The Structure and Diagnostic Value of the Starch Grain.

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In view of their common occurrence in plant tissues, starch grains have been used, especially in Pharmacognosy, to differentiate between plants. While there is a great variation in the size, shape and structure of starch grains, those of different members of a genus, or even of a family, often show a similarity. Hence, these group characteristics often may be used to identify a given starch as belonging to a certain group of plants. As medicinal action or value varies greatly between closely related members of the same genus, it is of the highest importance to establish the authenticity of the species, and in this determination the starch grain is often of the greatest diagnostic value.

The characters most often used in the identification of starches are the size, shape, and markings of the grains. The most distinct markings are the hilum and the concentric layers of starch. It is very commonly stated that the hilum is the point of attachment of the grain, and that it occupies a position on the surface, while in reality it is the part first formed, and is marked by a fissure or cleft in the interior, caused by the loss of moisture, and shrinking of the central portion.

The starch grain has a structure somewhat similar to that of the sphaero-crystal, and like it grows by the apposition of new materials. According to the best authorities, the grain is made up of minute crystals or miscellæ of soluable starch, or granulose, imbedded in a frame work of starch cellulose. Alternate layers seen in many grains, contain a greater proportion of granulose, and hence stain more deeply with iodine solution. This structure of the grain was demonstrated by the action of such solvents as chloral hydrate or diastatic solutions which dissolve the granulose very rapidly, and leave a framework of starch cellulose, of the same size as the original grain, but lacking the substances which produce the characteristic color with iodine solution.

Hence, is was stated by Meyer that the soluble starch was distributed throughout the grain in very small crystals or trichites. A consideration of the behavior of the grain upon swelling certainly demands a structure of this nature. The methods used in the preparation of this paper, and the results obtained agree with this structure, but indicate that the crystals

or trichites of soluble starch may be of a considerable size. Since the crystals have nearly the same refractive index as the remainder of the grain, they cannot be detected without special treatment.

The structure of the starch grain is most easily seen in such grains as those of the common potato, or of arrowroot, which show the layers of starch most plainly, or those of Bamboo Brier root, which show the characteristics of the central portion. When these starches are stained with certain dyes, of which safranin is the best example, and the excess of stain is extracted, the central portion retains the stain, and the crystalline nature of this portion becomes evident. In the center of the grain crystals are seen arranged with their longer axes extending radially from the hilum. The clefts in this crystalline mass mark the hilum, and the fissures caused by the loss of moisture follow the longer axes of the crystals, forming the fan-shaped markings often seen in the unstained grain. Fig. 1. When the grains are kept moist at 50° C. for some time, the moisture passes through the outer layers, causing but little change in their structure, but when the crystalline portion is reached, this swells up and slowly dissolves from its surface, becoming surrounded by a zone which stains deeply with iodine solution, indicating the presence of soluble starch. The solution and swelling of this inner portion pushes upward the outer layers, which finally rupture at the thinnest point, that is, at the hilum end in eccentric grains. It therefore appears that the greater moisture content of his portion is due to its higher solubility, and indeed this part may be entirely dissolved and carried away in solution before the outer layers are affected. This is commonly the case in such grains as those of the common yam, or of Bamboo Brier root. Fig. 4.

Something of the chemical nature of the various layers of the grain can be determined by treating with the following easily reducible silver solution, which may be compared to Fehling's alkaline cupric tartrate solution:

Silver Nitrate	1 gm.
Water	15 cc.
Ammonia water	q. s.

Add the ammonia water to the solution of the silver nitrate until the precipitate first formed just dissolves.

Potassium and Sodium Tartrate	2 gm.
Water	15 cc.

Dissolve.

