The „Silverthread" Disease of Coffee in Surinam

by

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(With Plate VII and VIII.)

Introduction.

The Surinam coffee (*Coffea arabica*) as well as Liberian Coffee (*C. liberica*) is attacked by a disease, which manifests itself by the hanging of one or more dead leaves on the top of the branches. These leaves are attached to the branch by a thread, which is formed by a great number of twisted hyphae; on further examination the branch shows, besides the leaves hanging from it, which are quite withered, some leaves with smaller or larger brown spots. On the underside of these leaves a cobweb-like tissue of hyphae is found, which in dry weather on leaves, not yet badly attacked, has a silvery white appearance, hence the name „silverthread" (Dutch: zilverdraad) has been given to the disease.

Usually bundles of hyphae spread out on both sides of the midrib; as they proceed further up the leaf they gradually spread as a delicate web over the whole leaf surface. In the other direction one can follow the strand along the branch, often even far down the mainstem of the tree.

I noticed the disease on all the estates which I visited.

Diseases such as this have been described by several

investigators: Ernst[1]) speaks of a disease, which he found in Venezuela and which is called there „candelillo"; it is identified by him with the „coleroga" of British-India, studied by Cooke.

Zimmermann[2]) deals with a „spinnewebziekte" (cob-web-disease) which exists in Java; an illustration is added to the description, which shows a great likeness to the silverthread

In the Annual Report of the Porto-Rico Agricultural Experiment Station for 1904, a similar disease is mentioned, of which Earle gave the first description and which later was investigated by Clinton.

The question, whether we have in all these cases the same disease, will be dealt with in detail further on.

I express my thanks to the agent of Royal West India Mail at la Guayra for kindly sending me leaves attacked by candelillo and to Dr. Clinton, who kindly gave me some information on the subject.

Description of the fungus, the culture-experiments and artificial inoculations.

a. Development and growth on the plant.

The fungus is to be seen only on the underside of the leaves; in very rare cases one may see some hyphae bending round the margin of the leaf and extending over a small patch on the front, but then I always was able to prove, that some leaves had touched each other, so

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1) A. Ernst, Botanische Notizen aus Caracas, Botanisches Centralblatt, 1880, 1er Jahrgang, S. 1178.

2) A. Zimmermann, Eenige pathologische en physiologische waarnemingen over koffie, Mededeelingen uit 's lands Plantentuin 67, 1904, pag. 46.

that a very moist atmosphere existed between them; most of these cases occur where some leaves on the top of a branch already killed by the fungus, are hanging on the rhizomorphous strands. (Fig. 2). For one can always see, that a myceliumstrand, which looks almost like a rhizomorph, passes from the branch to the leaf. This strand can be followed rather far over the leaf surface; first following the midrib, then crossing the leaf blade. (Fig. 1 and 2).

The strand, before passing from the branch to the leafstalk, first spreads over the thicker parts, which are formed round each node; it forms a very thin membrane, from which in the majority of cases several strands originate; one of them runs to each leaf, one to each of the axillary shoots, (when these are already developed) and one or more to the top of the branch. The development of the fungus is always acropetal; nor did I succeed in causing a basipetal growth by artificial inoculations. One may follow the strands along the branches in succeeding order down to the mainstem, but I never succeeded in finding the fungus less then one or one and a half feet from the ground, notwithstanding I investigated a good number of trees. Nor did I notice any sign of hyphae on the roots of plants, which were heavily attacked.

The colour of the strands varies from light to darkbrown; on the younger branches the strands may be lifted up as well as the delicate membrane on the leaves. Because this membrane is so very delicate, the best manner to take it from the leafsurface is by holding one of the rhizomorphous strands with tweezers.

When the leaf is still green, one may see the cobweb as a silvery-white tissue; very soon the colour grows darker, so that when the parts, which have been attacked are dried up and dark brown, the hyphae show a somewhat

lighter brown colour. The spots then look as if a brown powder had been strown over them; it might be called a granulated appearance. This description refers particularly to the Liberia coffee; in Coffea arabicà the colour of the hyphae soon ressembles that of the dead leaf.

