

# THE PHARMACOGNOSY OF THE ASPIDOSPERMA BARKS OF BRITISH GUIANA\*

## PART V. THE MICROSCOPY OF THE BARKS OF *Aspidosperma megalocarpon* MUELL. ARG. AND *Aspidosperma quebracho-blanco* SCHLECHT.

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Three samples of barks of each of the two species *Aspidosperma megalocarpon* Muell. Arg. and *Aspidosperma quebracho-blanco* Schlecht. have been examined histologically. The arrangement of tissues, cell structure and cell contents in each bark are accurately described and illustrated. The diagnostic characters by which the two barks may be identified and distinguished from each other are tabulated. Those characters by which the bark of *A. megalocarpon* differs from those of *A. album*, *A. excelsum* and *A. ulei* are also discussed.

THE macroscopical characters of the bark of *Aspidosperma megalocarpon* Muell. Arg. have already been described, illustrated and compared with those of the bark of *Aspidosperma quebracho-blanco* Schlecht<sup>1</sup>. The structure of the latter bark has been incompletely described by several authors<sup>2-8</sup>, it was official in the B.P.C. 1934<sup>9</sup> and the Dispensatory of the U.S.A. 1943<sup>10</sup>. The detailed histology of the bark of *A. megalocarpon* is described in this paper, together with a more complete series of drawings than has been presented by earlier workers, to illustrate the structural characters of the bark of *A. quebracho-blanco*.

### MATERIALS

Our samples of the bark of *A. megalocarpon* were those previously designated 3A, 3B and 3C collected in British Guiana in 1949, 1950 and 1954 respectively<sup>1</sup>. Our authenticated samples of the bark of *A. quebracho-blanco* were: 6A, collected by Dr. Martin Cardenas in 1953 at Cochabamba in Bolivia; 6B, obtained from the Museum of the Pharmaceutical Society (used by G. R. A. Short for his investigations in 1926<sup>11</sup>); and 6C, collected by the Conservator of Forests, British Guiana in British Guiana in 1954.

### EXPERIMENTAL

Line drawings to illustrate the diagnostic characters of outer and inner surfaces and of tissue distribution as seen in smoothed transverse sections of the barks of these two species are given in Figures 1 and 5, A, B and C.

#### *Histology of the Bark of A. megalocarpon* (Figs. 1, 2, 3 and 4)

The histological characters of this bark were in general agreement with those of the three other *Aspidosperma* species previously described<sup>1,12,13</sup>

\* The subject matter of this communication forms part of a thesis by one of us (J.D.K.) accepted by the University of Nottingham for the degree of Doctor of Philosophy in Pharmacy.

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but specific differences were found. The thick cork comprised some fifteen to fifty layers of tangentially-elongated cells, with inner and outer tangential walls thickened and lignified (Fig. 1, D, *ck* and Fig. 3, A, *ck*). A few groups of sclereids, similar to those present in the phelloderm (Fig. 1, C, *g.sc*), were also enclosed in the cork. One to three layers of thin-walled and tangentially-elongated cells formed the phellogen. A well-marked phelloderm contained many sclerenchymatous cells arranged in small groups (Fig. 1, D, *phe* and Fig. 3, A, *phe*), together with thin-walled slightly tangentially-elongated parenchymatous cells. Individual sclereids were isodiametric to tangentially-elongated with thick, lignified walls, traversed by simple or branched pits and with small to somewhat large lumen. Cortex of bands of parenchyma alternating with four to six more or less continuous bands of sclereids (Fig. 1, C and E, *b.sc* and Fig. 3, B, *b.sc*); parenchymatous cells with large intercellular spaces; sclereids similar to those of the phelloderm, a few of which contain a granular material which stained yellow with iodine solution and pinkish-red with Millon's reagent. Towards the inner region of the cortex were discontinuous groups of sclereids and at times a few isolated thick-walled fibres; no defined endodermis or pericycle were found.

Up to about 50 per cent of the bark is of phloem consisting of sieve tissue, parenchyma, fibres, medullary rays and sclereids. In the outer half of the phloem are groups of sclereids, and the cells of the medullary rays are sclerotic when traversing these groups; the inner half of the phloem contains no sclereids. The sieve tubes have oblique, compound sieve plates on the end walls (Fig. 2, D, E and F, *s.p*, Fig. 3, E, *s.p* and Fig. 4, A, *s.p*) and are less readily distinguished in the outer part of the phloem. Phloem parenchyma consists of thin-walled cells, with a few intercellular spaces, some vertical walls reticulately thickened and with compound pits. Fibres are scattered throughout the phloem and a few are embedded in the sclereid groups of the pericyclic region. The greater number of fibres with narrow lumen are mostly isolated (Fig. 2, C, D, E and F, *p.f* and Fig. 3, C, D and E, *p.f*) or very rarely in groups of two to three fibres (Fig. 2, B, *g.p.f*); a smaller number with large lumen are always isolated (Fig. 2, E, *p.f.l* and Fig. 4, A, *p.f.l*). Both types of fibres, which measure R and T = 28 to 80 to 134  $\mu$  and H = 600 to 1140 to 1650  $\mu$  are spindle shaped, with bluntly pointed ends and thick, lignified, stratified walls traversed by a few simple pits along which splitting may have occurred. A crystal sheath surrounds the isolated phloem fibres and groups of fibres, except when these are embedded in groups of sclereids. The medullary rays are very wavy, are one to two cells in width but becoming up to three cells wide (Fig. 2, A, *e.m.r*) towards the periphery of the phloem; they are 25 to 45 cells in height; individual cells are straight to somewhat wavy in outline and contain starch granules. When these medullary rays pass through groups of sclereids, the cells become sclerotic (Fig. 2, A and B, *sc.m.r* and Fig. 3, C, *sc.m.r*) with thick pitted walls and narrow lumen.