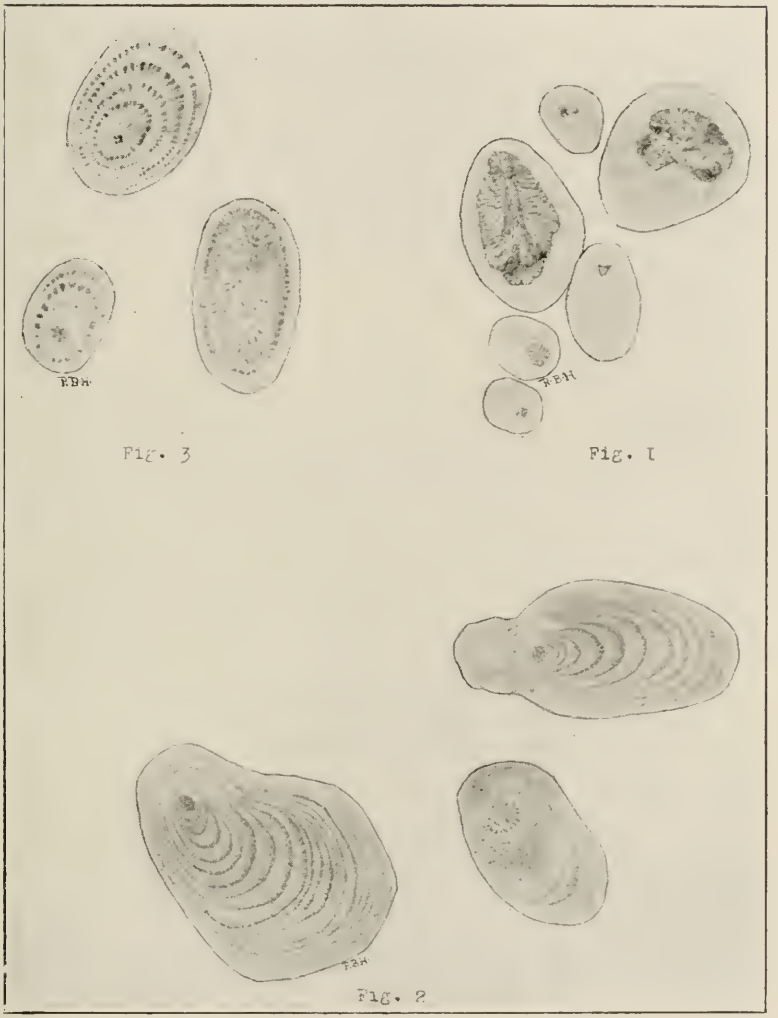


Fig. 3

Fig. 1

Fig. 2

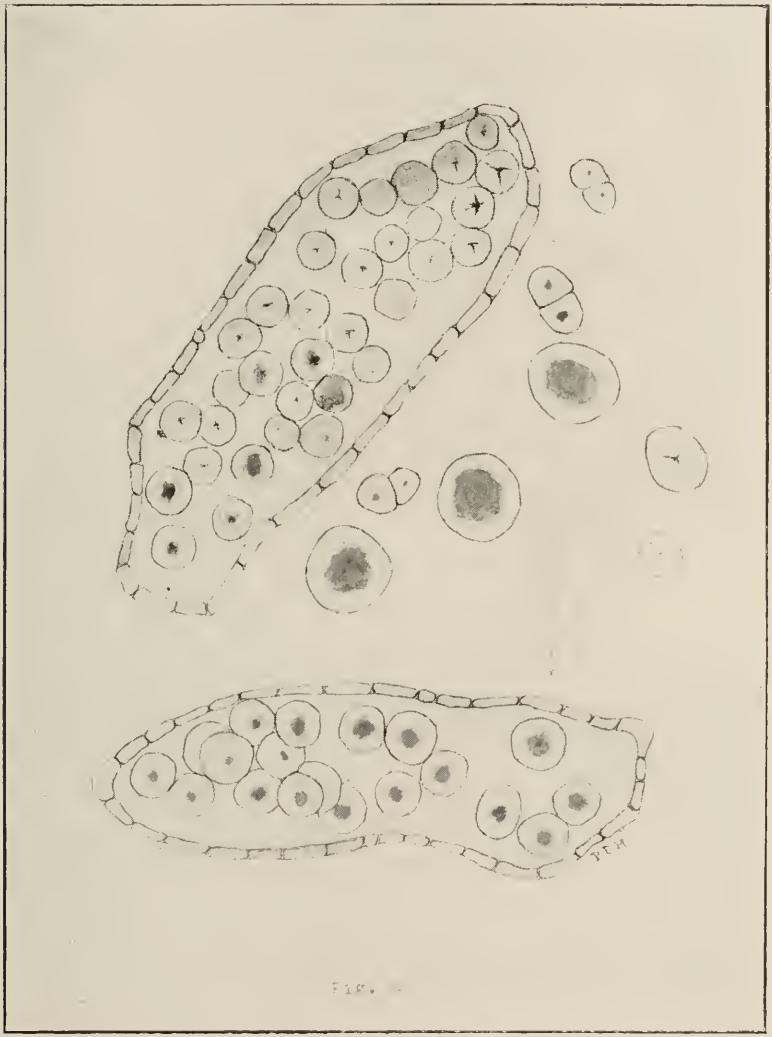
When starch grains are treated for a time with a small amount of the ammoniacal silver solution to allow the silver salt to be evenly distributed throughout the grain, and then an equal amount of the tartrate solution added, the silver salt is reduced by the granulose present in the grain, giving stains varying in color from purple through shades of brown to black, depending upon the state of reduction. Reduction occurs in a few hours at ordinary temperature, or may be quickly accomplished by warming very slightly although too strongly heating causes complete reduction of the solution and total blackening of the grain.

If it is desired to make permanent mounts by the above method, the starch should be stained in bulk until the proper depth of color is reached, and then the soluble and unreduced silver salt washed out with water to prevent further reduction. The grains may then be dehydrated and mounted permanently in balsam.

When starch grains are treated according to the above method, alternate layers and the central portion reduce the silver solution most rapidly, showing the presence of reducing substances which cause a deposit of silver. Fig. 2. The alternate layers which stain most deeply with iodine solution take on a granular appearance, and show the presence of crystals. The location and size of these crystals become more distinct upon further treatment of the mount with 1% chromic acid solution. Then they are seen to be of considerable size in some grains, and often to be arranged in a group with their longer axes radiating from the hilum. Quite large crystals occur toward the outer portion of each layer, and their presence accounts for the difference in refraction of the layers at this point, which produces the appearance of concentric rings. Fig. 3.