The granulated appearance of the leaves is caused by the formation of knots of hyphae, which manifest themselves as little yellowish brown spots.

The hyphae are very thick (5—7 μ) and run generally quite straight over the leaf surface, they have a number of short branch-hyphae, which make wide angles with the main hypha; so the microscope shows us a field, divided into almost rectangular figures (Fig. 5).

At certain places however accumulations of hyphae arise, when the straight hyphae throw out side branches, which in their turn make side branches of the second order. The small branches of different hyphae meet each other and become twisted together; so the little nodules are formed, which are visible to the naked eye (Fig. 5, 6, 7h).

As mentioned before, one finds the fungus already well developed on leaves, which do *not yet* show the discoloured spots. So the question arises how it can be explained that the spots are not seen at the same time.

Microscopical research showed, that hyphae are never seen *in* the green parts of a leaf, over the surface of which the membrane is already widely spread; they are found on the contrary in the brown spots; the hyphae stretch themselves just to the outside of the discoloured parts. A transverse section taken at a place where the silvery tissue was already well developed, but the brown colour was just noticeable, will throw some light over the manner in which the fungus damages the leaves. One may then see, that accumulations of hyphae are formed above the stomata, which lay a little under the leaf surface; then

the guard-cells become brown-coloured, i. e. they die off. Before these cells are quite dead, the hyphae do not penetrate the leaf tissue; first they grow intercellularly and not before a larger number of the surrounding cells have been killed, do they establish themselves there in and fill them up.

As a rule this process takes place very quickly, as will be seen from the artificial inoculations which will be described later on. This is also the cause, why it is rather difficult to find an exemple of this stage of the disease.

The nodules of hyphae giving the peculiar aspect to the leaves, lay for the greater part over the stomata and apparently the death of the leaf is caused by suffocation.

It is a curious fact, that, while coffee-leaves have stomata only on the underside, the epiderm on *both* sides becomes brown before the interlaying parenchymatous tissue.

In Liberian coffee in the first stage rather small spots develop, often along the margin, but also half way up the leaf; and these slowly extend themselves, till the whole leaf becomes withered. In Surinam coffee on the contrary usually the middle part of the leaf withers transversely immediately; then gradually the remaining parts, the apex and the basis, become brown. When the leaves are totally or almost totally withered, the stalk breaks quite near the node; at first I thought that the leafstalk was attacked primarly and that this caused the shedding of the leaves. Experiments showed this hypothesis to be false; when one allows a coffee branch with leaves to wither, they always fall in this manner. By microscopical observation dead cells are found under the strands on the stalks, but only superficially and not in such quantities that they do any damage.

b. Culture experiments.

Pure cultures of the fungus are easily procured by taking small pieces of the membrane and transferring them to a suitable medium. A still better way is to place a leaf which has been attacked in a damp glasbox; the fungus then develops an abundance of airhyphae which may easily be transferred purely to the nutritive medium. The best medium appears to be potato-agar, prepared according the following recipe of Appel and Wollenweber:

10 % of glucose
1½ % „ agar
1 % „ citric acid.

It produces an abundant growth, much better than a medium of only inorganic constituants. It may here be mentioned, that a fructification was never found in any of my media, though I used a great variety of them; so it is necessary to give a description of the mycelium as accurately as possible.

The most typical thing of the mycel is the formation of short branch hyphae, which bifurcate close to the main hyphae (Fig. 7*i* and *j*). The branches of the first order are often strongly septate, so that a number of almost cubic or globular cells arises (Fig. 7*e*).

Anastomoses of the branches of the same hyphae as well as of different hyphae very often occur. The phenomena, here described may be seen very clearly in coverslip-cultures with coffee-decoct, nutritive agar or with water. The figures 6 and 7*a—j* give a survey of the most typical forms of the mycelium. At places, where a good many side-branches have been formed, they twist together, this is the origin of the knots described above (Fig. 6 and 7*h*). Besides those knots hyphae spirally twisted may often be found in agar (Fig. 7*c*).