In the cortical and phloem parenchyma are abundant simple or 2- to 4-compound starch granules; individual granules with eccentric hilum

*ASPIDOSPERMA MEGALOCARPON*

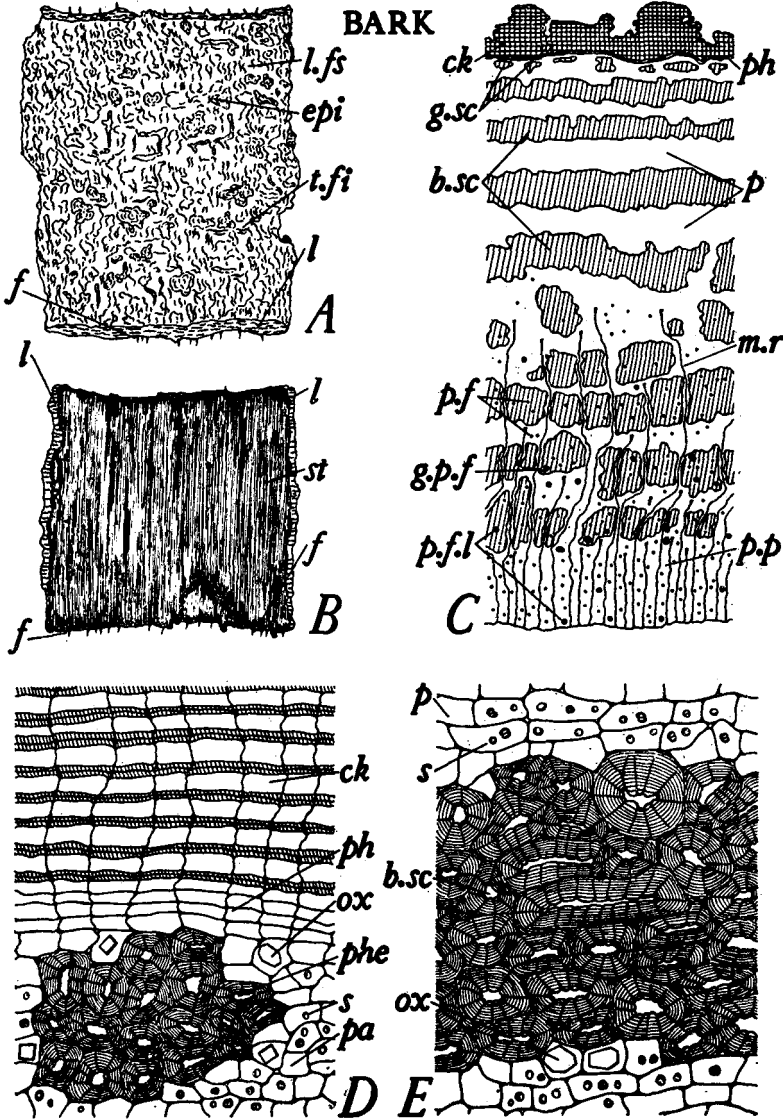


FIG. 1. *Aspidosperma megalocarpon* bark, macroscopical characters and T.S.:— A, outer surface  $\times \frac{1}{4}$ ; B, inner surface  $\times \frac{1}{4}$ ; C, smoothed T.S.  $\times 12$ ; D, cork, phellogen and phellogen; E, cortex; D and E,  $\times 200$ ; *b.sc*, band of sclereids; *ck*, cork; *epi*, epiphyte; *f*, fibre; *g.p.f*, group of phloem fibres with narrow lumen; *g.sc*, group of sclereids; *l*, laminations; *l.fs*, longitudinal furrow; *m.r*, medullary ray; *ox*, crystal of calcium oxalate; *p*, cortical parenchyma; *pa*, cortical parenchyma found associated with phellogen sclereids; *ph*, phellogen; *phe*, phellogen sclereids; *p.f*, isolated phloem fibre with narrow lumen; *p.f.l*, isolated phloem fibre with large lumen; *p.p*, phloem parenchyma; *s*, starch; *st*, longitudinal striation; *t.fi*, transverse fissure.

