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THE "ROUGH-BARK" DISEASE ON APPLE BRANCHES AND LEAVES.

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B. T. GALLOWAY, *Chief of Bureau.*

THE "ROUGH-BARK" DISEASE OF THE YELLOW NEWTOWN APPLE.

BY

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LETTER OF TRANSMITTAL.

U. S. DEPARTMENT OF AGRICULTURE,
BUREAU OF PLANT INDUSTRY,
OFFICE OF THE CHIEF,
Washington, D. C., December 14, 1912.

SIR: I have the honor to transmit herewith a paper entitled "The 'Rough-Bark' Disease of the Yellow Newtown Apple," by Mr. John W. Roberts, Assistant Pathologist of this Bureau, and recommend its publication as Bulletin No. 280 of the Bureau of Plant Industry. This paper has been submitted by Mr. M. B. Waite, Pathologist in Charge of the Office of Fruit-Disease Investigations, with a view to its publication.

This manuscript contains an account of an apple disease the cause of which has hitherto been unknown, together with a description of the fungus causing it and methods for its prevention.

Respectfully,

B. T. GALLOWAY,
Chief of Bureau.

Hon. JAMES WILSON,
Secretary of Agriculture.

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THE "ROUGH-BARK" DISEASE OF THE YELLOW NEWTOWN APPLE.

INTRODUCTION.

For some years plant pathologists have been familiar with a disease of the Yellow Newtown apple tree which makes its appearance as a cankered or roughened condition of the bark on both trunk and branches.

In the summer of 1909 the attention of the writer was called to this disease, which is known to growers as "rough bark." So prevalent is this condition of the tree in certain sections of Virginia that many orchardists consider it a characteristic of the variety and use it as a means of identification. From this fact it will be apparent that the disease is not only widely distributed through Yellow Newtown orchards, but that it is sufficiently developed on individual trees to present a most striking appearance.

One may find trees which show the disease not only on the young 1-year-old wood, but also on the trunk and on all branches save those of the current year's growth. In the case of old trees which have not been properly cared for, the roughness and the cankered condition of the trunk and branches are particularly striking.

DESCRIPTION AND BEHAVIOR OF THE DISEASE.

The first appearance of disease is to be found in the sinking of certain definite areas of the bark, which later enlarge, turn black, and finally crack or break open about their margins. An examination of such spots shows that the underlying layer is blackened and dead. It is the shriveling and subsequent warping of these dead tissues, coupled with the ragged margins at the points where they break off from the healthy bark, which bring about the typical "rough-bark" effect. Plate I and Plate II, figure 1, showing the disease in its different stages, seem to justify orchardists in naming it "rough bark." The disease may continue to develop, sometimes involving an entire branch, or it may apparently die out and the wound heal over. In most cases, however, the blackened areas slowly enlarge, sometimes killing small branches by completely girdling them or leaving roughened or cankered places on the trunk or larger branches.

In the case of trees growing in poor soil or impoverished from other causes the disease may be quite destructive, entirely killing large limbs and adding to the pale, sickly appearance of the already unhealthy foliage. In most cases, however, only an occasional branch is killed and the tree lives on with its branches showing many roughened areas, its vitality doubtless somewhat weakened but without serious injury as long as it is in good growing condition. The chief complete destruction seems to be to the smaller branches, as the larger limbs and the trunk, though usually injured locally and badly roughened, are seldom deeply girdled.

So far as is known to the writer, the Yellow Newtown is the only apple variety susceptible to the disease. At least, it is the only variety growing in Virginia which is very seriously affected.

CAUSE OF THE DISEASE.

ISOLATION OF THE FUNGUS.

Cultures were made from diseased areas by Mr. W. M. Scott in 1907, and a fungus was isolated. In 1909 the same fungus was isolated by the writer from diseased spots which had only begun to develop. At various times since then the fungus, a species of *Phomopsis*, has been isolated from diseased branches and leaves of the Yellow Newtown and from the leaves of the York, Winesap, and other varieties of apples growing in Virginia.

INOCULATION EXPERIMENTS.

In the spring of 1910 inoculation experiments were made, using 1-year-old and 2-year-old wood of the Yellow Newtown apple. Spores from pure cultures of the fungus growing on corn meal were inserted beneath the bark through tiny slits. Only sterile instruments were used, and care was taken not to make ragged wounds. In order to close up the slits at once the branches thus inoculated were immediately bound round with raffia, which was removed one week later. Within a month there appeared about most of the points of inoculation darkened areas, which became larger and more nearly black as the season progressed. These places at the end of the season resembled very closely the dark, sunken areas which are typical of the initial stages of the disease. In nearly every case the inner bark was black and dead and the diseased area was quite definitely marked off from the surrounding sound tissues. An examination of these branches, which was made in July the following season, showed in and about 18 of the 24 points of inoculation the sunken, roughened or cankered areas which are typical of the "rough-bark" disease.