In the knots and at the top of the short branches (Fig. 7*a*) round or ovalshaped cells often appear, which by their form give the impression of conidia; they contain a large quantity of plasmaic matter and are strongly vacuolised. Such cells are also found in the mycelium on the leaves, and when isolated by scratching a withered leaf with a needle, or shredding a piece of mycelium, they germinate easily. So I suppose that they play an important part in spreading the disease.

Fig. 8 shows some cases in which new hyphae are originating from such isolated cells. Undoubtedly also the direct contact of diseased and healthy leaves is an important factor in the process of infecting new branches, for in a moist atmosphere the fungus passes very easily from one leaf to another. I think that leaves may become loose, and be blown by the wind to other trees which they will infect if the conditions are favourable. When dead leaves are hanging from a branch only connected with a mycelium-strand it may often be seen that they touch new healthy leaves and that the hyphae begin to spread from the place where the leaves meet.

As I have already written, I never noticed any fructification, neither in the natural substratum, or in a damp chamber or any nutritive medium. Of all the different media potato-agar gave the best results; besides this I cultivated on sterilised coffee-branches, beanstalks, potato and carrot slices. In coffee-branches the above mentioned knots developed so abundantly, that almost globular bodies with 1—2 mM diameter arose; these consisted of a rather loose tissue of hyphae. When young they sometimes remind one of the necator stage of djamoer oepas; then their colour is white, but it soon becomes greyish or brown. Thinking that the fungus might belong to the Basidiomycetes, I started large cultures on bread with different

quantities of glucose in Erlenmeyer's conical flasks of 450 cM³.

Not one of these cultures which I kept from July 1911 till May 1912 showed any sign of spore-or fruitbody formation.

c. Artificial inoculations.

Infection is accomplished very easily by binding round a branch leaves already attacked; under favourable conditions c. q. a moist atmosphere the fungus develops rapidly.

On June 30th in this manner a branch of Liberian coffee was infected; above the place of inoculation it bore 8 leaves. On July 21st four leaves were already attacked so bad that they showed brown patches; on July 28th these patches had become larger and the fifth leaf was attacked; here the filament was still white, and no discoloured spot could be seen; but the following day, July 29th, the leaf had a large brown spot; this proves that death by suffocation, (the stomata becoming clogged) occurs very quickly, so it can be understood, that this stage is not easily found.

On August 6th all the leaves showed a bad infection while the day before no brown colour was seen on the eighth and last leaf.

Infection from a pure culture cannot be managed so easily; I obtained some good results by keeping freshly cut branches under a glass bell very moist, after putting little pieces of agar with the mycelium on the leaves. Of course the leaves had been thoroughly rinsed with sterilised water in order to remove a good many attacking spores. One can follow the infection under the microscope so that no confusion is possible. After some time the hyphae spread over the leaf; when I bound those so attacked round coffee branches on the tree, the infection soon gave positive results.

As the disease discribed by Zimmermann [1]) holds an isolated place, I will first deal with this one.

Zimmermann says it shows a very typical characteristic, the „anker cellen". *I am quite sure* that I never saw anything like them in this fungus, so the „spinnewebziekte" must be something else, though the growth on leaves and branches resembles in many respects the silverthread.

Zimmermann notices l. c. page 50, that this „spinnewebziekte" cannot be identical with the *Pellicularia Koleroga*, a disease described by Cooke, which attacks coffee in Ceylon. So I pass on to the next literature on this subject, namely the fungi mentioned by Cooke and by Ernst. I have not been able to peruse Cooke's original publication and so I had to be content with the information which Delacroix [2]) gives in his book, and with the description given by Massee in his „Diseases of cultivated plants and trees". A short account of the fungus is found in Kew Bulletin 1893, pag. 67.

Though the general description shows a rather close resemblance to the silverthread fungus, yet there are some important differences.