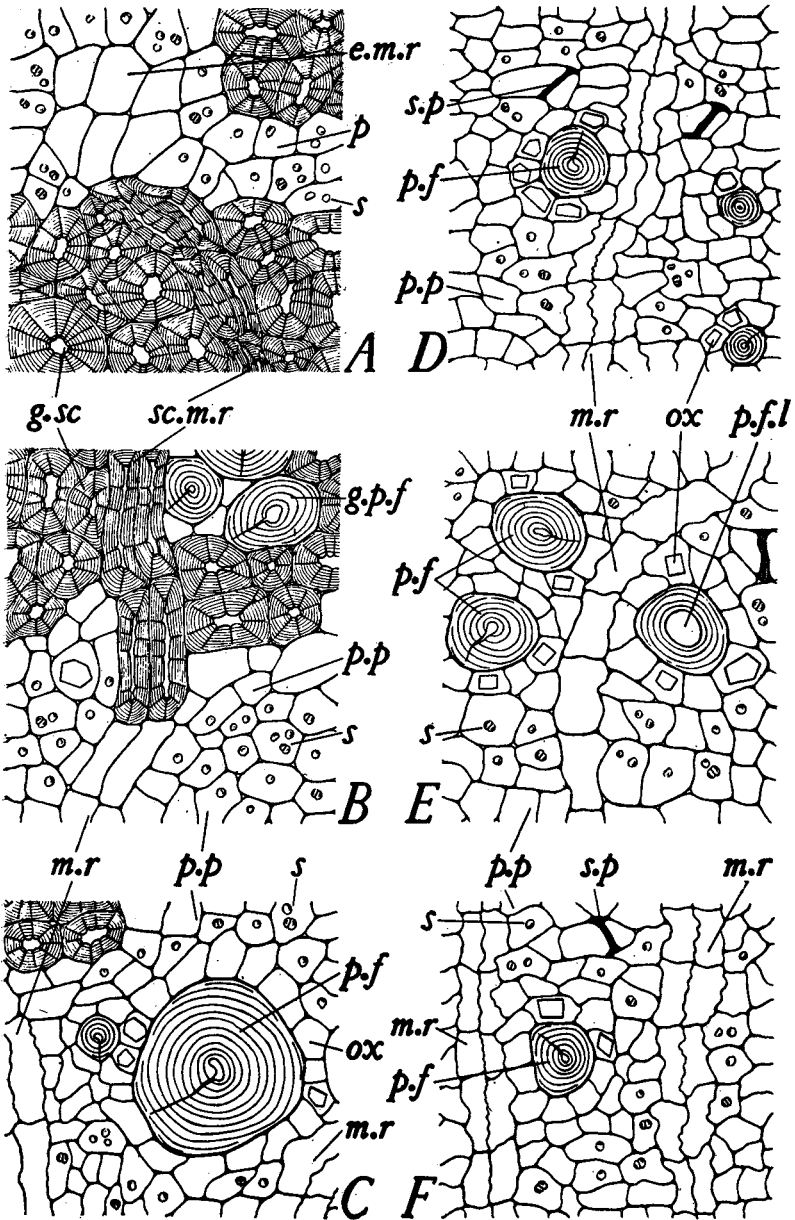


FIG. 2. *Aspidosperma megalocarpon* bark in T.S.:—A, innermost cortex and outermost phloem; B, outer phloem; C, D and E, phloem; F, innermost phloem; all  $\times 200$ ; e.m.r., end of medullary ray; g.p.f, group of phloem fibres with narrow lumen; g.sc, group of sclereids; m.r, medullary ray; ox, crystal of calcium oxalate; p, cortical parenchyma; p.f, isolated phloem fibre with narrow lumen; p.f.l, isolated phloem fibre with large lumen; p.p, phloem parenchyma; s, starch; sc.m.r, sclerotic medullary ray; s.p, sieve plate.

are spherical, ovoid or plano-convex and up to  $32\ \mu$  in diameter (Figs. 1, 2, 3 and 4, *s*). Calcium oxalate, in square, rectangular or obliquely rectangular prisms or small cubes up to  $55\ \mu$ , occurs as a crystal sheath around the fibres or groups of fibres (Figs. 1, 2, 3 and 4, *ox*) and, at times, in association with the sclereid groups.

*Powder.* The powder is very light brown in colour and exhibits characters typical of those structures described above. These include the cork cells which are polygonal in surface view and slightly reddish brown in colour, with thick, lignified walls. The sclereids occur in masses the individual cells being  $25$  to  $75$  to  $115\ \mu$  in length and  $15$  to  $45$  to  $60\ \mu$  in width; they are thick walled and lignified, the lumen is narrow but is somewhat larger in those sclerotic cells containing granular contents (Fig. 4, B, *sc.g*) which stain with iodine solution and with Millon's reagent. The phloem fibres are usually broken during the powdering of the bark; fragments with very narrow lumen, either isolated or in groups of two to three, preponderate; fewer fragments of isolated fibres with large lumen are present, which may be associated with sclereids. The calcium oxalate crystal sheath surrounding these fibre fragments or groups of fragments is readily detected (Fig. 4, B, *f.l.s*, *g.p.f* and *p.f*). Calcium oxalate crystals (Fig. 4, B, *ox*) are also present in association with a few of the groups of sclereids. Phloem parenchyma (Fig. 4, B, *p.p*) associated with cells of the medullary rays (Fig. 4, B, *m.r*) and cortical parenchyma (Fig. 4, B, *p*) of tangentially-elongated cells all contain starch granules.