In double or compound grains a crystalline mass is seen at the center of each part, indicating that compound grains are formed by the deposition of material around a number of points of crystallization, and the subsequent growth of each part until fusion occurs. In the outer layer of the grain no crystals appear, and this portion seems to be made up for the greater part of starch cellulose, which explains its lesser solubility in diastatic solutions. Fig. 3.

An application of the differences in the structure of starch grains is of value in the examination of such closely related species as the common sarsaparillas of the genus Smilax. All the members of this group, which are commonly met with, have similar histological structures. All show the presence of raphides crystals of calcium oxalate and parenchyma cells



filled with starch grains, which are commonly spherical, or united in groups of two or three grains. Honduras sarsaparilla, Smilax officinale, is characterized by starch grains varying from 7 to 20 microns in diameter, and raphides crystals 6 to 8 microns in length. The common Bamboo Brier root shows similar characteristics when examined in fine powder, but shows a variation in the starch grain. The starch grains of this drug greatly resemble those of Honduras sarsaparilla, being either single or united in groups, and show a similar structure in the interior of the grain; but upon measurement they range in size from 9 to 40 microns, averaging 29 microns in diameter. Hence, this means may be used in the differentiation of these plants when the drug is examined as a fine powder.

By the application of the above or of similar methods, it is possible to differentiate between very closely related plants. As our knowledge of the structure of starch grains is more fully developed, their value in the differentiation of such closely related species becomes apparent, and they are recognized as one of the greatest aids in Pharmacognosy.

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