuals and Human Cultures (Cambridge Mass.: Botanical Museum of Harvard University, 1939), p. 49.

3 Alphonse DeCandolle, The Origin of Cultivated Plants (London: Kegan Paul, Trench Company, 1884), pp. 323-325.

4 N.K. Iyengar, "Cytological Investigation on the Genus Cicer," Annuals of Botany, 3: 271-305, 1939.

5 Ames, loc.cit.

as "Cicer". "The botanical specific name arietinum owes its origin to a not altogether fanciful resemblance of the seed, when first forming in the pod, to a ram's head."[6] The common name for Cicer arietinum in India is "Bengal gram" or simply "gram."

Sarin and Qureshi state that "gram is by far the most extensively consumed of all cattle feed in India." The chick pea is also used "by men, especially those of the poorer classes."[7]

The plant itself is seldom used for forage because of an exudate of oxalic acid. This acid, which is injurious to cattle is collected by the Ryots - a group of natives of India who cultivate the soil - and is used instead of vinegar in their curries. The natives also eat the seeds in their cakes and curries.[8]

Sarin and Qureshi thought that the plant might have wider use as a dietary constituent if the starch could be extracted. In the process of extraction they found that Cicer arietinum contained 43.5% starch. One of the by-products of this process consisted of a residual pulp that is of value as cattle feed. The second by-product was water. The extracted starch proved to be useful in textile sizing. "Tests have shown it suitable for giving a light finish to cotton, wool and silk cloth."[9]

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6 Iyengar, op. cit., pp. 271-305

7 J. L. Sarin and M. H. Qureshi, "Starch from Gram," Industrial and Engineering Chemistry, 30:1318-1319, 1938.

8 Colonel H. Drury, Useful Plants of India (Madras: Higgenbotham and Company, 1873) p. 134.

9 Sarin and Qureshi, loc. cit.

As to other uses of the plant and seeds Drury states that medicinally the chick pea is used as a refrigerant in fevers. For the treatment of dysmenorrhea (painful menstruation), the Portuguese place the plant in hot water and have the patient sit over the steam. The plant has also been used in cases of dysuria (difficult urination).[10] Hill relates that the seeds are sometimes employed as a substitute for, and as an adulterant of, coffee.[11]

Statement of organization. The material is organized to cover (1) methods and procedure, (2) gross morphology, (3) primary and secondary root, (4) root-stem transition, (5) primary and secondary stem tissues, and (6) leaf primordia.

The chapter on methods and procedure includes a description of the conditions under which the seedlings were grown, the percentage of germination, the possible factors influencing such a percentage and the histological technique employed in the preparation of the material for study.

Under the heading of gross morphology will be found a description of the germinating seed and the relation of the cotyledons to the main axis of the embryo. Further data are also included from studies of the morphology of roots, stems and leaves at maturity.

The chapters on the primary tissues of the root and the stem deal with the ontogeny of the histogens, the stelar arrangement of the primary axis

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10 Drury, loc. cit.

11 Hill, loc. cit.

and the origin and development of the secondary tissues.

A study of the mesocotyl includes information concerning the transition from a tetrarch condition in the root to the dictyostelic stem. The leaf primordia deal with the leaf origin and arrangement.

CHAPTER II

REVIEW OF THE LITERATURE

In a paper dealing with the developmental anatomy of Cicer arietinum Sister Raphaelis Gehlen states that the chick pea root is tetrarch, having four xylem arms and four intermediate phloem areas and a central pith. The pericycle occupies a single layer of cells over the phloem, but is double over the xylem arms.[12]

Gehlen describes the xylem of the primary root as differentiating from the meristematic tissue more rapidly than does the phloem.[13] Shortly after this change occurs the protoxylem and the metaxylem shift so that the protoxylem becomes internally located in the bundle and the metaxylem assumes an external position. This arrangement of xylem tissue is continued upward in the root to a point several millimeters below the cotyledonary node. Here the phloem areas, which have enlarged and elongated, become situated outside the xylem areas instead of between them.

There are now four xylem areas capped by four phloem regions. Eight collateral bundles result from a division of the four xylem and phloem areas.

In the transition area which, according to Gehlen, occurs about three millimeters below the cotyledonary node, the eight bundles of the

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12 Sister Raphaelis Gehlen, "Stelar Anatomy of Cicer arietinum and Glottidium floridanum," American Journal of Botany, 16:781-788, 1929.

13 Gehlen, loc. cit.

14 Gehlen, loc. cit.

root are rearranged to form the six bundles found in the stem. This change from eight root bundles to six stem bundles begins with the outward passage of polar bundles 1 and 2. These two bundles unite to form a cotyledonary trace. Shortly afterwards bundle 5, at the opposite side of the hypocotyl, splits to form two bundles. The half of bundle 5 nearest to bundle 6 converges with that bundle to form a petiolar trace.

The remaining half of bundle 5 forms the fifth vascular strand. To complete the normal vascular bundle number 8 divides. These six bundles, 3, 4, 5, 7, 8, 8a, enter the stem. The transition occurs in a region less than six millimeters in length.

Harris and co-authors in their paper on the Vascular Anatomy of the Dimerous and Trimerous Seedlings of Phaseolus vulgaris show the transition differences between these two groups.[15]

In the dimerous seedling, the root contains four bundles - two in the cotyledonary plane and two in the intercotyledonary plane. These bundles, v-shaped with the protoxylem appearing at the apex of the v, surround a large pith area. The bundles each bifurcate to form a total of eight bundles and "typical stem structure with the protoxylem in an endarch position begins to be assumed." Each pair of bundles formed from the division of one of the four original bundles is known as a primary double bundle. "Thus the level of transition from root structure to stem structure is low, being practically at the base of the hypocotyl."[16]

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15 J. A. Harris, E. W. Sinnott, J. Y. Pennypacker and G. B. Durham, "The Vascular Anatomy of Dimerous and Trimerous Seedlings of Phaseolus vulgaris," American Journal of Botany, 8:63-102, 1921.

16 Harris, loc. cit.

Members of the four pairs soon separate forming eight bundles which persist through the hypocotyl to the cotyledonary node.

At the cotyledonary node the "two members of each of the two original pairs of bundles in the cotyledonary plane become widely separated and each member fuses with the adjacent member of the intercotyledonary pair." This produces four bundle aggregates each of which breaks up into three parts. "The lateral members of each group of three which is in the cotyledonary plane approach the corresponding bundle of the next group of three, and these two strands become the cotyledonary traces and enter the base of the cotyledon. The lateral members of each group of three, which is in the intercotyledonary plane approach the corresponding bundle of the next group and fuse with it."[17]

Above the cotyledons the six remaining bundles approach one another closing the cotyledonary gaps and forming a ring, the members of which almost immediately divide. These twelve bundles are continued throughout the first internode.

