BRAZILIAN JALAP

PART I. HISTOLOGY

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INTRODUCTION

BRAZILIAN JALAP has been offered for sale in England quite frequently during recent years. It is stated to contain up to 20 per cent. of resin which, according to W. L. Scoville[1], bears some resemblance to the resin of Vera Cruz jalap.

E. M. Holmes[2] first described the morphological characters of Brazilian jalap in 1915 and gave the botanical source as *Piptostegia pisonis*, Mart. It is referred to by this name in various text books of pharmacognosy but a review of the older literature reveals some confusion regarding the botanical source of the drug. O. A. Farwell[3] concluded that the correct name of the plant was *Operculina macrocarpa* Linn Urban, but in 1947 Dr. J. Hutchinson, Royal Botanical Gardens, Kew, re-examined the nomenclature of the drug and stated that the botanical source of this material was *Ipomœa tuberosa* Linn[4]. It is referred to by this name in the British Pharmaceutical Codex, 1949.

The histology of the root has been described by Farwell[3] but this description is very inadequate and, if he was describing the same drug as that referred to in this paper, sometimes inaccurate. Moreover, there are no drawings. It seemed desirable, therefore, that a further examination of Brazilian jalap should be carried out with the object of determining the characters which would enable it to be identified in the powdered condition and to be recognised in admixture with other Convolvulaceous roots, particularly Vera Cruz jalap and Orizaba jalap.

The microscopical examination of several commercial samples revealed that although they were all based on the same anatomical structure there were some differences in detail between the drugs. It was obviously essential that authentic tubercles from the plant which yields the drug should be obtained and examined, and during the past four years many attempts have been made to obtain specimens of whole tubercles of *Ipomœa tuberosa* Linn, all without success.

It is thought desirable, however, to publish an account of the work that has been carried out on the samples of commercial Brazilian jalap.

MATERIAL

Sample A. A commercial sample obtained from a wholesale house in February, 1947.

Sample B. A commercial sample presented to the School of Pharmacy, College of Technology, Bristol, by Dr. T. E. Wallis in 1940.

Sample C. A commercial sample obtained from a wholesale house in August, 1947.

Sample D. A commercial sample obtained from a wholesale house in April, 1948.

MACROSCOPICAL CHARACTERS

The general description of the samples examined agrees with those previously given by Farwell[3] and Holmes[2] and with that given in the British Pharmaceutical Codex 1949.

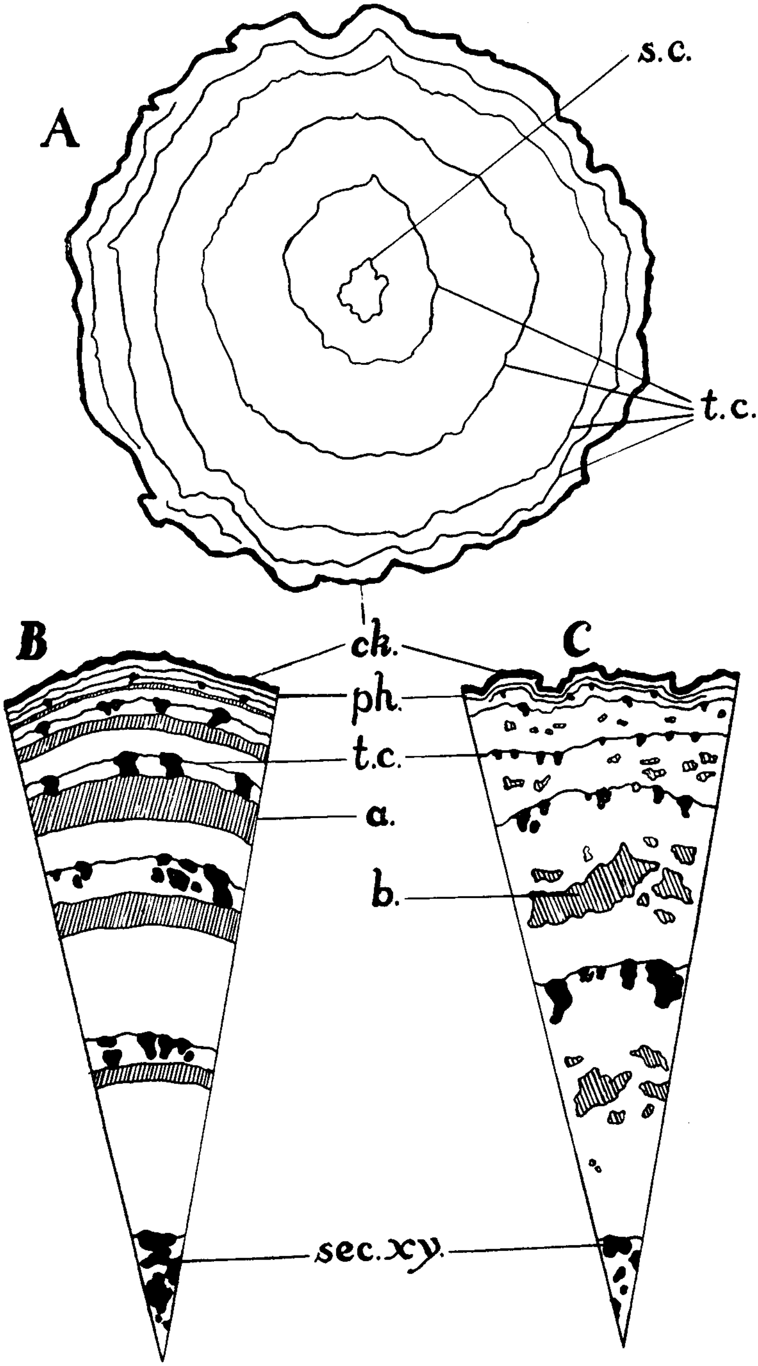
They consist of transverse or oblique slices of root from 2·5 to 7·5 cm. in diameter and from 0·3 to 1·5 cm. thick, and some wedge-shaped pieces up to 3·0 cm. thick. The colour in bulk is greyish-brown. On the outside of the root is a thin brown wrinkled cork. The transverse surface varies in colour from pale buff to yellowish-grey and exhibits from three to eight irregular concentric dark narrow bands alternating with wider and paler bands. The dark bands are often sunk below the level of the neighbouring tissue. The xylem elements which occur in the paler bands are small and widely separated and they often protrude above the surface of the drug. Masses of yellowish brown resin are frequently visible on the surface. The fracture of the drug is short. There is practically no odour and the taste is slightly acrid.

