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ORIGIN AND DETECTION OF MUCILAGE IN PLANTS.

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By the term "mucilage in plants" is meant those substances which are soluble, or at least swell very perceptibly in water, and which, upon the addition of alcohol, are precipitated in a more or less amorphous or granular mass. Mucilage originates in the plant either as a part of the contents of the cell or as a part of the wall thereof. When it occurs as a part of the cell contents (as cell-sap), mucilage is produced either as an "Auscheidung" from the protoplasm, or it may possibly arise in some cases as a disorganization product of some of the contents. When it occurs as a "membrane mucilage," it owes its origin to several causes, viz.: either as a form of secondary thickening or addition product to the wall; or as a metamorphosis of the cell wall, at least in part. In the latter case it may arise either as a disorganization product of the primary wall as an intercellular substance; or of the subsequent lamelae making up the pith, medullary ray, parenchyma and other cells. In addition to these two well-authenticated cases of the origin of mucilage, viz., as a cell contents and cell-membrane, we have mucilage given out by some secreting hairs (glands). In such instances we can simply say that the mucilage appears between lamellae of cutin on the one hand and of cellulose on the other hand, of the epidermal cells of the secreting hair.[1]

For convenience of reference, the following table, containing some of the important official plants yielding a mucilage, together with the origin of the same, is given:

I. ***Cell-Contents Mucilage***.

1 Corm of *Orchis* sp. (Salep).
2. Rhizome of *Agropyrum repens*, L., Beauvois (*Triticum*).
3. Bulb of *Urginea maritima*, L., Baker (Squill).
4. Bulb of *Allium* sp. (onion, garlic).
5. Stem, leaf and elements of flower, excepting stamens, of *Viola tricolor*, L.
6. Flower-stalks of *Hagenia abyssinica*, Bruce, Gmelin (Cusso).
7. Pulp of fruit of *Musa paradisiaca* (Banana).
8. Succulent plants, as Aloe, etc.

II. ***Cell-Membrane Mucilage***.

A. ***Secondary thickening of wall***.

1. Root of *Althaea officinalis*, L., (Althaea).
2. Bark of *Cinnamomum* sp. (Cinnamon).
3. Bark of *Rhamnus Frangula*, L. (Buckthorn).
4. Bark of root of *Sassafras variifolium*, Salisbury, o. Kuntze (Sassafras).

[1] De Bary, *Vergleichende Anatomie*, S. 98.

5. Inner bark of *Ulmus fulva*, Mx. (Ulm).
6. Leaves of *Barosma betulina*, Thunberg, Bart. et Wend, and *B. crenulata*, L., Hooker (Buchu).
7. Seed-coat of *Cydonia vulgaris*, L., (Quince).
8. Seed-coat of *Linum usitatissimum*, L. (Flaxseed).
9. Seed-coat of *Sinapis alba*, L., Hook. fil et Thompson, and *S. nigra*, L., Koch (White and Black Mustard).

B. ***Metamorphosis of Cell-wall***.

a. ***Pith and Medullary Ray Cells***.

1. *Astragalus* sp., yielding Tragacanth.[2]

Ba. ***Parenchyma cells of wood and bark***.[3]

1. Cherry-gum, yielded by some of the Amygdalaceae.

Bb. ***Various cells of the bark***.

1. *Acacia Senegal*, Wild., yielding Gum Arabic.[4]

Bc. ***Primary wall as intercellular substance***.

1. Thallus of *Chondrus crispus*, Stackhouse, and *Gigartina mamillosa*, J. Agardh (Irish moss).

III. ***Secreting Hairs (Drüzenzotten)***.

1. Leaf and calyx of *Viola tricolor*, L.
2. Leaf of *Coffea arabica*, L. (Coffee).
3. Leaf of *Prunus avium*.[5]

The origin of mucilage as an "Auscheidung" from the protoplasm has been pointed out in the corms of various species of *Orchis*, by Frank,[6] Meyer[7] and Hartwich.[8] All have contributed more or less confirmatory researches toward this end. Frank says that in the young cells of the tuber of *Orchis*, near the nucleus occurs a bundle of needle-like crystals, around which a clear mucilage drop is formed. This becomes larger and larger, and finally when fully grown replaces the protoplasm and nucleus, and occupies the lumen of the cell. Arthur Meyer, on the other hand, claims that the protoplasm in *Orchis* species does not disappear, but that it is to be found in the older stages of the cell, and that the peripherical layer of protoplasm secretes the mucilage which lodges in between the "plasmanetz" (ie., in the vacuoles). Hartwich confirms the labors of Meyer in that he finds the peripherical layer of protoplasm in the latter stages of the life of the cell, but he does not observe the "plasmanetz." He also says, in cells of *Orchis latifolia* and *O. Morio* that do not contain raphides, the mucilage does not arise in the middle of the cell.