*Detailed Histology of the Bark of A. quebracho-blanco* (Figs. 5, 6 and 7)

The general histological characters of this bark were in agreement with those exhibited by the three barks of the genus, described previously and already compared with the bark of *A. quebracho-blanco*<sup>1,12,13</sup>. The distribution of the tissues in *A. quebracho-blanco* differs from those described in this present paper for *A. megalocarpon* in a number of important respects. Thus, the cork is composed of about twenty to seventy-five layers of cells which are pronouncedly elongated (Fig. 5, C, *ck*, Fig. 6, A, *ck* and Fig. 7, A, *ck*). The phelloderm is well marked as a somewhat discontinuous band of sclereids about eight to twelve cells in radial breadth, together with a small amount of parenchyma. In the cortex the abundant sclereids are arranged in a number of masses (Fig. 5, C, *g.sc*, Fig. 6, B, *g.sc* and Fig. 7, B, *g.sc*). The phloem, comprising up to about 70 per cent of the thickness of the bark, may be divided into three very unequal zones. The greater part contains abundant sclereid groups, also fibres with narrow lumen which are mostly isolated or very rarely in groups of two fibres and at times embedded in the sclereid groups; medullary rays become sclerotic when passing through these sclereid groups. Towards the inner region of phloem, isolated fibres with narrow lumen are present but sclereid groups are absent. The innermost region contains isolated fibres with narrow lumen and a few isolated fibres with large lumen, sclereid groups are absent. The medullary rays (Fig. 6 and 7, *m.r*) are very wavy, due to displacement by groups of sclereids; they are one to three cells in width but becoming up to five cells wide (Fig. 6,

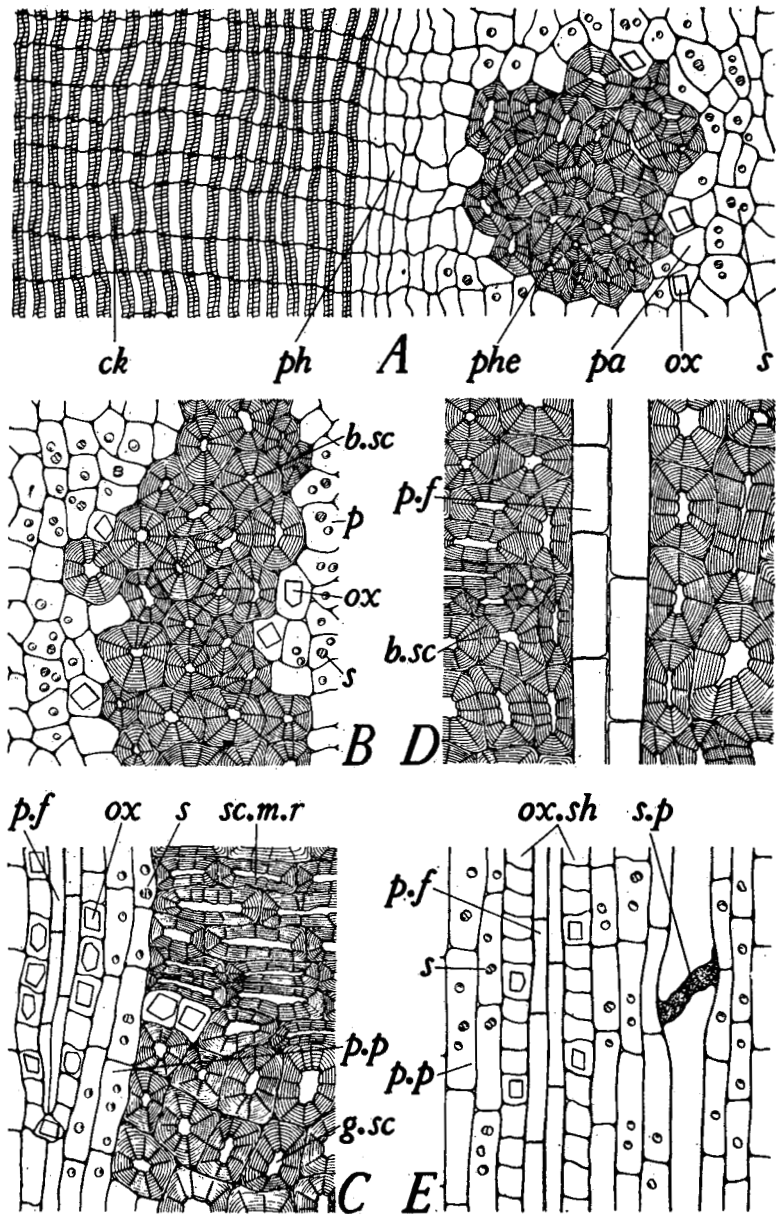


FIG. 3. *Aspidosperma megalocarpon* bark in L.S.:—A, cork phellogen and phello-derm; B, cortex, C, outer phloem; D and E, phloem; all  $\times 200$ ; *b.sc*, band of scleroids; *ck*, cork; *g.sc*, group of scleroids; *ox*, crystal of calcium oxalate; *ox.sh*, calcium oxalate crystal sheath; *p*, cortical parenchyma; *pa*, cortical parenchyma found associated with phellodermic scleroids; *ph*, phellogen; *phe*, phellodermic scleroids; *p.f*, isolated phloem fibre with narrow lumen; *p.p*, phloem parenchyma; *s*, starch; *sc.m.r.*, sclerotic medullary ray; *s.p*, sieve plate.