In the trimerous seedlings with three cotyledons and three primary leaves, the transition is accomplished in a similar manner. The process is slightly more complex because of the increased number of bundles concerned in the transition.

Foster in his paper on leaf differentiation states that the leaves originate from the "outer cell layers at the side of the shoot growing point." The actual emergence of the leaf primordia is the result of the expansion of the apex to form one or more axial portions of the shoot.

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17 Harris, loc. cit.

"The number of strands present.....is directly related to the phyllotaxis and the relative width of insertion of the primordia."[18]

The primordial growth soon forms the lamina, the petiole and the stipules of the leaf. Following the petiole-midrib specialization the lamina is developed. The marginal initials of the lamina begin growth, but are soon replaced by surface growth which eventually leads to the maturation and differentiation of the laminal tissues.

Bell describes the ontogeny of the primary axis of Soja max. He has found that the tetrarch radial protostele is changed in the transition area into an endarch collateral dictyostele of eight bundles. This is accomplished by the divergency of the double cotyledonary traces from the two vascular groups of the root.[19]

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18 Adriance S. Foster, "Leaf Differentiation in Angiosperms," Botanical Review, 2:349-372, 1936.

19 Willis H. Bell, "Ontogeny of the Primary Axis of Soja max," Botanical Gazette, 95:622-635, June, 1934.

CHAPTER III

METHODS AND PROCEDURE

In the cultivation of Cicer arietinum for study three methods were used, each with varying degrees of success. In the first method the seeds were sown in earthen pots in a clay soil. The temperature during the growing period was nearly constant at 25° centigrade. The seeds were watered when sown and several times thereafter. In this case the germination time was from ten to fourteen days and the percentage of germination was less than twenty-five per cent.

The second method varied somewhat from the first. The culture jars in which the seeds were germinated were disinfected with chlorinated lime as a preventative measure against fungal development. The temperature, in this case as in the first, was fairly constant at 25° centigrade. The moisture needed for germination was supplied by wet paper towelling in the bottom of the culture jar. The germination time was from ten to fourteen days and the percentage of germination was approximately forty per cent.

Method number three consisted of sowing the seeds out-of-doors in a loam soil. These seeds were watered more frequently than those in either of the other two methods. In this case the temperature, cooler at night, varied from 13°-32° centigrade. The growing period was seven to ten days long and the percentage of germination was about thirty per cent.

The percentage of germination was low in all three cases. Possibly

the most important factor influencing the low germination was the viability of the seeds. This conclusion has been reached because the other factors during germination, such as temperature and amount of moisture, were constant enough to eliminate the possibility of their causing such a low percentage.

Histologic Technique. The histologic technique employed in the preparation of the material for study was similar to that given by Chamberlain.[20] Dr. Chamberlain's method has been slightly altered to suit the needs of this particular plant. The following method is used:

Killing and fixing

1. Weak killing solution of chromo-acetic acid, 1-3 days
2. Material washed in cold running water, 1 day

Hardening and dehydrating

3. Alcohol series 10%, 20%, 30%, 40%, 50%, 60%, 70%, two grades a day; 85%, 95%, twenty-four hours; absolute alcohol, twenty-four hours changed two or three times

Clearing and embedding

4. 1/3 xylol - 2/3 alcohol, 1/2 xylol - 1/2 alcohol, 2/3 xylol - 1/3 alcohol, twenty-four hours; pure xylol, twenty-four hours changed two or three times
5. 1/2 xylol - 1/2 paraffin, six to twelve hours; pure paraffin pouring off the first bath immediately to re-

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20 Charles J. Chamberlain, Methods in Plant Histology (Chicago: Chicago University Press, 1928), 349 pp.

CHAPTER IV

GROSS MORPHOLOGY

The mature chick pea is an herbaceous annual legume, growing to a height of from one to two feet. The compound pinnate leaves are composed of from nine to thirteen leaflets. Each leaflet is ovate, serrate and equal, with stipules lanceolate and somewhat toothed. The grayish white appearance of the plant is due to the presence of fine pubescence on the leaves and stem. The plant matures in about ninety days and "is well adapted to arid and semi-arid regions."[21]

The single white flowers are rather inconspicuously borne in the axils of the leaves. The calyx is five-lobed surrounding a papilionaceous corolla. After pollination the flowers mature rapidly into a short pod bearing one or two seeds.

When mature the seeds are gibbous, mucronate and are usually white or straw colored. There are varieties of Cicer arietinum existing which produce seeds varying in color from red to yellow and from almost black to brown.

The root system of Cicer arietinum is a tap root. There is one, sometimes two, mature roots with many lateral roots and root hairs. The growth and development of the root is rapid during the early life of the plant. Before the epicotyl becomes fully developed and mature

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21 Albert F. Hill, Economic Botany (New York: McGraw - Hill Book Company, Inc.,) pp. 354-355.

the root often doubles or triples this structure in length.

The mature chick pea seed consists of a hard, woody seed coat, two cotyledons, a hypocotyl and a plumule; endosperm is lacking. The seed coat is composed of several layers with an outer palisade layer of 70-120 microns in width. These palisade cells are thin, elongate and are very closely packed together. Underlying the palisade cells is a layer of columnar cells, loosely but regularly arranged. Each cell of this second layer is separated from the next one by an air chamber. The columnar cells average forty microns in length. A third layer consists of parenchymatous cells and varies widely from three to four irregular layers to many layers. (fig. 15)

The cotyledons are composed of many large cells filled with starch granules. These seed leaves serve, in the absence of endosperm, as food storage organs.

In the mature seed the hypocotyl, that part of the primary axis between the primary root and the cotyledons, is about two to three millimeters long. The hypocotyl is laterally joined by two bridges of tissue, one bridge uniting with each of the cotyledons. Joining the hypocotyl at the cotyledonary node is the plumule.

Growth in the hypocotyl consists of increase in the number of cells and increase in size, causing the emergence of the primary root through the seed coat. Growth of the primary root continues for several centimeters before the plumule begins to grow. The absence of hypocotyledonary elongation causes the cotyledons to remain within the soil. They exist until after the plant has produced photosynthetic leaves and is able to manufacture food.

CHAPTER V

THE PRIMARY ROOT

The development of the primary root of Cicer arietinum corresponds to that recognized by Janczewski as the fourth angiosperm type.[22] Hayward citing Janczewski stated that in the fourth angiosperm type "there is a common meristematic zone which extends across the apex of the root, and sharply delimited histogens are lacking."[23] Cells destined to become root cap differentiate from the apical portion of the meristem early in the root development. The stele and cortex of the root are formed from the differentiation of the central cells away from the apex, while the epidermis results from the division of the peripheral cells of the meristem.