In the first place I never noticed conidia, which Cooke says occur in his fungus; the possibility remains that their absence was caused by unfavourable cultural conditions, but it is scarcely likely that the natural substratum should never show them.

Delacroix says in his description, pag. 69: „Quand

1) Zimmermann, l. c. pag 46.

2) Delacroix, Les maladies et les ennemis des Caféiers, Paris, 1900.

le temps est humide ce revêtement devient visqueux....:" Massee[1]) also speaks of „leaves covered with a slimy, gelatinous matter" and of „colourless hyphae, embedded in a mucilaginous matrix". These characteristics I never noticed. However damp the wether was, the powdery appearance never vanished. Then Delacroix mentions brown-coloured drops, which flow from the diseased leaves when the air is always damp; this I never saw in the silverthread.

Cooke says (Delacroix, pag. 69) that the berries may be attacked and fall in great numbers; I never saw, even in trees badly attacked, this in the case of our disease. So I conclude that *silverthread and Pellicularia are not identical.*

A third disease, named candelillo, which occurs in Venezuela is described by Ernst. There seems to be a detailed publication in the Spanish language; as the pages 1178—1179 in the Botanisch Centralblatt are nothing more than a „Notiz". He called the fungus *Erisyphe scandens,* but he adds that it is only a preliminary identification. Then Ernst says: „derselbe (der Pilz) scheint manche Aehnlichkeit mit *Pellicularia Koleroga* Ck. zu haben." Delacroix seems to have read the Spanish publication by Ernst; for he gives some particulars which are not to be found in the German article. According to Delacroix Ernst sent specimens to Cooke, which gave the following result: „Or, l'examen d'échantillons envoyés à Cooke par le Dr. Ernst lui même a permis au mycologue anglais d'affirmer l'identité des deux parasites dans le candelillo et le *Koleroga*". Notwithstanding this statement it seems to me, that one may be somewhat sceptical about this identity. The genus *Pellicularia* is a very in-

1) Massee, l. c., pag. 461.

completely defined one; I wish to draw the attention to a recently published article by Colemann [1]), who shows that the *Koleroga* disease on *Areca catechu*, according to Cook, identical with that found on coffee, is due to a *Phytophthora*. Moreover I received leaves attacked by candelillo from Venezuela. The fungus occurring on them, shows only a small resemblance to the *Pellicularia* as described in the publications already mentioned. Conidia could not be found, so that it was impossible to deny or affirm the identity with any certainty. However I made an accurate comparison between those leaves and the Surinam specimens.

In the first place the general appearance differs greatly. In candelillo the withered leaves have a dull grey velvety colour, which I never noticed in silverthread. This colour is caused by small hyphae, standing perpendicularly on the surface of the leaf, which show a very curious growth as may be seen in fig. 9*a*—*d*. The branchhyphae have at their apex almost globular cells, which form in their turn new globular cells, a growth more or less similar to that seen in Saccharomyces. I never saw it in silverthread; the aspect of silverthread under the microscope is shown in fig. 5, while in candelillo these typical short hyphae are seen, often united to large bodies of 50—100 μ diameter; one gets the impression of a greyish body with a crenate margin. Of course the possibility exists that this formation is caused by different climatic circumstances, though I do not think the climate would differ there so much from that of Surinam. I made several attempts to make cultures from the specimens sent to me; notwithstanding the leaves were quite dry, I succeeded in getting some fungus growth;

1) L. Colemann, Department of Agriculture, Mysore State, Mycological series, Bull. 2. 1911.

of course there are a good many spores from various fungi in the candelillo layer, so that it is rather difficult to say whether it is the candelillo which germinates. Several times I got the same fungus, but never a fungus identical with the silverthread.

I think I may be allowed from these investigations to declare that there is no identity between candelillo and silverthread.