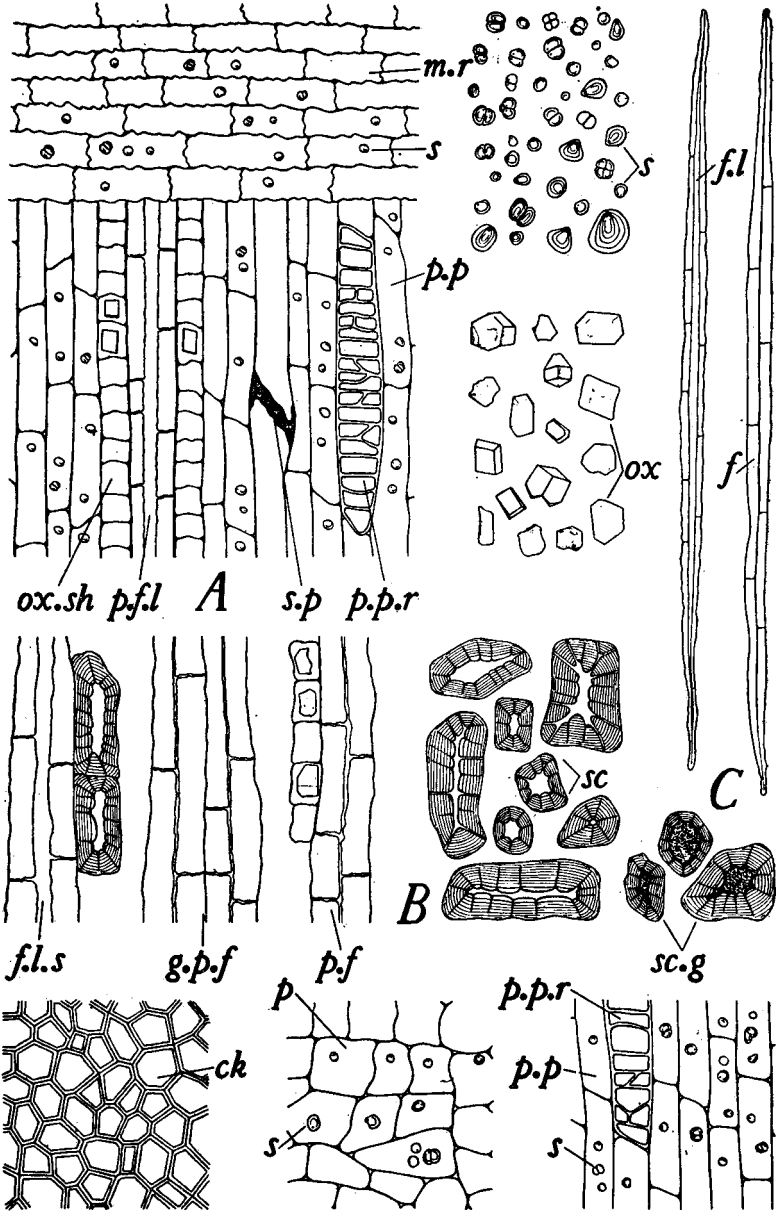


FIG. 4. *Aspidosperma megalocarpon* bark in L.S., powder and macerate:—A, innermost phloem  $\times 200$ ; B, various components as seen in powder  $\times 200$ ; C, fibres, isolated by maceration  $\times 50$ ; *ck*, cork; *f*, phloem fibre with narrow lumen; *f.l*, phloem fibre with large lumen; *f.l.s*, sclereids attached to large lumened fibre; *g.p.f*, group of phloem fibres with narrow lumen; *m.r*, medullary ray; *ox*, crystal of calcium oxalate; *ox.sh*, calcium oxalate crystal sheath; *p*, cortical parenchyma; *p.f*, isolated phloem fibre with narrow lumen; *p.f.l*, isolated phloem fibre with large lumen; *p.p*, phloem parenchyma; *p.p.r*, phloem parenchyma with vertical walls reticulately-thickened; *s*, starch; *sc*, sclereids; *sc.g*, sclereids with granular material; *s.p*, sieve plate.

C, *e.m.r*) towards the periphery of the phloem and are 16 to 25 cells in height.