The apical cells of the meristem cease rapid division and enlarge, differentiating into the first formed tissue - the root cap. This is followed by a division away from the apex of the peripheral meristematic cells to form the dermatogen. In the region immediately above the meristem, which is 135-140 microns wide, the dermatogen is two to several layers thick. These several layers merge into one layer higher in the root.

Several microns above the meristematic region the cells are seen to differentiate into a central core, the plerome, and into a region

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22. E. Janczewski, "Recherches sur l'accroissement terminal des racines dans les Phanerogames," Annals Science Naturales, 20:162-201, 1874.

23. Herman E. Hayward, Structure of Economic Plants (New York: The Macmillan Company, 1938) 674 pp.

intermediate to the plerome and the dermatogen, the periblem. At 240 microns the cells of the periblem are large, loosely spaced and nucleate, while the plerome cells are smaller and less deeply stained.

The vascular tissue of the root begins development as a group of centrally located procambial cells. The protoxylem cells are the first formed from the procambial tissue. The protoxylem tissue, for the first 50-100 microns of its development, forms a tetrarch radial protostele (fig.1). This condition changes shortly as the central protoxylem cells break up to form the protoxylem parenchyma (fig.2). The primary xylem development is endarch with the protoxylem differentiating centrifugally. The metaxylem of the primary xylem is formed on the inner face of the xylem arms (fig.2).

While the primary xylem development is taking place, the primary phloem and the pericycle are forming. Approximately 600 microns from the root tip the primary phloem first appears. One and one-half millimeters from the tip the protophloem is well established and the pericycle surrounds the vascular tissue as a single row of cells.

The primary phloem begins growth as a small group of procabbial cells. This tissue enlarges as growth continues and finally there are phloem fibers differentiated on the outer edge of the primary phloem (fig.4). The primary phloem consists of sieve tubes, companion cells and phloem parenchyma.

The root as it now exists (fig.3), with four xylem areas and four intermediate phloem areas, undergoes no further change for several millimeters except the addition of new cells. Near the transition area other changes

occur. These are given under the chapter on root-stem transition.

With the origin of secondary growth in thr root the vascular tissues become united around the central parenchyma cells. The secondary xylem if formed prior to the development of the secondary phloem. The meristematic cells of the procambial tissue between the primary xylem and the primary phloem have by this time become cambial cells. Previous to the development of the secondary tissues the cambial cells, while present throughout the xylem and phloem development, remain inactive. At the time of secondary thickening the cambial cells become active and begin to divide, each division resulting in the formation of new xylem or phloem cells from one half of the split cell. The other half remains cambial and continues to cut off the secondary tissue.

Along with fascicular development the cambium between the vascular bundles begins to divide with the result that the four xylem areas become merged with each other to form a continuous circle. The secondary xylem development in the root is more rapid than the secondary phloem formation.

The secondary phloem is cut off on the outer side of the cambium. As this is happening the phloem fibers formed as a part of the primary tissues have been pushed outward. These groups of fiber cells never unite with each other, but remain as separate areas throughout the root.

The tissues of the root following secondary thickening are (1) epidermis - one cell layer thick, (2) cortex - a group of large parenchymatous cells, (3) endodermis - a not too well defined layer of cells immediately outside the pericycle, (4) pericycle - a single layer of cells defining the outer boundary of the stele, (5) phloem tissue - in-

cluding phloem fibers, companion cells and sieve tubes, (6) cambium - one layer thick, (7) xylem tissue - vessels and tracheids and (8) pith.

The lateral roots of Cicer arietinum are tetrarch as is the primary root. The two root types are identical except for the absence of xylem parenchyma in the central region of the lateral root. This area in the lateral root is occupied by the protoxylem and is made up of large conducting vessels.

These lateral or secondary roots arise from the pericycle of the main root. The pericycle outside one of the xylem arms of the main axis becomes meristematic and begins to cut off new cells which push outward through the cortex of the root. This growth is continued in a manner similar to that of the primary root with the pericyclic tissue forming the meristematic region of the root. After meristematic growth is well established, the xylem elements begin to develop within the lateral root. This xylem development is followed shortly by the formation of the phloem tissue in the lateral root. The vascular system of the lateral root is thus in direct contact with the conducting elements of the main axis.

CHAPTER VI

THE ROOT-STEM TRANSITION

The change from the vascular tetrarch arrangement in the root to the six, later ten, bundle dictyostele stem occurs in a region about three to four millimeters long. Just below the transition area the vascular tissue of the root is arranged in four xylem points with four intermediate phloem areas. The sylem areas are triangular in appearance with the apex of the triangle pointing outward toward the cortex of the root. On the inner face of the xylem triangle the metaxylem is developed. The intermediate phloem areas are made up largely of protophloem with the phloem fibers forming early on the outer edge of the area.

About one and one-half millimeters below the cotyledonary node the metaxylem on the inner face of the xylem area begins to split (fig.3). The metaxylem moves outward until four more or less oval xylem areas are formed with the metaxylem capping the ends of each area and with the protoxylem now centrally located. The four xylem areas then split into eight xylem areas, all still retaining the same position.

At one millimeter below the cotyledonary node the eight xylem regions shift so that the protoxylem is turned inward while the metaxylem faces outward. As this change in position is occurring, the phloem areas split and move to a position peripheral to each of the eight xylem areas. There are now wight xylem and eight corresponding phloem areas (fig.5). The phloem areas composed of phloem fibers and protophloem, are separated

from the primary xylem by several layers of cambial cells.

These cambial cells, now functioning tissue, were formed early in the root development as a part of the procambial strand. During the changes in the xylem and phloem of the root the cambial cells are inactive, but by the time the four vascular bundles have split to form eight bundles, the cambiam is complete and functioning. The cambium is wider at the bascular bundles than it is between them because of greater growth activity in this region.

The eight bundles continue upwards to a point about 275-300 microns from the cotyledonary node. At this point the bundles rearrange themselves sending off traces to the cotyledons. The method of transition in Cicer arietinum is fairly constant with only slight variations occurring.

The bundles are arranged so that there are two in each of the cotyledonary planes and two in each of the intercotyledonary planes. Bundles A and B, E and F are cotyledonary bundles while C and D, G and H are found in the intercotyledonary plane (fig.5).