Smoothed transverse surfaces of the roots, when examined with a hand lens, show abnormal secondary growth which is characterised by the formation of concentric rings of tertiary cambiums (Fig. 1 A). These cambiums, along with the innermost ring which is a secondary cambium, give rise to a considerable amount of tissue on the outside but to much less tissue on the inside, the extent of tissue development decreasing with each successive tertiary zone, so that towards the outside of the root the tertiary cambiums are very close together. On the outside of the root there is a well developed cork and a narrow phelloderm.

Although it has not been possible to study the formation of the tertiary cambiums by examination of the authentic root in various stages of growth, it is probable that after the first cambium forms a ring of bundles close to the primary xylem, a tertiary cambium arises in the pericycle and this is followed by others in close succession and having similar origin. All the layers of tissue continue to function so that while the cambiums give rise to vascular tissue the pericycle cells built up a vast parenchyma. Thus alternate bands of widely separated vascular bundles and proliferated pericycle are formed. However, the appearance of the phloem and of the proliferated pericycle is not identical with that in

FIG. 1. *Brazilian jalap.*

A. Diagram of the transverse surface showing the arrangement of the tertiary cambiums. × 1½.
B. Diagram of a segment of the transverse surface of sample B. × 3.
C. Diagram of a segment of the transverse surface of sample C. × 3.
s.c., secondary cambium; t.c., tertiary cambium; ck., cork; ph., phelloderm; sec.xy., secondary xylem; a., band of elongated stone cells; b., islands of lignified parenchyma.


A
s.c.
t.c.
B
C
ck.
ph.
t.c.
a.
b.
sec.xy.

Orizaba jalap and it may be that the actual development of the cambiums is not exactly as outlined above.

When the smoothed transverse surface of the root is stained with phloroglucinol and hydrochloric acid and examined with a hand lens, the differences between the four samples become apparent.

Sample A shows lignified cells scattered throughout the phelloderm and bands of lignified cells in the outer region of the proliferated pericyclic parenchyma. The extent of the band of lignified cells decreases with each successive zone of tertiary growth (Fig. I B). Further examination shows that the cells are lignified stone cells.

Sample B shows no modification from the structure already described, no lignified tissue other than the vascular bundles being present.

Sample C shows irregularly shaped islands of lignified cells situated between successive zones of tertiary growth (Fig. I C). Further examination shows that these cells are lignified parenchyma.

Sample D shows similar islands of lignified parenchyma but they are very much smaller and far less numerous.

There is a complete absence of lignified stone cells from Samples B, C and D.