In the Annual Report of the Porto-Rico Experiment Station for 1904 such a fungus is mentioned, which is investigated by Clinton. Dr. Clinton kindly sent me some dried speciemens from the Connecticut Experiment Station Herbarium, where he is now employed. These leaves showed a close resemblance to the diseased ones from Surinam trees; the aspect under the microscope of small magnifying power is quite the same. In the Porto-Rico climate, different from that in Surinam, the fungus shows exactly the same phenomena; I think this strengthens the conclusion that candelillo is something different from silverthread; if it were identical, there would be a great chance that the aspect and growth would be the same under the almost similar circumstances in Venezuela and Surinam.

It may be still mentioned, that in Kew Bulletin 1893, page 67, a note is found, that leaves, attacked by a similar disease were sent from Jamaica to Kew by Cockerell. Perhaps it is the same disease as that in Porto-Rico.

Method of control.

As the fungus grows almost entirely externally, the best method to combat it is to spray with a fungicide. I used 2% and 3% solutions of sulfate of copper; the stronger solution gave good results, but the foliage was

damaged by it. So it is better to use 2% Bordeaux mixture, which gave good results. When a small number of trees is badly attacked, it may be of great advantage to collect the withered leaves hanging from the branches and those lying under the tree, and to burn them.

Generally the damage caused by the disease is only of small importance, and spraying will not be necessary in the majority of cases; I should advise it for plots of trees, badly attacked, which seem to be a centre of infection in the midst of the cultivation.

The damage done in Surinam coffee is greater, the trees often lose so many leaves that they become almost bare.

Summary.

The silverthread disease attacks *Coffea arabica* as well as *C. liberica*.

It forms rhizomorphous strands, which grow acropetalous, and at the nodes of the branches, they spread over the under side of the leaves.

The hyphae clog the stomata; no sooner are the guard cells killed, then they also penetrate the leaf. Brown spots arise on the leaves; finally these wither completely and hang from the top of the branches by the mycel strands.

The fungus does not show any fructification; the mycelium has some typical characteristics.

Artificial inoculation gave positive results.

It is not identical with *Pellicularia Koleroga*, nor with candelillo, the same disease exists in Porto-Rico.

Spraying with Bordeaux Mixture is a good means of combating it.

LITERATURE.

A. ERNST, Botanische Notizen aus Caracas. Botanisches Centralblatt, 1880, 1_{er} Jahrgang.

A. ZIMMERMANN, Eenige pathologische en physiologische waarnemingen over koffie. Mededeelingen uit 's Lands Plantentuin 67, 1904.

G. DELACROIX, Les maladies et les ennemis des Caféiers. Paris, 1900.

G. MASSEE, Diseases of cultivated plants and trees. Kew Bulletin 1893.

APPENDIX.

After this pamphlet had gone to press I was able to study the contents of a newly published work on diseases of tropical plants by G. Delacroix. [1]) However the plants here mentioned do not warrant any change in my conclusions about the fungus, yet some peculiarities are sufficiently important, to be mentioned here.

Delacroix himself investigated leaves, which he received from the French West-Indian Islands, and which were attacked by a disease called „l'Enfer du Caféier". He reports that the disease has a great resemblance to *Koleroga*; however he was not able to observe some characteristics; for instance: conidia were not seen and the tissue was not gelatinous.

Gallaud [2]) saw a similar disease in New Caledonia, which appears to him to be same as *Koleroga*.

1) G. Delacroix, Maladies des Plantes cultivées dans les pays chauds. Paris, 1911.

2) I. Gallaud, Un nouvel ennemi des caféiers en Nouvelle-Calédonie, Comptes Rendus de l'Académie des Sciences, Nov. 1905, t. CXLI, pag. 898.

Gallaud gives as his opinion, that the leaves die because the hyphae attach themselves very closely to the cuticula of this leaf. Neither Cooke, nor Gallaud ever saw hyphae penetrating the leaf tissue.

EXPLANATION OF PLATES.

PLATE VII.

FIG. 1. Branch of Liberian coffee with a strand of the silverthread fungus.

In *a* the mycelium spreads over the leaf; the light-coloured spot on the side of the leaf near the basis is *already* dead.

FIG. 2. Result of artificial inoculation.