The cells of the cork of *A. quebracho-blanco* differ from those of *A. megalocarpon* in being more pronouncedly tangentially-elongated and have walls which are thin and are either unligified or only slightly ligified. The other cell forms are similar in the two barks: these include the phellogen; the sclereids of phelloderm, cortex and phloem together with those sclereids containing granular contents which stain with iodine and Millon's reagent; the two types of phloem fibres with narrow or wide

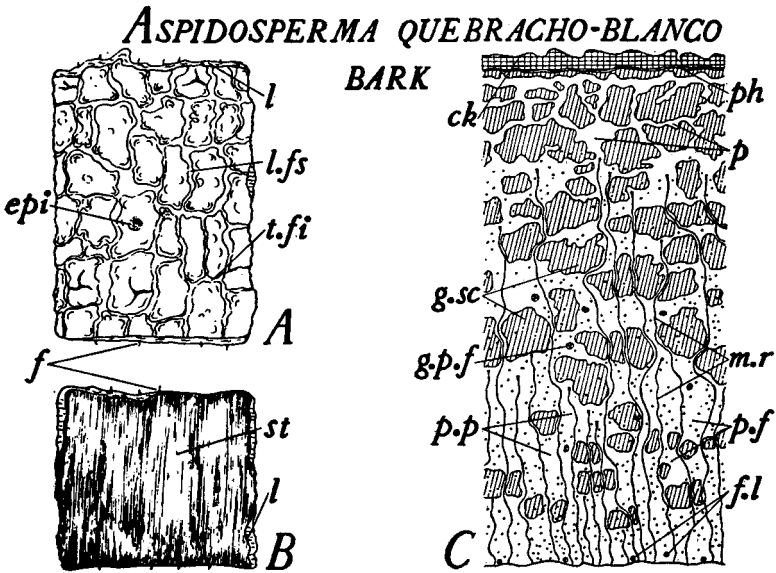


FIG. 5. *Aspidosperma quebracho-blanco* bark, macroscopical characters and T.S.:—A, outer surface  $\times \frac{1}{4}$ ; B, inner surface  $\times \frac{1}{4}$ ; C, smoothed T.S.  $\times 8$ ; *ck*, cork; *epi*, epiphyte; *f*, fibre; *f.l*, isolated phloem fibre with large lumen; *g.p.f*, group of phloem fibres with narrow lumen; *g.sc*, group of sclereids; *l*, laminations; *l.fs*, longitudinal furrow; *m.r*, medullary ray; *p*, cortical parenchyma; *ph*, phellogen; *p.f*, isolated phloem fibre with narrow lumen; *p.p*, phloem parenchyma; *st*, longitudinal striation; *t.fi.*, transverse fissure.

lumen; the sieve tubes; the parenchyma of phelloderm, cortex and phloem, also of the medullary rays which become sclerotic when associated with the sclereid groups. The calcium oxalate crystals and starch grains present agree in their structure and distribution in the two species.

**Powder.** The powder is light pink to light brown in colour and exhibits the characters described above for the entire bark. The powder resembles that of powdered *A. megalocarpon* bark in the presence of the following: characteristic sclereids in masses, some of which have granular contents that stain yellow with iodine or pinkish-red with Millon's reagent; fragments of two types of phloem fibres with either narrow or wide lumen; sieve tissue; parenchyma containing either calcium oxalate or starch. Calcium oxalate crystals are up to  $32 \mu$  in maximum length; starch