Microscopical Appearance

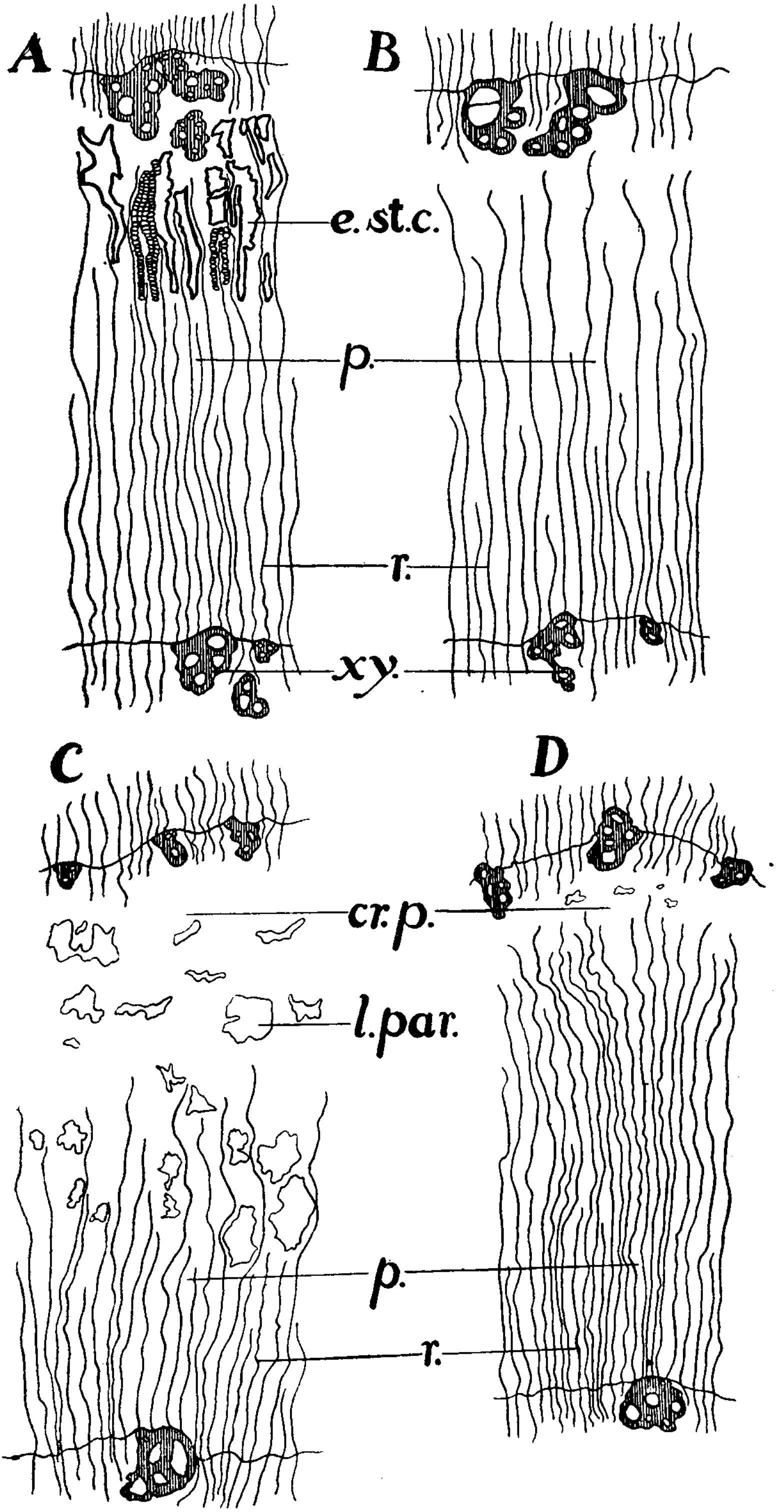
The general anatomical structure of the root can be further elucidated by examination of transverse, radial longitudinal and tangential longitudinal sections with the low power objective. It is then seen that the proliferated pericyclic tissue contains two distinct types of cell. Transverse sections, after clarifying by warming with chloral hydrate solution, show dark linear rows of cells, radially arranged, slightly wavy and extending throughout the entire tissue. They are continuous with similar cells in the parenchymatous tissue on the inside of the tertiary cambium. Longitudinal sections show these rows of cells to have all the features of secondary vascular rays. They are strap-like bands of tissue consisting of rounded cells, usually 1 or 2 cells wide, and from 4 to 10 cells deep. In order to distinguish this tissue from the remainder of the proliferated parenchymatous pericycle, they are referred to as " tertiary rays." The number of tertiary rays per mm. in all four samples of Brazilian jalap varies from 13·3 to 18·7 with average numbers for the actual samples as follows :—A, 15·1; B, 15·5; C, 15·3; D, 15·9.