The first step in the transition is the division of bundle D to form bundles D and D_1. This is shortly followed by the formation of two bundles from bundle C (fig. 6). Bundles E and F next divide with F bisecting before E (figs. 7 and 8). A few millimeters above this bifurcation of E and F, bundles C_1 and D unite as do D_1 and E (fig.9). E_1 departs as a cotyledonary trace. The cotyledonary bundle B divides (fig.10), and B_1 moves to join bundle C (fig.11). Bundles F and F_1 which have previously bisected now pull apart, and F becomes the cotyledonary partner of E_1.

F_1 forms the third intercotyledonary bundle. The cotyledonary bundles are Now A and B, E_1 and F, while the intercotyledonary bundles are B_1C, C_1D, D_1E, F_1, G and H (fig.11). The cotyledonary bundles now depart as cotyledonary traces leaving six intercotyledonary bundles which enter the stem (fig.12). In the transition process bundles A, G and H did not divide, A passing into the cotyledon and G and H forming two of the stelar bundles of the stem.

In one series of slides the changes involved in the transition area are less complex in arrangement than those cited above. The bundle arrangement is similar, with eight bundles, A-H, in the root. A and B, E and F the cotyledonary bundles, pass as two traces to each cotyledon without dividing. Of the remaining intermediate cotyledonary bundles D and G bifurcate forming two bundles which, with bundles C and H make up the normal stem compliment of six bundles.

Both of these cases differ slightly from the reorientation of the vascular tissue given by Gehlen. She found that only one of the cotyledonary bundles, B, divides. The second half of bundle B forms the third intercotyledonary bundle on one side, and the lacking bundle in the opposite group is formed from a division of intercotyledonary bundle H.

The number of vascular strands from the root is in all cases constant as is the number passing to the stem, but the manner in which they reorient seems to differ with the individual plant.

All the changes occurring in the transition area take place in the region below the cotyledonary node. After the two traces pass to each

cotyledon, there is no further division of bundles, and the six bundles already present become the vascular tissue of the stem. Occasionally there are more than six bundles after the cotyledonary traces depart, in which case there is a divergence of bundles to form the six passing into the stem.

CHAPTER VII

THE STEM

In considering the ontogeny of the stem it is well to note the structure of the embryo before germination. The epicotyl, or that part of the embryo above the first node, is the mature stem in miniature. The epicotyl consists of the stem meristem surrounded by two perfectly formed minute leaves, several developing primordia and two stipules.

The upper-most region of the stem, known as the apex is structurally a group of small, large-nucleate cells possessing the power of rapid division. This apex, like the meristematic region of the root, is responsible for the formation of all primary and, indirectly, all secondary tissues of the stem.

At the time of germination the meristematic cells, occupying 15-30 microns of the conical stem tip, begin to divide rapidly both anticlinally and periclinally. The anticlinal divisions are responsible for the formation of the central group of cells in the undifferentiated stem, known as the corpus, while the periclinal divisions account for the tunica or external layers. Elongation and maturation of the cells behind the meristematic region cause an increase in stem length. During the process of growth the meristematic tip has been pushed farther and farther away from the first or cotyledonary node by the intervening cells.

The vascular tissues of the mature stem are connected to the meristem by strands of cells which have retained their meristematic activity,

while the surrounding cells have ceased to divide and have begun to elongate. As the provascular strands are differentiated away from the apex, there is protoxylem formed on the inner face of the strand. Developing almost simultaneously with the protoxylem is the primary phloem. Between these two regions of newly differentiated tissue the provascular cells still retain their identity. This provascular tissue will eventually give rise to the cambium of the mature stem. The vascular strands, known as collateral bundles, now consist of an inner xylem area, a median procambium and an outer phloem region. The collateral bundles, ten in number, continue down the stem dividing and anastomosing with the vascular strands of the leaves.

As the stem matures secondary thickening is added to the already present stem tissues. The meristematic procambial cells form a single layered ring of cambium from which is differentiated secondary xylem and phloem. The cambial cells divide forming two daughter cells, one of which will remain cambium, the other becoming xylem of phloem depending upon which side of the cambium the cell was cut off. The secondary cells, unless crowded, tend to arrange themselves radially in rows from the cambium.

The primary vascular tissues of the stem cause the stele to appear dictyostelic, but with the development of secondary thickening the stem is transformed into a siphonostele. With the addition of secondary tissue in the vascular bundles a complete ring of tissue is formed. This is accomplished by the augmentation of new cells from the cambium of each bundle, and by the development of the interfascicular tissue. The cambial

layer is continuous around the stem, however, in the formation of the primary tissue only the fascicular cambium is involved. With the addition of secondary thickening the interfascicular cambium also becomes functional. Interfascicular development results from an increased activity of the cambial cells between the bundles. Hence a combination of fascicular and interfascicular growth causes the secondary tissue to be formed in a ring.

The mature stem of Cicer arietinum is oriented in the following manner; the outer layer of the stem is the epidermis, which is usually one, sometimes two layers in thickness. A thin cuticle covers these epidermal cells. Inside the epidermis is found the cortex - a region composed of food storage parenchyma cells. Moving inward from this point are found endodermis, pericycle, phloem, cambium, xylem and pith. After secondary thickening occurs the endodermis and pericycle appear broken up and are rather indistinct. The phloem fibers of the primary phloem have been pushed to the outer edge of the vascular cylinder and now lie immediately inside the pericycle. The secondary phloem, consisting of sieve tubes and companion cells, occupies the position between the primary phloem and the cambium. On the inner side of the cambium in located areas are the secondary xylem and the primary xylem respectively. The center of the stele is occupied by the pith. The rate of formation of secondary xylem exceeds the rate of secondary phloem development with the result that the amount of xylem is often double or triple the amount of phloem produced.

In the chapter on the root-stem transition the number of vascular bundles in the stem was given as six. This number persists only a short time,

then because of a splitting of bundles G and C, D to form G_1 and $(C_1D)_1$ the number is increased to eight. A few microns above this previous division the same bundles cut off two new members on the other side with the result that there are now ten bundles ascending the stem.

The manner in which the stipular and petiolar traces leave the stem is identical for each leaf. The ten bundles are lettered from A-J. Bundles A, then G divide increasing the number to twelve. Following this division bundles B, D and F begin to move outward toward the epidermis. While this is taking place, bundles C and E divide forming C_1 and E_1. Bundles B, D and F have now reached their outermost limits and the tissues between these three bundles and the stem stele begins to separate. The two bundles B and F are the traces to the stipules, and trace D becomes the vascular tissue to the petiole of the leaf. While the stipular and petiolar traces are still united with the main axis, bundles C_1 and E_1 move outward following the petiolar trace, and finally unite with each other to form the floral bud traces.