It is not unlikely but that in all of the plants mentioned as having a "cell-contents mucilage," and which contain rhapides of calcium oxalate as scilla (*Fig. 6*), that the origin is similar to that of Salep, viz., as an "Auscheidung" from the protoplasm.[9]

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[2] Mohl, *Bot. Zeit*., 1857, S. 33.

[3] Tschirch, *Angewandte Pflanzenanalomie*, S. 210.

[4] Wigand, Pringsheim's jahr., 111, 115, Moeller, Wiener Akad., 1875, 219. Tschirch, *loc. cit.*, 213.

[5] J. Reinke, *Bot. Zeit*., 1874, 47 and 59.

[6] *Pringsheim's Jahr. f. wiss. Bot*., V, p. 161.

[7] *Arch. d. Pharm*., 1886, P. 325.

[8] *Arch. d. Pharm*., 1890, P. 563.

[9] See also Tschirch, *loc. cit.*, S. 109, 125.

In *Viola tricolor*, L.[10] is found, as has been previously shown by the author, peculiar sub-epidermal mucilage cells, which occur in all the leaf-like elements of the plant with exception of the stamens.

These are easily ascertained by placing fresh specimens of the leaf in an alcoholic solution of methylene blue of the following strength:

ALCOHOLIC METHYLENE, BLUE SOLUTION.

Methylene blue . . . . . 0.400 grammes.
Alcohol (95 per cent.) . . . . . 100 c.c.

The specimen is left in this solution for at least several hours, after which surface or transverse sections may be made and are transferred to a slide having a few drops of the following mixture:

GLYCERIN METHYLENE BLUE SOLUTION.

Methylene blue . . . . . .0.400 grammes.
Alcohol (95 per cent.) . . . . 20 c. c.
Glycerin (nearly anhydrous) . . . . . 80 c. c.

The cover-glass is put upon the section, and in a short time the mucilage cells are stained blue (as *Figs. 1, 2, 3*) and are readily distinguished from the remaining unstained cells. Preparations so prepared may be kept indefinitely. In fact in a few weeks the contrast is even more marked than on the first day.

The nature of the contents of the sub-epidermal mucilage cells may be best studied by sectioning alcoholic material and mounting in alcohol. If to a preparation of this kind an alcoholic solution of iodine is added, the protoplasm nucleus and plastids become light brown in solor. If to a transverse section in alcohol (as Fig. 4) is added chlorzinciodide, the mucilage gradually swells, and the subepidermal cell becomes about twice the size as when seen in alcohol. The wall between the two cells becomes blue, but the mucilage is not altered in color.[11] Surface sections of the leaf, treated with an alcoholic iodine solution, and then with an alcoholic sulphuric acid solution (containing two volumes of alcohol to one volume of concentrated H_2SO_4), allow for a closer study of the nature of the contents.

(*Fig. 5*) The nucleus is central and is surrounded by a small amount of cytoplasm. From here to the peripherical layer of the protoplasm extend numerous threads of the same substance. In the periphery are a number of small plastids, being fewer and smaller than in the epidermal cells.

The fresh pulp of banana fruit treated with an alcoholic methylene blue solution, and from which sections are made and examined in a glycerin methylene blue solution, indicate that along the phloem portion of each of the fibro-vascular bundles we have a

[10] *Inaugural Dissertation*, Marburg, 1897.

[11] For behavior of mucilages toward iodine compounds and other reagents, see Arthur Meyer, Wissenschaftliche Drogenkunde, S. 47 and Tschirch, *loc. cit.*, S. 206

chain of cells (four to six times larger than the starch-containing parenchyma cells) which contain a mucilage. They take on a deep blue color (*Fig. 7*) and occur only associated with the fibro-vascular bundles. Their position and arrangement lead to the opinion that they are in the nature of membered mucilage tubes similar to the tannin tubes of the oaks.