In between the tertiary rays are radially arranged rows of larger rectangular parenchymatous cells, and scattered throughout this tissue are small groups of sieve tissue, often crushed. The rectangular parenchymatous cells are frequently modified into latex cells and in the outermost parts of the tissue in Sample A they are also modified into stone cells.

FIG. 2.—*Brazilian jalap.*

A, B, C and D. General arrangement of the tissues between the second and third tertiary cambiums of samples A, B, C and D respectively. × 20.

e.st.c., groups of elongated stone cells; p., parenchyma; r., " tertiary ray"; cr.p., crushed parenchyma; xy., xylem; l.par., lignified parenchyma.


A
B
e. st.c.
p.
r.
xy.
C
D
cr. p.
l. par.
p.
r.

Up to and including the third tertiary tissue the modification is very pronounced, the modified cells being axially elongated stone cells, but the extent of modification in later tissues decreases with each successive tertiary growth so that in the outermost regions there are often no stone cells. Furthermore, the extent of elongation of the cells themselves also decreases so that the stone cells in the outer regions are often isodiametric.

The xylem bundles, which contain up to 24 vessels, occur widely separated in the narrow band of parenchyma on the inside of the tertiary cambium. Where the inner layer of parenchyma of any one tertiary cambium impinges upon the outer layer of parenchyma from the previous cambium, there are tangential bands of crushed tissue, which vary in width in the different samples. In Sample D the tangential band of crushed tissue is quite narrow and some of the cells have slightly lignified walls. In Sample C, this zone is much wider and contains large, irregular shaped islands of lignified parenchyma. Fig. 2 (A, B, C and D) shows the general arrangement of the tissues seen in transverse section, between the second and third tertiary cambiums of samples A, B, C and D respectively.

Histology

The cork consists of two kinds of cells arranged in layers. Arising from the phellogen are 3 to 12 rows of thin-walled polygonal tabular cells, the walls of which are suberised and slightly lignified. Further layers of similar cork cells alternate with layers consisting of three or four rows of polygonal tabular cells having thickened tangential walls which are strongly suberised but not lignified. These cells often contain dark brown contents, though some are so compressed that in many cases the lumen is entirely obliterated. Measurement of the cork cells in surface view gave dimensions 40 to 120 μ long by 20 to 90 μ wide. The cork arises from a layer of thin walled phellogen which produces on its inside a fairly extensive phelloderm. The outer layers of the phelloderm consist of regularly arranged rectangular tabular cells while the inner layers consist of irregularly shaped cells with few intercellular spaces. The cell walls are thin and composed of cellulose. The cells contain abundant starch grains and numerous cluster crystals of calcium oxalate. The starch grains vary in size from 5 to 30 μ wide. The smaller grains may be in aggregations containing up to 12 grains, but these are not numerous. The medium-sized grains occur in groups of 2 to 6 components while the larger grains are almost always single or in pairs. The smaller grains are oval or subspherical in shape but the majority of the larger grains are semifaceted or muller shaped, and typical of the family Convolvulaceae. The hilum and striations are not visible but the grains exhibit a well-defined maltese cross when examined under polarised light. (Fig. 6, B.) The cluster crystals of calcium oxalate found in the phelloderm are from 8 to 25 μ in diameter with some occasional crystals measuring up to 40 μ.

The stone cells which occur singly or in groups containing 2 to 5 cells in Sample A, measure from 40 to 160 μ long and 30 to 60 μ wide in transverse section. Similar dimensions are obtained when the measure-