ASPIDOSPERMA BARKS OF BRITISH GUIANA. PART V

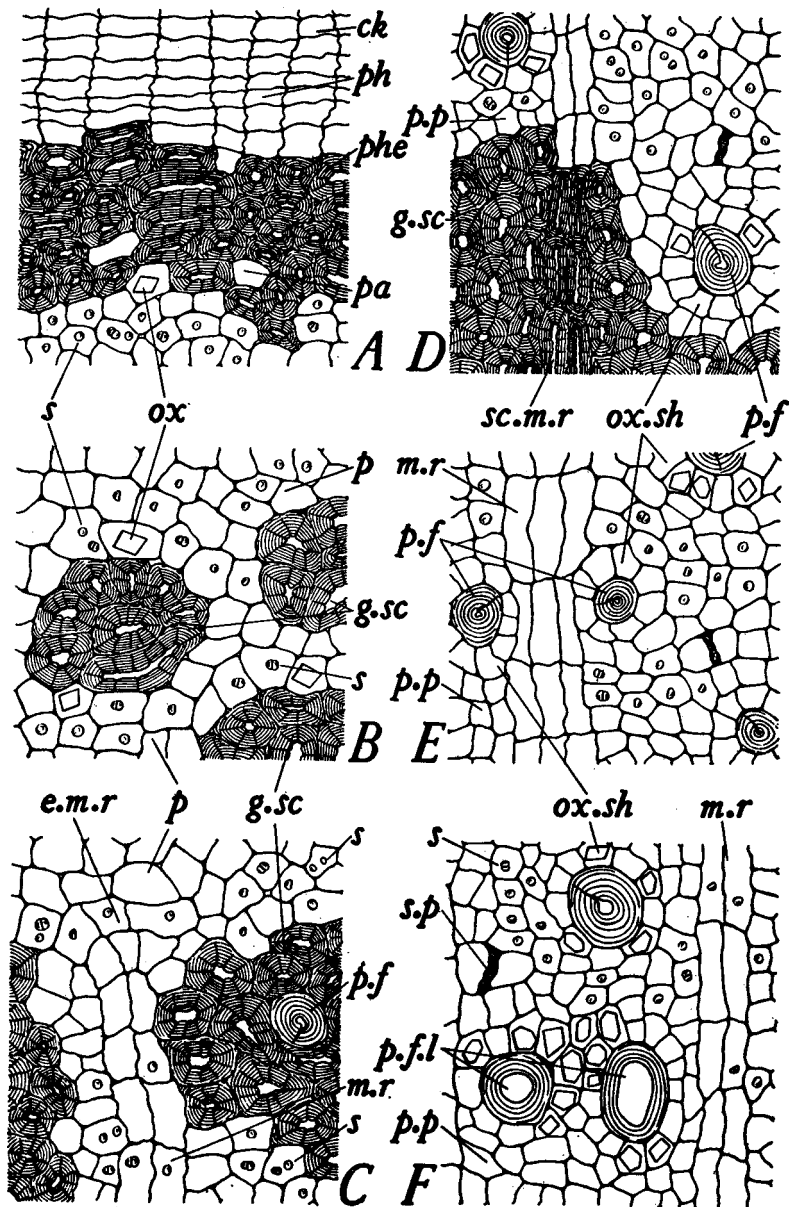


FIG. 6. *Aspidosperma quebracho-blanco* bark in T.S.:—A, cork, phellogen and phelloderm; B, cortex; C, innermost cortex and outermost phloem; D and E, phloem; F, innermost phloem; all  $\times 200$ ; *ck*, cork; *e.m.r.*, end of the medullary ray; *g.sc.*, group of sclereids; *m.r.*, medullary ray; *ox.*, crystal of calcium oxalate; *ox.sh.*, calcium oxalate crystal sheath; *p.*, cortical parenchyma; *pa.*, cortical parenchyma found associated with phellodermic sclereids; *ph.*, phellogen; *phe.*, phellodermic sclereids; *p.f.*, isolated phloem fibre with narrow lumen; *p.f.l.*, isolated phloem fibre with large lumen; *p.p.*, phloem parenchyma; *s.*, starch; *sc.m.r.*, sclerotic medullary ray; *s.p.*, sieve plate.