The origin of "cell-wall mucilage" may be best studied in the young roots and stems of althaea.[12] In the very young elements the mucilage cells are found to be much larger than in the surrounding parenchyma. If in this stage sections of fresh material are treated with alcohol, there is no indication of any of the cells containing mucilage. A little later, however, we find in older specimens that, in addition to the protoplasm and nucleus, there is a granular layer formed on the inside of the wall. Upon the addition of water this layer swells, and upon subjecting it to the methylene blue treatment it is found to be a mucilage layer. The cell wall grows in thickness by the successive depositions of lamallae of mucilage, and finally in old cells we observe only a small lumen with a little protoplasm, the mucilage wall occupying the remainder.

The study of these mucilage cells may be performed on commercial specimens of althaea root. If the latter are first washed in water until softened, and then put into strong alcohol, we have material that may be easily sectioned. Sections are made and are put into an alcoholic methylene blue solution for several hours or longer. These are then mounted on a slide in a glycerin methylene blue solution. In the course of a few hours the mucilage cells have taken on a prominent blue color and are distinct from the other parenchyma cells (*Fig. 8*). The lamellae are pronounced, and in the centre a small, irregular lumen with a small amount of protoplasm is observed.

If a specimen of elm bark of commerce is treated similarly to that of althaea root, equally, if not more, characteristic are the blue colored mucilage cells (*Fig. 9*).

Mucilage occurs as a secondary thickening beneath the cuticle of a number of seeds, as Sinapis alba (*Fig. 10*) and Linum (*Fig. 11*), etc. If sections of the dry seeds are mounted first in a solution of glycerin and water and then a glycerin solution of methylene blue added, the mucilage layer becomes a permanent blue color, stronger in Sinapis alba than in Linum.

The secretion hairs (or glands, as they are sometimes called) which we find at the apex of the divisions of the leaves of *Viola tricolor*, L., and other plants, yield a mucilage. In the leaves of *Viola tricolor*, L., at the apex of the divisions of the lamina and stipulae, we find large secretion hairs. These may possess a head-like portion alone, or may have in addition a stalk. They arise very early, especially upon the stipulae. They serve some function in the developing lamina and then disappear. Later are formed similar hairs upon the crenate margins of the lamina. When not fully mature, the epidermal cells of the secretion hairs possess a relatively large amount of protoplasm, a large nucleus and a few rather large vacuoles (*Fig. 12*). When fully matured the secretion hair usually resembles that shown in *Fig. 13*. The upper portion or head is rounded and consists of two kinds of cells, those upon the periphery being larger and of irregular shape, those below these being smaller and nearly isodiametric. The cells of the stalk resemble closely those of the remaining

[12] See Tschirch, *Anatomischer Atlas*, Lief. VI, S. 128.

portion of the leaf. The epidermis of the upper portion or head is thicker than that of the stalk and consists of lamellae of cutin and cellulose between which, however, later arises the mucilage. If a hair that is about to discharge its mucilage is removed from the leaf and mounted in a few drops of picronigrosin or basic lead acetate solution, or glycerin methylene blue solution and slight pressure brought to bear upon the cover-glass, the cutin layer is ruptured, the mucilage is discharged, becoming faint blue with picronigrosin or methylene blue solution, or granular and almost colorless with basic lead acetate solution.

The use of methylene blue as a reagent for mucilage in plants, as outlined by the author, has the advantage that it is decisive, as only some lignified cell-walls otherwise take up this color, and it may be applied by proper manipulation to dry as well as fresh plant material.

DESCRIPTION OF ILLUSTRATIONS.

(1) Surface section of the epidermis of the under surface of the leaf of *Viola tricolor*, L.

(2) Surface section of the epidermis of the upper surface of the leaf *Viola tricolor*, L.

(3) Surface section from the under surface of the spurred petal of *Viola tricolor*, L., var. *vulgaris*, Koch.

(4) Transverse section of leaf of *Viola tricolor*, L., showing epidermal cell and sub-epidermal mucilage cell.

(5) Surface section of sub-epidermal mucilage cell of *Viola tricolor*, L., treated with alcoholic iodine and sulphuric acid solution.

(6) Longitudinal section through a commercial specimen of a fleshy scale of bulb of scilla, showing mucilage layer around acicular crystals of calcium oxalate.

(7) Longitudinal section through the pulp of banana fruit near a fibro-vascular bundle, showing large membered mucilage secretion cell.

(8) Transverse section through a commercial specimen of *Althaea* root treated as described in the test, showing large mucilage cells with lamellae and irregular protoplasmic contents.