Plate II, figure 2, shows the disease as produced by artificial inoculations. All show the black, sunken areas which have begun to

crack and break off. The fungus had nearly girdled the specimen shown in the center of this figure. The control slits had healed over nicely and showed no roughening or adjoining dead areas.

The trees selected for these inoculations were healthy specimens near the center of a 4-year-old orchard that was in fairly good growing condition and which was some distance away from the nearest orchard in which the disease was prevalent.

Branches thus diseased through inoculation with spores of the fungus were brought into the laboratory, and from the margins of the spots or cankers the fungus was reisolated, usually in pure culture.

In like manner the trunks and branches of young trees growing in the greenhouse were inoculated. In two months well-developed sunken spots possessing all the characteristics of the disease appeared. The fungus was reisolated both by plating the spores from the few pycnidia which had formed and through cultures from the diseased tissues.

Attempts were made to bring about infection by simply spraying spores on branches and on green twigs and water sprouts of the current year's growth, but in no case did the fungus gain an entrance. An abrasion or wound seems necessary before infection can take place.

Numerous inoculations made in orchards during the fall of the year have been uniformly unsuccessful. It would seem, therefore, from these experiments and from the observations as to the size of spots due to natural infection that the earlier part of the season is the time during which the fungus gains entrance and begins its development.

Since this same species of *Phomopsis* had also been isolated from leaf spots along with *Sphaeropsis malorum* even as early in the season as April, it was thought desirable to test its parasitism on leaves.

When spores were sprayed on healthy leaves, either in the orchard or in the greenhouse, no spotting resulted under ordinary conditions; but when a seedling apple tree growing in the greenhouse was thus sprayed and covered with a bell jar within which the air was kept saturated with moisture, an occasional leaf showed a few minute spots, from which the fungus was reisolated. However, the conditions and not the fungus may easily have initiated the spots, after which the fungus might readily have entered. Apple leaves, when moistened and covered with a bell jar, are likely to be spotted, especially if exposed to strong sunlight. That the fungus will grow on such dead spots is shown by the fact that when, in either orchard or greenhouse, spores were sprayed on leaves which had previously been burned slightly with a hot needle, the fungus infested the dead areas and developed pycnidia on them within a week. The leaves

used in the orchard experiment were covered with a paper sack for 24 hours to prevent excessive drying out, while those used in the greenhouse were covered with a bell jar for the same length of time. The spots did not seem to enlarge much, if any, but became gray with reddish margins. As shown in Plate I and in Plate III, figure 1, such spots with numerous pycnidia are strongly suggestive of *Phyllosticta*, and indeed the fungus under such conditions might easily be mistaken for a *Phyllosticta* or a *Phoma*. When plates were poured and the fungus reisolated it fruited as a *Phomopsis*, having both *Phoma* spores and the long, slender, curved sort which Shear¹ calls *scolecospores*.

The fungus will also cause a rot of the apple. York Imperial apples, about one-third grown, very hard and green, were inoculated through needle pricks with spores from corn-meal cultures and then placed in moist chambers. In one month rotted spots from 0.5 to 1 centimeter in diameter were noted about most of the points of inoculation, and on the larger spots the pycnidia of a fungus were quite conspicuous. (See Pl. III, fig. 3.) A microscopic examination of pycnidia and spores showed them to be identical with those of the fungus concerned in the inoculations. The pycnidia contained both the *Phomalike* and the long, slender, hooked spores, the latter being particularly abundant. Cultures from the margins of the rotted areas produced the fungus in pure culture, showing conclusively that it had been the cause of the decay.

Full-grown but solid, not fully ripe, specimens of Yellow Newtown apples were inoculated by inserting spores through breaks in the skin and then placed in moist chambers. In two weeks rotted areas 4 to 5 centimeters in diameter appeared about the points of inoculation, while the controls showed no decay. From the margins of these rotted spots also the fungus was reisolated in pure culture. Typical pycnidia and spores were also formed, as in the case of the green apples. It will thus be seen that the fungus, especially on ripe Yellow Newtowns, is able to produce considerable rotting, though such a rot apparently occurs seldom in nature.

DESCRIPTION OF THE FUNGUS.

On corn meal, corn-meal agar, and sterile apple wood the fungus fruits quite readily, but pycnidia formed naturally on apple branches are difficult to find, being rather obscure and apt to disappear rather quickly after spore discharge has taken place.

The globose pycnidia develop singly or sometimes in a raised stroma, which may be from 0.5 to 1 centimeter across. They are

¹ Shear, C. L. The ascogenous form of the fungus causing dead-arm of the grape. *Phytopathology*, v. 1, p. 116-119, 5 fig., 1911.

quite variable in size, those which develop in corn-meal cultures being rather large, while those on leaves are comparatively small. Average-sized pycnidia have a diameter of about 0.5 millimeter, but the range of size runs from 0.1 to 1 millimeter in diameter.