The floral bud separates from the main axis on its inner side and from the petiolar trace on its outer side. Meanwhile the tissue between the stipular traces and the petiolar traces begins to constrict. Before the stipules are separated from the leaf petiole, the stipular traces divide and send a bundle into the petiole of the compound leaf. The petiole, after separating from the stipules, is composed of three bundles--one median and two lateral bundles.

The median trace is persistant throughout the leaf, but the two lateral traces pass to the first two leaflets. The traces to the remaining leaflets

are cut off the median bundle alternately as the bundle passes up the stem to end in the apical leaf.

The normal vascular complement in the main axis is regained by the division of bundles C and E to form C_2 and E_2, which fuse almost immediately. There are now in the stem bundles A, A_1, C, C_2E_2, E, G_1, G, H, I, J. The same process is repeated for the next of the alternate leaves, this time the stipular bundles are G and A, and the petiolar trace is formed from I.

CHAPTER VIII

LEAF PRIMORDIA

The development of the leaf primordia, like the stem, begins before germination. In the development of the apical meristem of the stem certain cells of the conical tip become initial cells for the leaf. These cells are formed, not haphazardly, but in a definite pattern following the phyllotaxic arrangement common to this species. Since the leaves are arranged alternately in the mature plant, a primordium is developed first on one side of the apex and then on the other.

The cells of the leaf primordium continue dividing for a short time before the primordia of the leaflets begin to form. As the apical meristem of the leaf grows away from the main axis, the primordia of the leaflets are initiated with the primordia at the base of the leaf developing first, and the apical leaflet primordium at the tip being last formed. This type of development is acropetal with the primordial succession going from the base to the tip.

Upon germination the leaf and leaflet primordia renew the growth that was started early in the embryological development. The apices grow rapidly for a short time and then growth ceases. Development of the leaf is continued by means of intercalary growth.

In the early stages of development the leaf in cross section shows three regions; the epidermis, the mesophyll and the provascular tissue. The epidermal structures found on both the stem and the leaves are interesting to note. There are two types of epidermal hairs formed; one a multicellular, glandular hair, the other a simple hair.

The glandular trichome is living tissue composed of from four or five to many cells (fig.14). The epidermal cell from which the hair arises is enlarged to two or three times the size of the adjacent epidermal cells. From this basal cell is formed a filament of cells each possessing a nucleus and a somewhat dense cytoplast. At the very tip of the hair is found a knob of several cells resembling a sporangium of a fern. This structure is usually composed of four or more cells.

The simple hair is less complicated in structure than the glandular hair, (fig.13). This trichome, like the glandular, is also supported by a single epidermal cell. Immediately above the epidermal cell is a small, nearly spherical cell. The remainder of the hair is a single, elongate, tapering cell which appears to be devoid of a definite nucleus.

At maturity the cross section of the leaf shows further differentiation (fig.16). The mesophyll of the embryonic leaf remains undifferentiated and can be distinguished as a group of parenchymatous cells. Such provascular tissue as is present undergoes relatively few changes upon reaching maturity. Some of the cells of the dermatogen divide to form the guard cells surrounding the stomata. These guard cells when mature are nearly spherical and possess, on the outer edge, two ridges of tissue. The ridges appear to act as a seal on the guard cells when they are closed. The number of guard cells present in the epidermis is greater on the lower side of the leaf than on the upper. Covering all the epidermal tissues is a relatively thick cuticle.

The mesophyllic tissue present in the immature leaf differentiates into two types of mature cells. On the upper side of the leaf the meso-

CHAPTER IX

SUMMARY

This investigation was made for the purpose of studying the developmental anatomy of the primary axis of Cicer arietinum.

Cicer arietinum, the only economically important member of the genus Cicer is found in Egypt, Southern Europe, Asia, Africa, Central America, Chile and Mexico. Its original home is thought to have been in the region south of the Caucasus where it probably became established about 1500 B. C.

The chick pea is a member of the family Leguminosae, subfamily Papilionatae, tribe Vicieae, genus Cicer and species arietinum.

The plant has been used in various ways, as human and cattle feed, in textile sizing, by the natives of India in cakes and curries, as a medicine and as a substitute for coffee.

Three methods were used in growing the material for study. In the first method the seeds were grown in earthen pots with a low percentage of germination resulting. The second method tried was to sow the seeds on moist paper in culture jars. The third method consisted of sowing the seeds out-of-doors in a loam soil. Results in the last two methods surpassed those of the first attempt.

The plant is an hervaceous, annual legume growing to a height of one to two feet. The flowers are white, papilionaceous and are borne in the axils of the pinnately compound leaves. The seeds, formed in pods, are seldom more than two or three in number per pod. At maturity the seeds

consist of a hard, woody seed coat, two cotyledons, a hypocotyl and a plumule. There is no endosperm present.

The primary root corresponds to Janczewski's fourth angiosperm type and is composed of plerome, periblem and dermatogen. Growth of the vascular tissue begins as a group of provascular cells which differentiate into a central xylem region. This xylem region is of short duration with the central cells forming xylem parenchyma. Phloem is formed from the provascular tissue after the xylem arms have separated. Metaxylem forms on the inner face of the xylem arms and then swings outward, finally reaching a position external to the protoxylem and internal to the procambial tissue. The division of the four xylem arms, during the metaxylem shift, to form eight is accompanied by a division of the phloem areas so that below the hypocotyl there are eight xylem regions internal to the eight phloem areas.

In the transition area the eight collateral bundles, A - H, bifurcate and reorient passing into the stem as six vascular strands. Bundles A, B, E and F are the cotyledon bundles and C, D, G and H are the intercotyledonary members of the stele. The first bundle to divide in the transition area is bundle D, this is followed by C. E and F next divide, and shortly above this bifurcation, bundles C_1 and D unite as do D_1 and E. E_1 becomes a cotyledonary trace while cotyledonary bundle B divides and B_1 joins C. Bundle F becomes the cotyledonary partner of E_1. The bundles remaining after the cotyledonary traces depart are B_1C, C_1D, D_1E, F, G and H. These pass into the stem where bundles G and C_1D divide twice to form ten stem bundles.

The secondary tissues of the root and stem are formed from the cambium which developed from procambial cells established early in the stelar development.

The immature leaf is composed of three regions, the epidermis, the mesophyll and the provascular tissue. The epidermal tissue forms the upper and lower epidermis and the two types of trichomes which have their origin in the epidermis. The mesophyll develops into palisade and spongy parenchyma tissue, and the provascular regions form the conducting cells of the leaf.