(9) Transverse section through a commercial specimen of *Ulmus* treated similarly to the *Althaea* specimen. The large mucilage cells are surrounded by parenchyma cells containing starch and calcium oxalate crystals and a few bast fibres.

(10) Transverse section of seed coat of *Sinapis alba*, showing outer mucilaginous wall.

(11) Transverse section of seed coat of linum. showing outer mucilaginous wall.

(12) Nearly mature secretion hair on leaf of *Viola tricolor*, L. The cells upon the periphery contain a large nucleus, protoplasm and a few relatively large vacuoles.

(13) Fully matured secretion hair on leaf of *Viola tricolor*, L., with stalk.

(14) Mature secretion hair of *Viola tricolor*, L., that was ready to discharge its mucilage. Upon mounting in a glycerin methylene blue solution or in picronigrosin and pressing upon the cover-glass with a needle or pencil, the mucilage is discharged and takes up the stain.

FIG. 1.

FIG. 2.

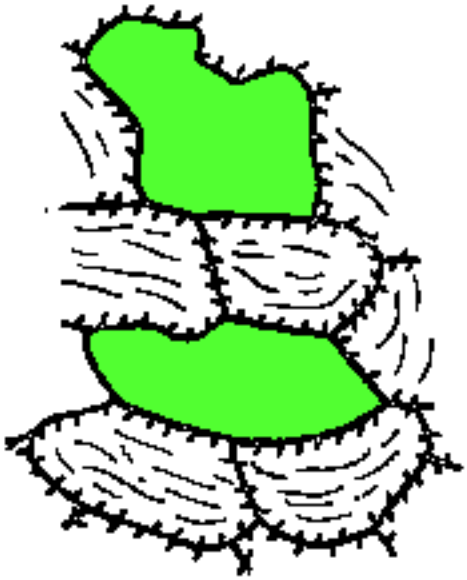

Fig. 3.

Fig. 4.

Fig. 5.

Fig. 6.

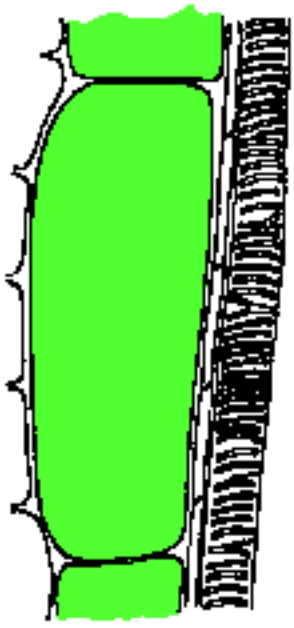

Fig. 7.

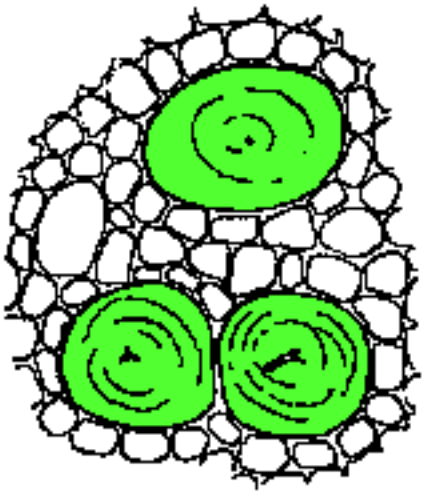

FIG. 8.

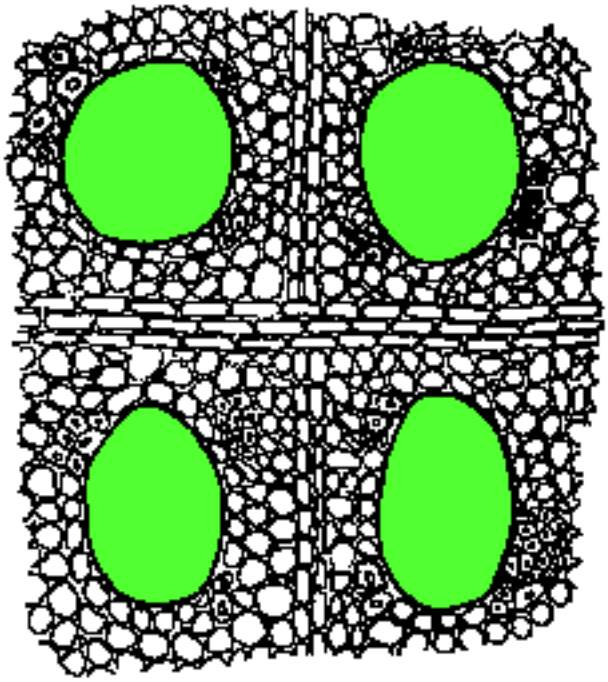

FIG. 9.