A myceliumstrand may be seen on the branch, the withered leaf hangs by this strand (*b*). The other leaf shows three brown spots.

PLATE VIII.

FIG. 3. Stoma just killed by the fungus. The striped part is dead. Magn. 400 ×.

FIG. 4. Stoma with a large number of cells already dead. Hyphae penetrating the leaf tissue. Magn. 400 ×.

FIG. 5. The membrane on the leaf surface. Somewhat simplified. Magn. 234 ×.

FIG. 6. Nodule of hyphae on a coffee branch in a pure culture. Magn. 234 ×.

FIG. 7. *a—j*. Typical myceliumforms. Magn. 234 ×.

a. to the left: anastomoses of hyphae; to the right: cells, resembling conidia.

b. short hyphae.

c. spirally twisted hyphae on agar.

d. and *f.* anastomoses in a hanging drop.

e. globular cells on agar.

g, *i*, *j*. hyphae with short bifurcated branches.

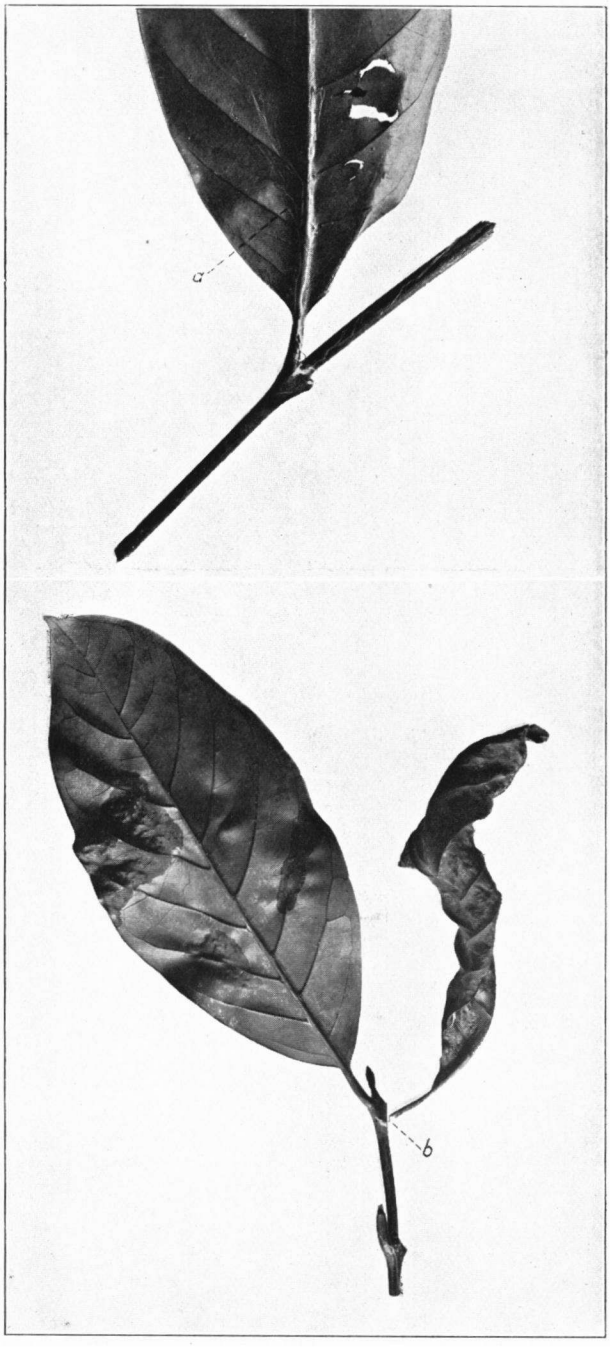
h. first sign of the formation of a nudule over a stoma. Magn. 400 ×.

FIG. 8*a* and *b*. Isolated cells germinating. Magn. 400 ×.

a. Old cell from a leaf.

b. Cell with much plasma from a pure culture.