LEGEND

- PROTOXYLEM
- METAXYLEM
- PHLOEM
- PHLOEM FIBERS

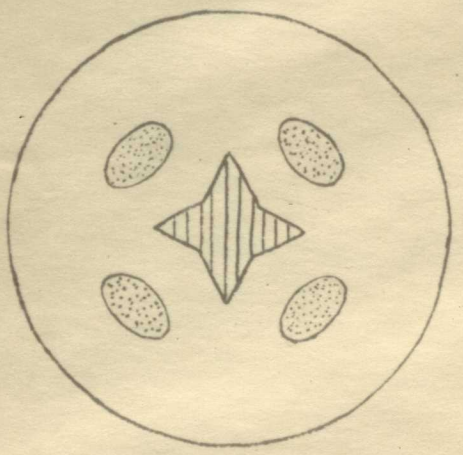
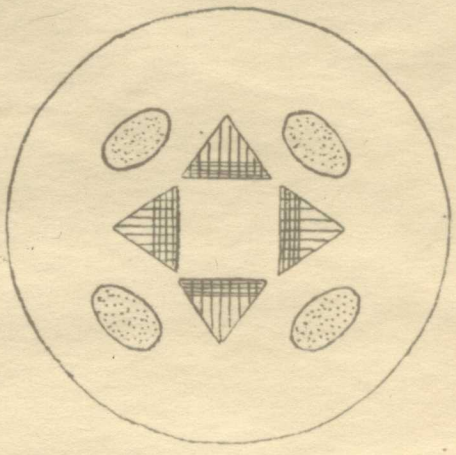


FIGURE 1



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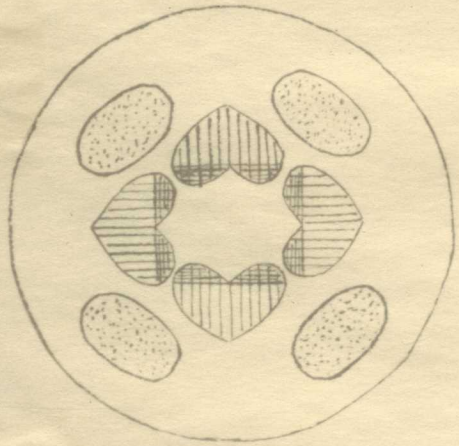

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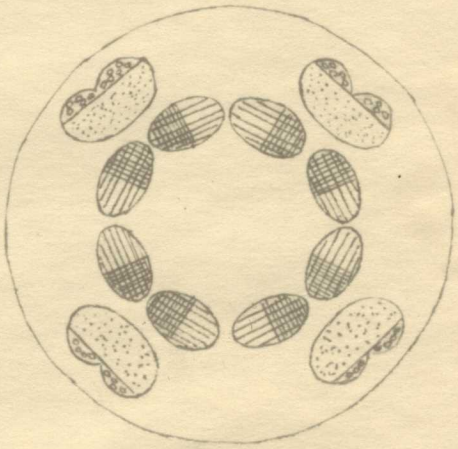

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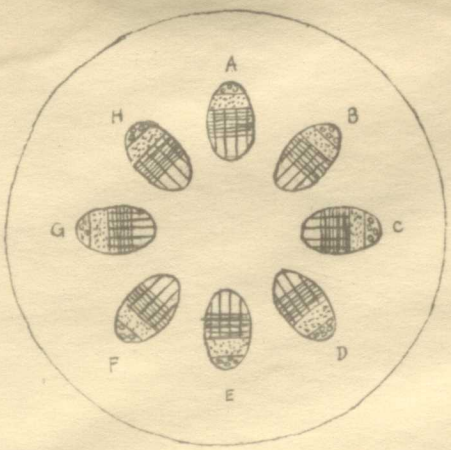

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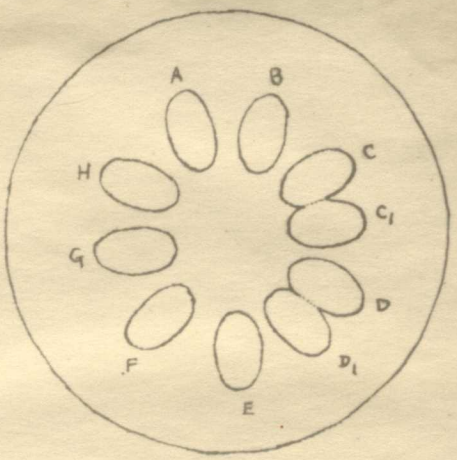

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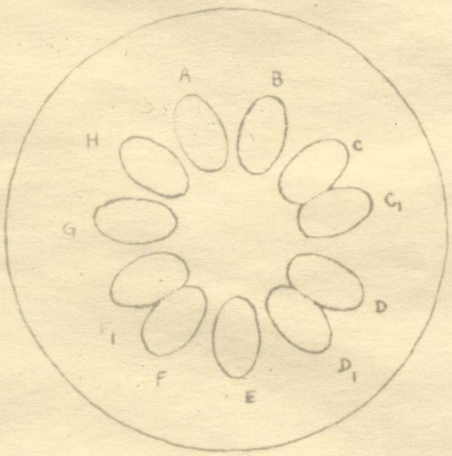

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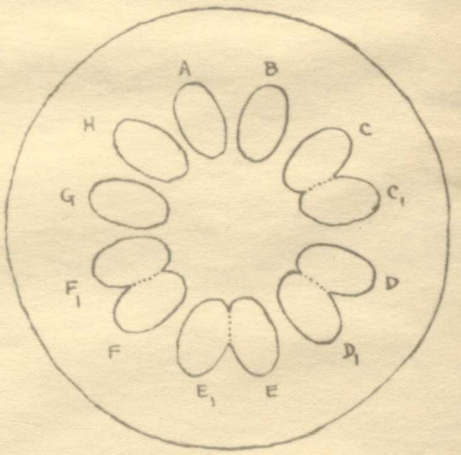

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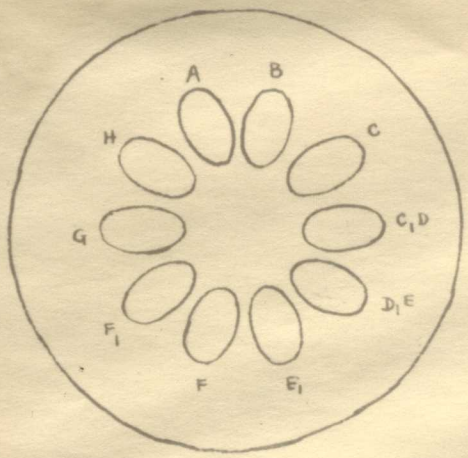