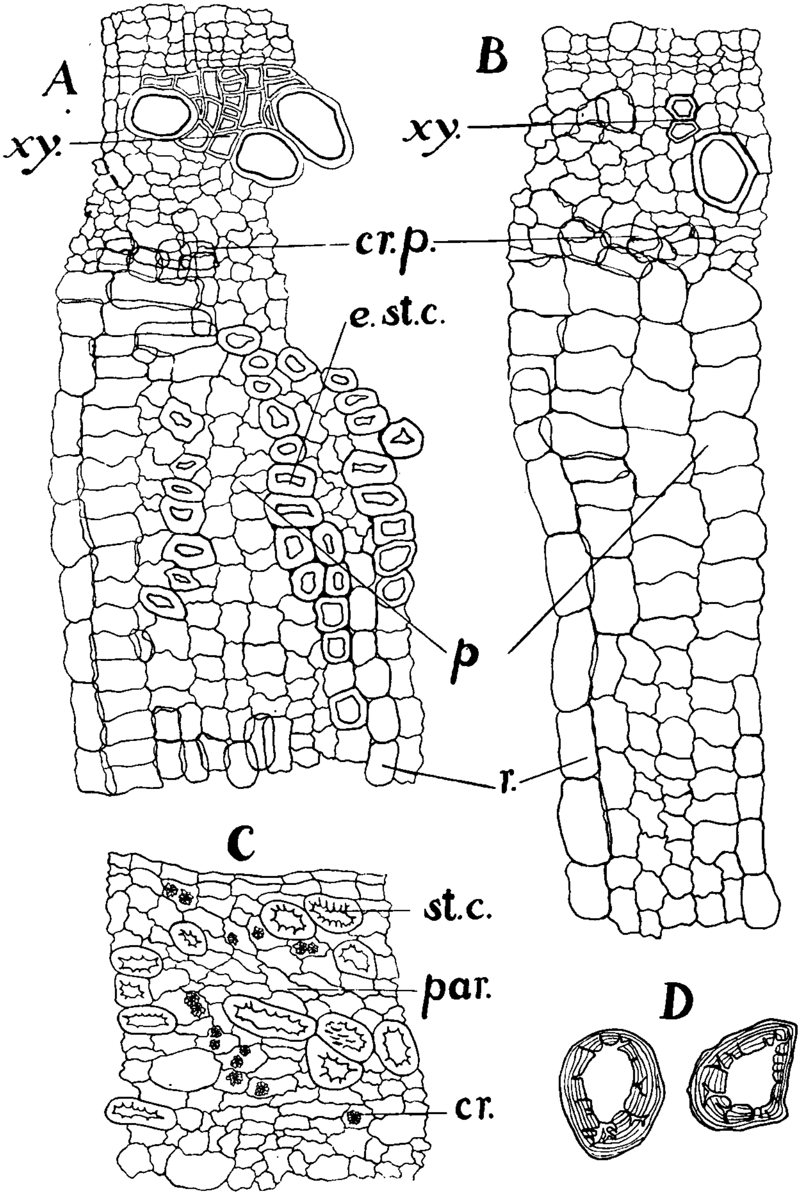


FIG. 3.—*Brazilian jalap.*

A. Transverse section sample A showing xylem and proliferated pericyclic tissue.
B. Transverse section sample B showing xylem and proliferated pericyclic tissue.
Both × 80.
C. Transverse section phelloderm sample A. × 80.
D. Stone cells from phelloderm sample A. × 180.
xy., xylem; cr.p., crushed parenchyma; e.st.c., elongated stone cells; par., parenchyma; r., "tertiary ray"; st.c., stone cell; p., proliferated pericyclic parenchyma; cr., crystal of calcium oxalate.