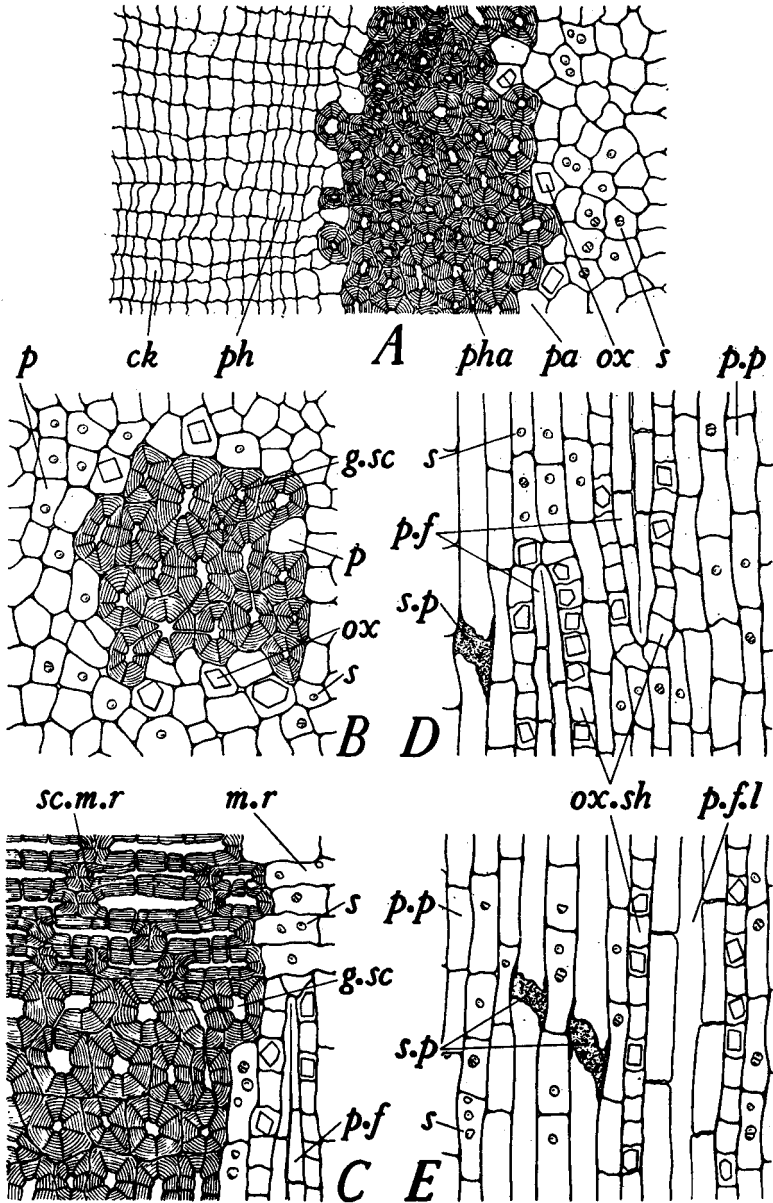


FIG. 7. *Aspidosperma quebracho-blanco* bark in L.S.:—A, cork, phellogen and pheloderm; B, cortex; C, outer phloem; D, phloem; E, innermost phloem; all  $\times 200$ ; *ck*, cork; *g.sc*, group of sclereids; *m.r.*, medullary ray; *ox*, crystal of calcium oxalate; *ox.sh*, calcium oxalate crystal sheath; *p*, cortical parenchyma; *pa*, cortical parenchyma found associated with phellogen sclereids; *ph*, phellogen; *pha*, phellogen sclereids; *p.f.*, isolated phloem fibre with narrow lumen; *p.f.l.*, isolated phloem fibre with large lumen; *p.p.*, phloem parenchyma; *s*, starch; *sc.m.r.*, sclerotic medullary ray; *s.p.*, sieve plate.

granules are simple or 2- to 4- compound, the single grains being up to  $25\ \mu$  in diameter. Powdered *A. quebracho-blanco* bark differs from that of *A. megalocarpon* in the presence of tangentially-elongated cork cells with thin walls which are unligified or only slightly lignified: fibres with large lumen are not found in association with groups of sclereids. The following cell sizes in *A. quebracho-blanco* also differ: sclereids 28 to 58 to  $72\ \mu$  in length and 14 to 28 to  $44\ \mu$  in width; phloem fibres  $H = 500$  to **850** to  $1200\ \mu$ , and  $R$  and  $T = 25$  to **46** to  $62\ \mu$ . The two powders also differ in the number of sclereids per mg. (S), the number of fibres per mg. (F) and in the S/F ratio<sup>14</sup>.

#### DISCUSSION

The diagnostic characters of the barks of three *Aspidosperma* species, namely *A. ulei*, *A. excelsum* and *A. album*, have been described and compared with those of the bark of *A. quebracho-blanco* in previous communications<sup>1,12,13</sup>. The macroscopical and microscopical characters of the bark of *A. megalocarpon* are here described and compared with those of *A. quebracho-blanco*. A complete discussion of the diagnostic characters of the barks of six *Aspidosperma* species will follow in a subsequent publication.

The diagnostic characters common to the barks of both *A. megalocarpon* and *A. quebracho-blanco* are:—

1. The cork is furrowed and fissured externally and may bear epiphytic lichens or liverworts.
2. Sclereids are present as groups or bands in the phelloderm, cortex and phloem; individual cells with thick, stratified, lignified, pitted walls and relatively narrow lumen.
3. A few sclerotic cells with somewhat large lumen contain a granular material which stains yellow with iodine and pinkish-red with Millon's reagent.
4. Sieve tubes with compound sieve plates on the oblique end walls.
5. Medullary rays narrow and becoming sclerotic when passing through the groups of sclereids.
6. Phloem fibres, of two types, the majority with narrow lumen and either isolated or in groups of two to three; fewer, with large lumen, always isolated. Both types of fibres large, spindle-shaped, with thick, stratified and lignified walls, traversed by a few simple or compound pits.
7. Starch, simple or 2- to 4- compound; individual grains with eccentric hilum, spherical, ovoid or plano-convex.
8. Prismatic calcium oxalate crystals in a parenchymatous sheath surrounding the fibres or groups of fibres, also at times in association with the sclereid groups or bands.

The characters diagnostic of the bark of *A. megalocarpon* are:—

- 9a. Cork cells thickened and lignified on the inner and outer tangential walls.
- 10a. Sclereids arranged in very small groups in the phelloderm and in four to six more or less continuous bands in the cortex.

11a. Phloem comprising up to 50 per cent of the thickness of the bark ; groups of sclereids are present in the outer half only of this phloem but fibres with either narrow or wide lumen are scattered throughout.

The characters diagnostic of the bark of *A. quebracho-blanco* are:—

9b. Cork cells thin-walled and unligified or very slightly ligified.

10b. Sclereids arranged in a more or less discontinuous band in the phelloderm and in groups of varying sizes in the cortex.

11b. Phloem comprising up to 70 per cent of the thickness of the bark, throughout the majority of which are abundant groups of sclereids and fibres with narrow lumen ; two narrow bands, devoid of sclereids, occur towards the inner region of the phloem, both containing fibres with narrow lumen but the innermost also contains a few fibres with wide lumen.

The thickened and lignified walls of the cork and the occasional sclereid groups enclosed in the corky layer of *A. megalocarpon* distinguish this bark from the barks of *A. ulei*<sup>1</sup> and *A. album*<sup>13</sup> in which the cork cells are unligified or very slightly ligified ; in the bark of *A. excelsum*<sup>12</sup>, both ligified and unligified cork cells are found. The absence of latex canals, presence of sclereids with granular material which stains with iodine and Millon's reagent, the presence of sclerotic medullary rays and the scattered fibres with large lumen throughout the phloem, also seen associated with sclereid groups, all differentiate the bark of *A. megalocarpon* from those of the three species named above.

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