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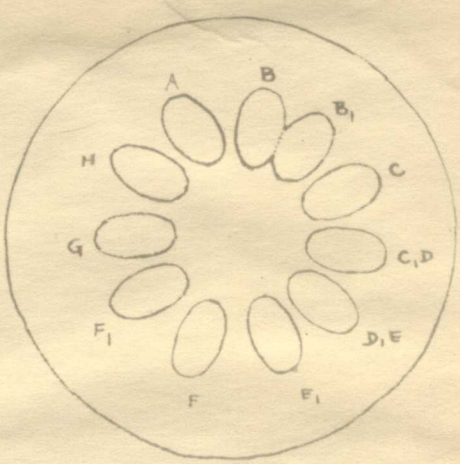

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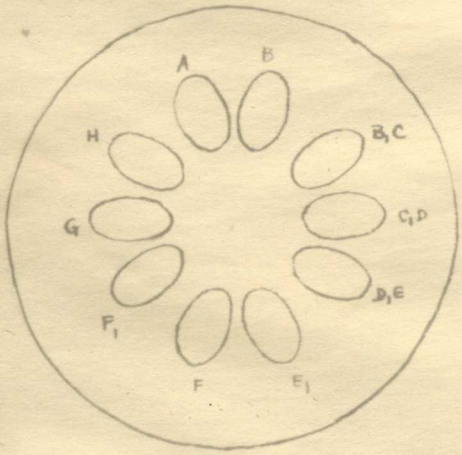

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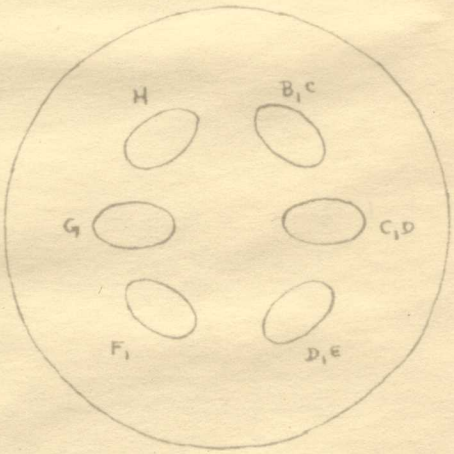

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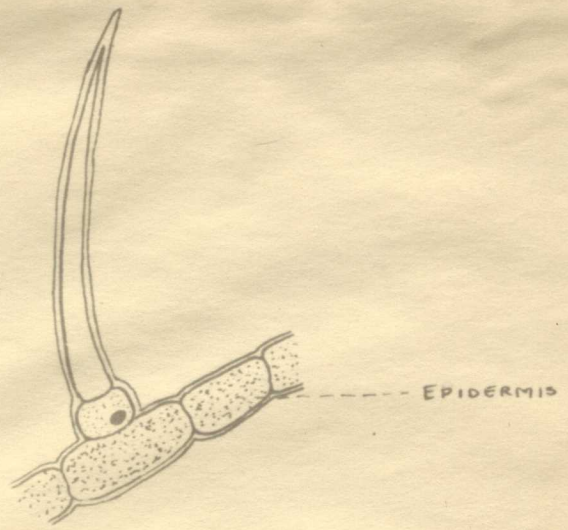
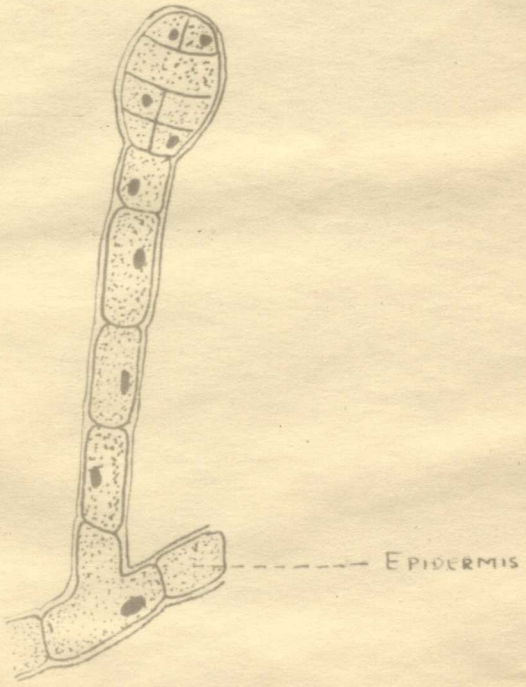


FIGURE 13



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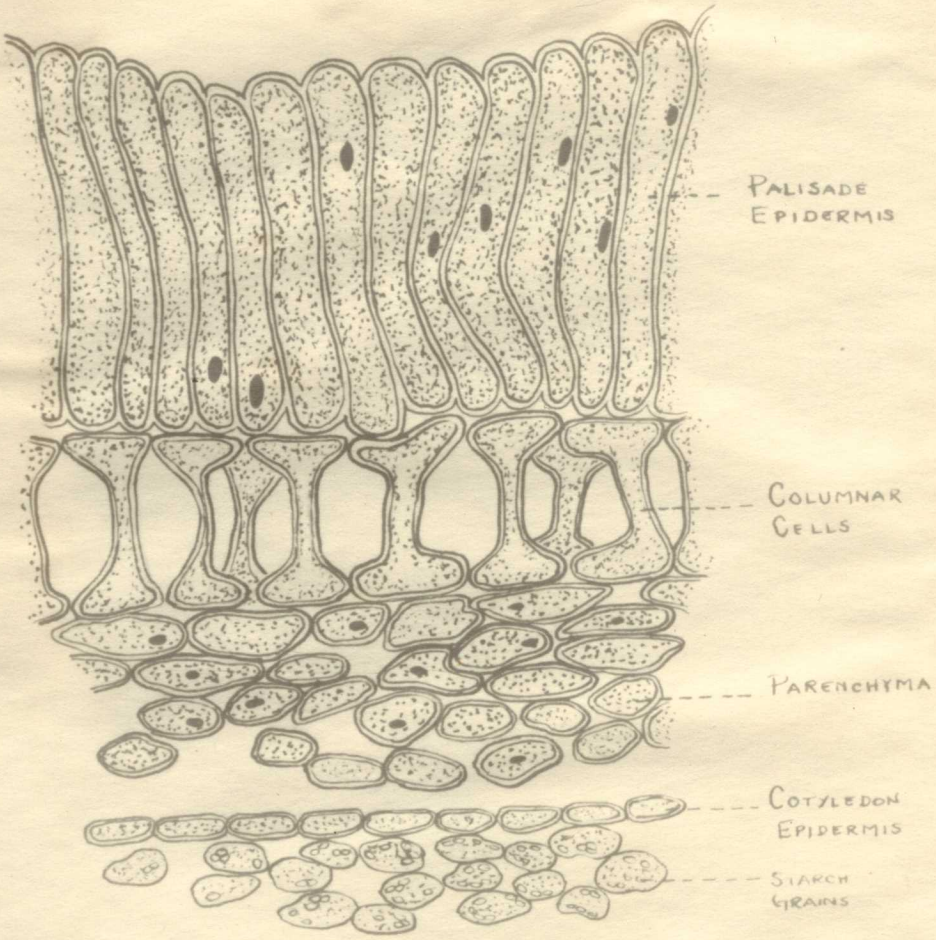
PALISADE
EPIDERMIS
COLUMNAR
CELLS
PARENCHYMA
COTYLEDON
EPIDERMIS
STARCH
GRAINS