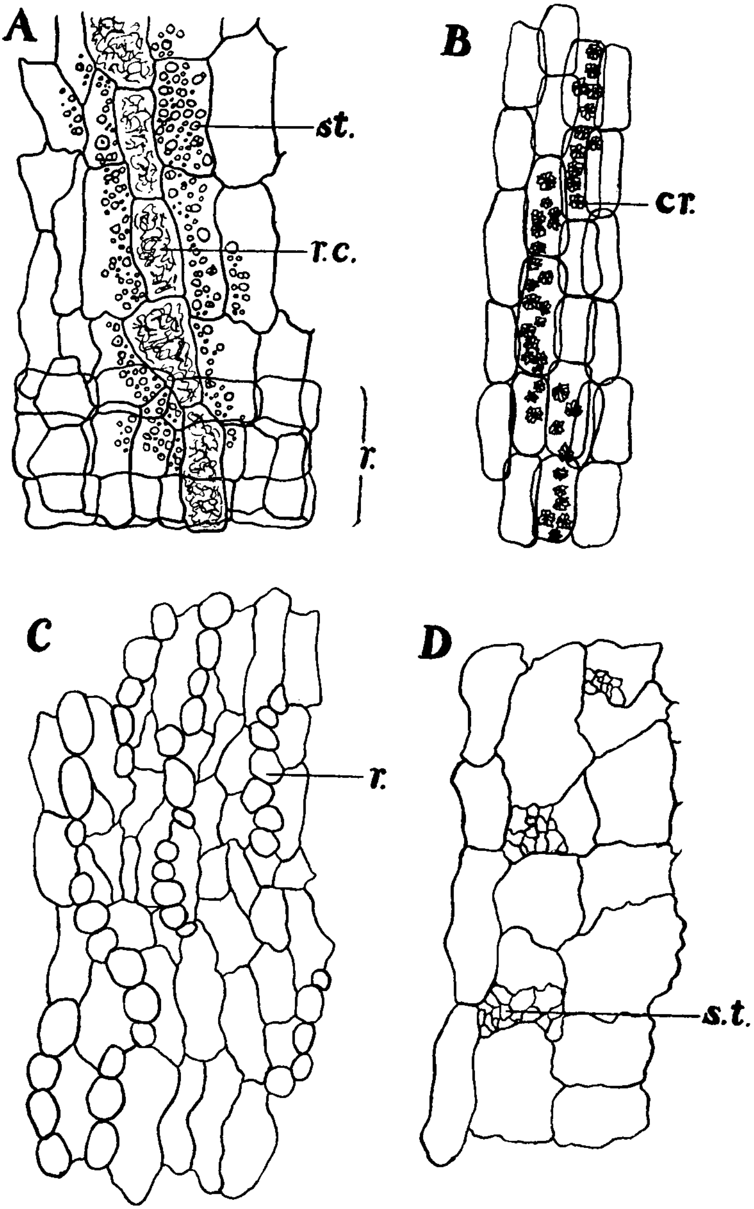


FIG. 4.—*Brazilian jalap.*

A. Longitudinal section showing latex cells.
B. Longitudinal section showing files of calcium oxalate crystals.
C. Tangential section showing " tertiary rays." All × 120.
D. Transverse section showing crushed sieve tissue. × 270.
st., starch; r.c., latex cell; r., " tertiary ray "; s.t., sieve tissue; cr., cluster crystals of calcium oxalate.

ments are made in longitudinal sections. The walls of the cells are from 12 to 15 μ thick; they are strongly lignified and show well-marked strati-

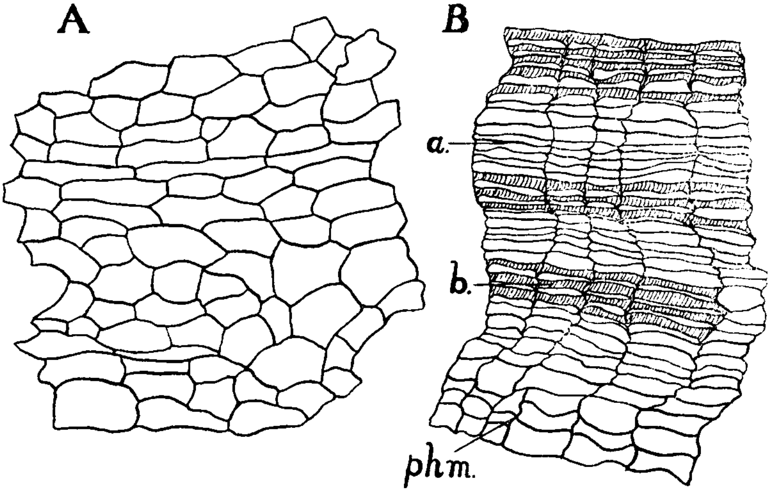


FIG. 5.—*Brazilian jalap.*

A. Surface section of cork sample D.
B. Transverse section cork sample B. Both × 120.
a., cork with thin, slightly lignified walls; b., cork with suberised tangential walls.

fications and simple branched pits. The cells of the " tertiary rays " have thin cellulosic walls, though a few are slightly suberised. They are rounded or oval in shape and may be seen particularly well in tangential longitudinal section (Fig. 4, C). The cells are filled with starch grains. The parenchyma of the proliferated pericycle consists of thin walled rectangular cells which tend to increase in size the further they are from the cambium. This is particularly true of Sample B. Almost every cell can be traced back directly to the cambium as they occur in regular radial rows. The cells furthest from the cambium often have larger tangential than radial measurements, while nearer to the cambium the reverse is usually the case. Quite frequently the cells have slightly suberised walls and contain brownish granular contents which are soluble in solution of sodium hydroxide and chloral hydrate solution and which stain bright lemon yellow with dilute solution of iodine, red with tincture of alkanet, and pink with solution of corallin soda. Longitudinal sections show that these latex cells occur in files of 4 to 6 cells (Fig. 4, A). The parenchymatous cells contain starch grains and numerous cluster crystals of calcium oxalate which occur in longitudinal files containing up to 30 crystals (Fig. 4, B). The crystals are very regular in size, being chiefly from 15 to 20 μ in diameter.