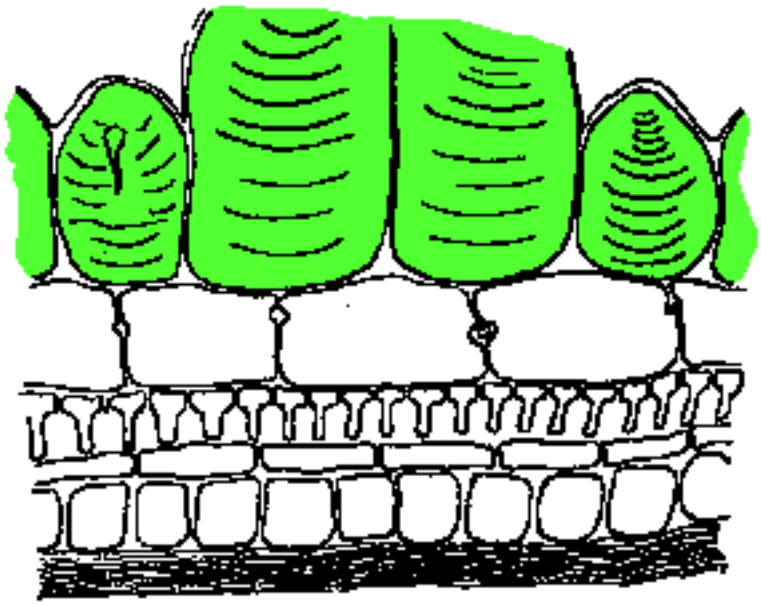

FIG. 10.

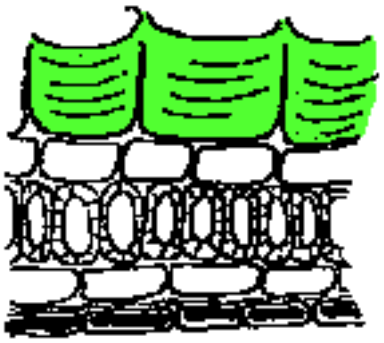

FIG. 11.

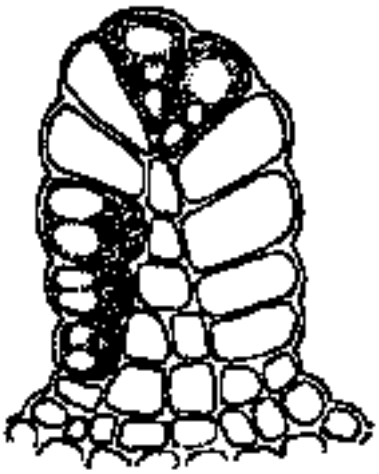

FIG. 12.

FIG. 13.

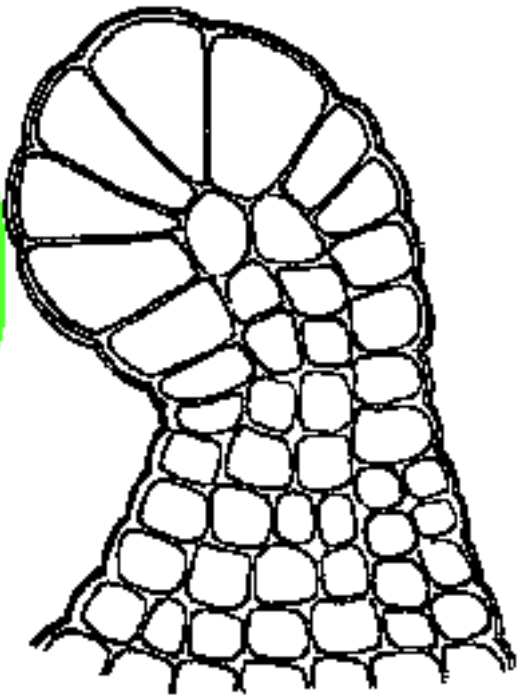

FIG. 14.