FIG. 9 *a*—*d*. Hyphae typically branched, which cause the velvety appearance in candelillo. Magn. 400 ×.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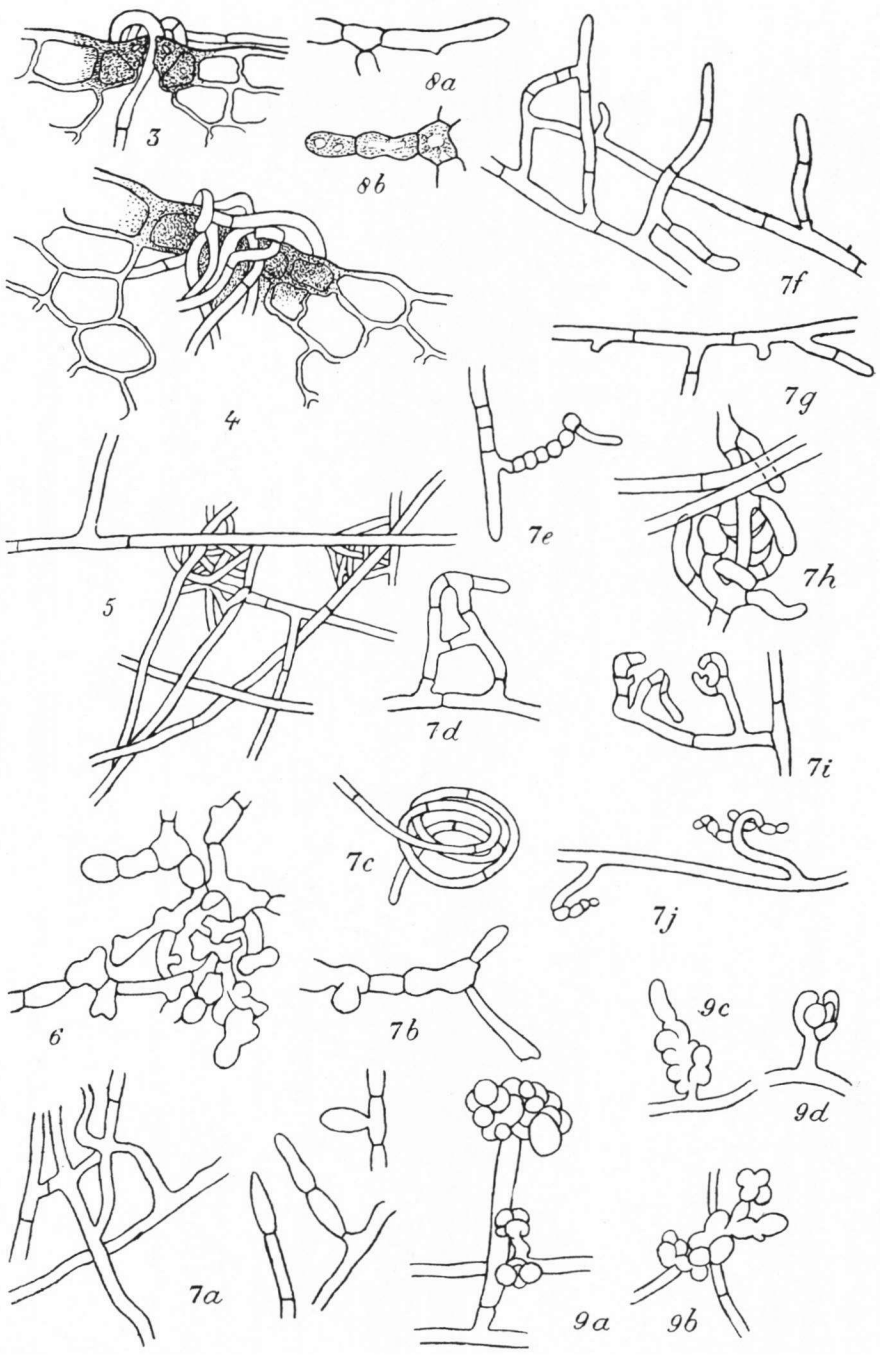
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