The elongated stone cells occurring in sample A are up to 600 μ long and 60 μ wide. They decrease in size with each successive tissue, and in the outer tissues may be similar in size to the stone cells in the phelloderm. The stone cells have lignified walls from 8 to 12 μ

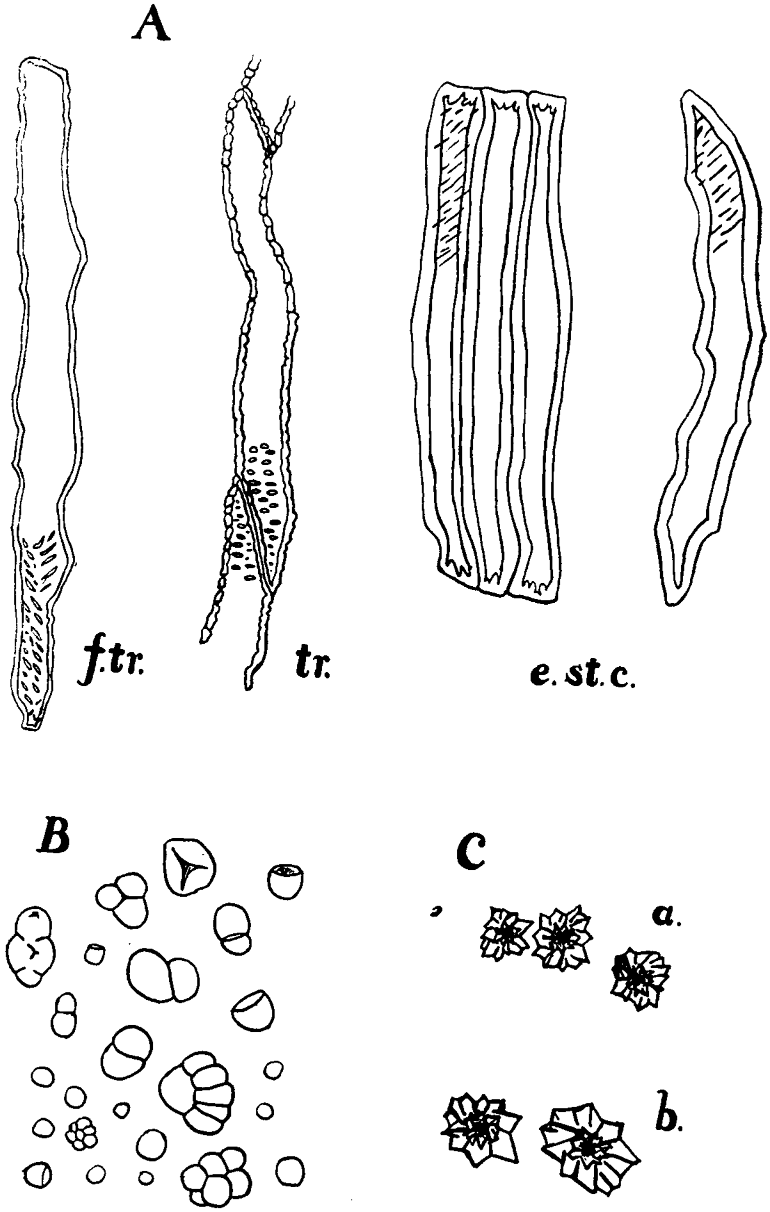


FIG. 6.—*Brazilian jalap.*

A. Elements from macerated tissue sample A × 150.
B. Starch × 270.
C. Cluster crystals of calcium oxalate × 360.
f.tr., fibre tracheid; tr., tracheid; e.st.c., elongated stone cell; a., calcium oxalate from pericyclic parenchyma; b., calcium oxalate from phelloderm.

thick with oblique elongated simple pits. Most of the cells have square ends, but occasionally there are cells with bluntly pointed end (Fig. 6A). The sieve tissue, consisting, of sieve tubes and companion cells, occur in isolated groups, often crushed, between the tertiary rays. The xylem elements consist of lignified vessels, tracheids and fibre tracheids. No true fibres were observed in any of the samples. The vessels are scalariform-reticulately thickened and are from 30 to 120 μ in diameter. The