FIGURE 15

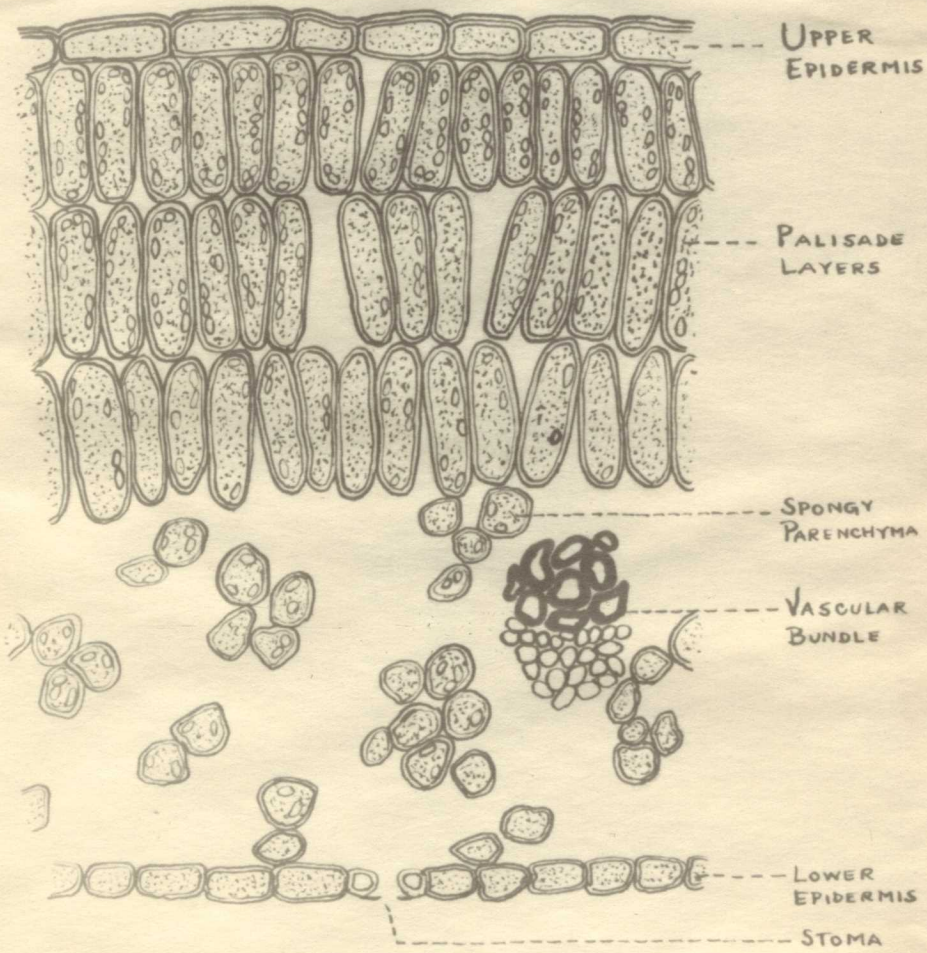
UPPER EPIDERMIS
PALISADE LAYERS
SPONGY PARENCHYMA
VASCULAR BUNDLE
LOWER EPIDERMIS
STOMA

FIGURE 16

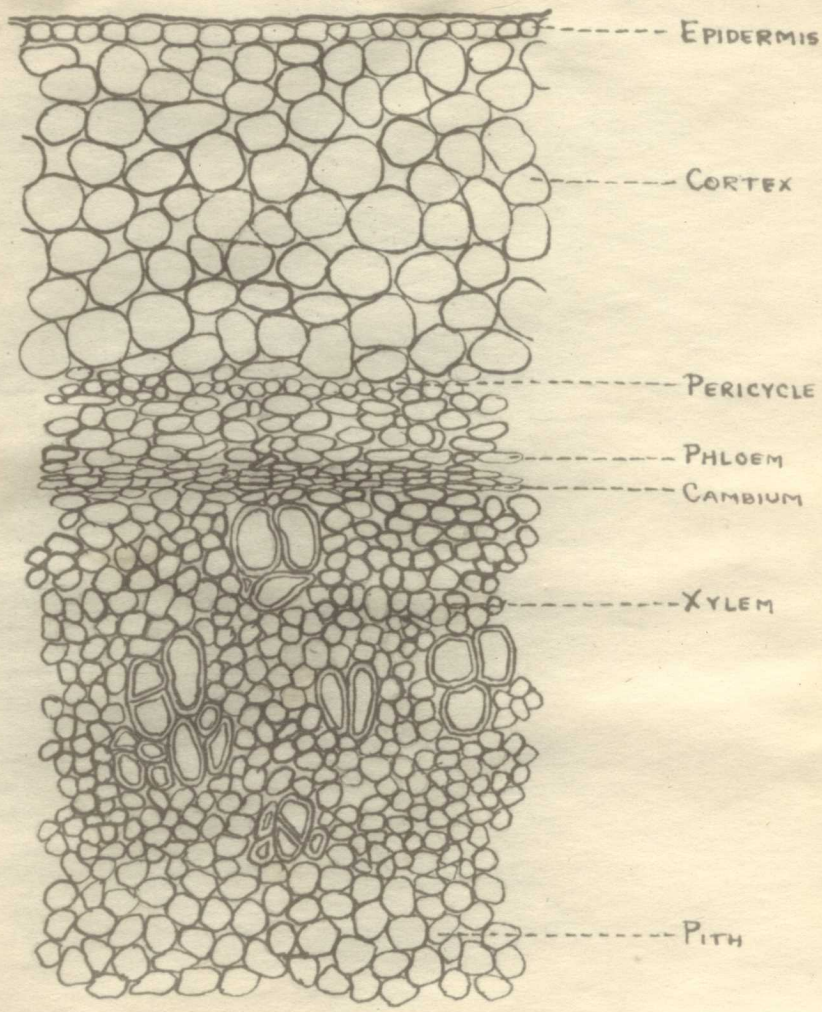

FIGURE 17

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