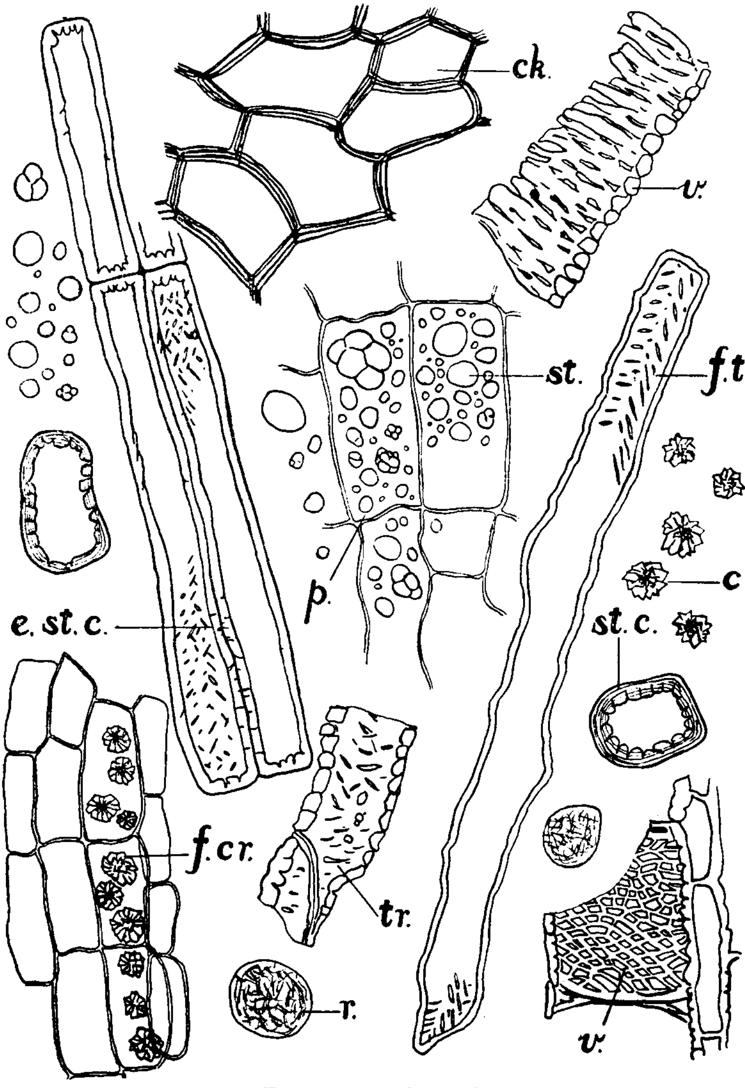


FIG. 7.—*Brazilian jalap.*
Powdered drug sample A × 200.
ck., cork; v., vessel; e.st.c., elongated stone cells; st.c., stone cell; p., parenchyma packed with starch; st., starch; tr., tracheid; f.tr., fibre-tracheid; r., mass of resin; f.cr., files of calcium oxalate crystals in parenchymatous tissue.

tracheids measure from 250 to 500 μ long and 30 to 60 μ in diameter, and have elongated bordered pits. The fibre-tracheids measure from 480 to 750 μ long and 25 to 50 μ in diameter. They have oblique slitlike pits.

Examination of the Powdered Drug

The following features are characteristics of all four samples:—

(1) Abundant starch, 3 to **12** to **18** to 30 μ in diameter. The smaller grains are rounded and often in aggregations; the larger grains are muller shaped and usually single or in groups of two or three.

(2) Portions of parenchymatous tissue packed with starch.

(3) Numerous cluster crystals of calcium oxalate 8 to **15** to **25** to 40 μ wide.

(4) Portions of parenchymatous tissue, containing files of cluster crystals of calcium oxalate, 15 to 20 μ wide.

(5) Masses of brownish granular latex which stain bright lemon yellow with dilute solution of iodine, red with tincture of alkanet and pink with solution of corallin soda.

(6) Brown cork in surface view, some cells of which are slightly lignified and measuring 40 to 120 μ long by 20 to 90 μ wide.

(7) Fragments of reticulate-scalariform vessels. Absence of vessels with round or oval bordered pits.

(8) Tracheids, often entire, measuring 250 to 500 μ long by 30 to 60 μ wide, with elongated bordered pits.

(9) Fibre tracheids, often entire, measuring 480 to 750 μ long by 25 to 50 μ wide, with oblique slit-like pits.

(10) Absence of true fibres.

Sample A also contains:—

(1) Stone cells 40 to 160 μ long by 30 to 60 μ wide with lignified walls 12 to 25 μ thick.

(2) Elongated stone cells up to 600 μ long and 60 μ wide, with lignified walls 8 to 12 μ thick.

Sample C and, to a slight extent *Sample D*, also contain:—

(1) Portions of thin walled irregularly sharped lignified parenchymatous tissue.

In the absence of authentic specimens of *Ipomœa tuberosa*, Linn, it is impossible to define the features by which the tubercles of this plant may be identified. The 4 samples of commercial Brazilian jalap examined, while based upon the same anatomical structures and having many features in common, show certain well-defined differences. Further investigation will therefore be necessary in order to define exactly the histological features of Brazilian jalap.

I would like to express my sincere thanks to Dr. T. E. Wallis, who has shown keen interest in this work and given me much valuable advice.

References

1. Scoville *J. Amer. pharm. Ass.*, 1918, **7**, 785.
2. Holmes, *Pharm. J.*, 1915, 671.
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