A cross section of a pycnidium (fig. 1) shows a dark outer layer of large-celled pseudoparenchyma; inside this is a thick, lighter colored layer of rather loose texture, making up the bulk of the pycnidial wall; next is a densely woven olivaceous or brownish layer, and within is a closely woven hyaline layer. The interior of the pycnidium is usually more or less plurilocular through the upward growth or invagination of the base or by an inward protrusion from the side.

FIG. 1.—Section through a pycnidium of *Phomopsis mali* grown on corn meal.

The spore masses when mature are extruded either in an indefinite cream-colored or yellowish mass or in slender ropelike masses from 1 to 3 centimeters long. (Pl. III, fig. 2.)

The spores are of two kinds: Those called by Diedicke¹ α spores are spindle shaped or nearly so, measure 7 to 10 by 3 to 4 μ , mostly 8 by 3 μ , contain two oil drops, and are borne on cylindrical basidia measuring about 20 by 2.5 μ (fig. 2). They germinate readily in distilled water.

FIG. 2.—The two forms of spores of *Phomopsis mali* produced in one pycnidium.

Those which Diedicke designated as β spores and called by Shear scoleospores are threadlike, curved at the center, hooked at one end, sigmoid or nearly straight, somewhat attenuated and borne on short, conical basidia. They vary considerably in size, the

average being 28 by 1.5 μ . Repeated attempts have been made to germinate these spores, but without success.

A pycnidium may produce spores all of one kind, but usually both kinds are found in the same pycnidium, though there is often a greater quantity of the one than of the other. Pycnidia developed on leaves show less tendency to form the long, curved sort than

¹ Diedicke, H. Die Gattung *Phomopsis*. Annales Mycologici, v. 9, p. 8-35, pl. 1-3, 1911.

those growing on either fruit or branches; in fact, this form of spore is rather difficult to find in pycnidia from leaves. For this reason the fungus as it appears on leaves may easily be mistaken for *Phoma mali* Schulz. and Sacc. On apple wood and fruit pycnidia produce both spore forms in abundance, some of them producing only one form, while others produce both. A localization of the production of the two kinds of spores in separate portions of the same pycnidium, as mentioned by Diedicke, has never been observed.

The fungus seems to differ essentially from other fungi described as occurring on the apple. *Phoma mali* Schulz. and Sacc., shown by Lewis¹ to cause a canker and soft rot of the apple, resembles it in the size of spores and growth on certain media, but differs from it in the absence of secondary spores, in the structure of the pycnidium, and in having less tendency toward stroma formation. The reactions of the two fungi toward certain culture media are also very different.

Fusicoccum viticolum, described by Reddick² and shown by him to cause a disease of the grape, resembles this fungus in the production of two similar spore forms, but differs from it in the size of spores and in that the pycnidia consistently develop in a stroma.

Phomopsis citri, described by Fawcett³ as the cause of a stem-end rot of citrus fruits, differs materially from this fungus both in pycnidial and spore characters, as does also the pycnidial form of *Diaporthe batatatis*, as set forth by Harter and Field,⁴ though both of these are evidently closely related to this fungus.

Accepting Saccardo's conception of the genus *Phomopsis* and Diedicke's investigations of the genus, the fungus in question is clearly a *Phomopsis*.

Of the described species of this genus the fungus resembles most closely *Phomopsis ambigua* (Nits.) Trav., described as occurring on the pear in Europe, but differs from it in having much larger secondary spores and is apparently different in other characters, though it is difficult to tell from the meager published description. Furthermore, *Phomopsis ambigua* is described as the imperfect form of *Diaporthe ambigua* Nits., associated with it on the pear, but no species of *Diaporthe* has ever been found in connection with this fungus.

¹ Lewis, C. E. Apple diseases caused by *Coryneum follicotum* and *Phoma mali*. Maine Agricultural Experiment Station, Bulletin 170, 1909.

² Reddick, Donald. Necrosis of the grapevine. New York (Cornell) Agricultural Experiment Station, Bulletin 263, 1909.

³ Fawcett, H. S. The cause of stem-end rot of citrus fruits (*Phomopsis citri* n. sp.). *Phytopathology*, v. 2, p. 110-113, pl. 8-9, 1912.

⁴ Harter, L. L., and Field, E. C. *Diaporthe*, the ascogenous form of sweet potato dry rot. *Phytopathology*, v. 2, p. 121-124, 4 fig., 1912.

It has therefore seemed best to the writer to describe it as a new species under the name of *Phomopsis mali*, as follows:¹

Phomopsis mali sp. nov.—Pycnidiis subglobosis, sparsis, gregariis vel in stromate, nigris, carbonaceis, plurilocellatis, ostiolatis. Sporulis subfusoides biguttulatis, continuis, hyalinis, $7-10 \times 3-4 \mu$. Basidiis subulatis $20 \times 2.5 \mu$. Sporulis ordinis secundi filiformibus, uncinatis vel sigmoideis, attenuatis, $20-36 \times 1.5 \mu$.

Hab. in ramis truncisque et foliis Piri mali in Virginia, America boreali.

[Pycnidia subglobose; scattered aggregate or in a stroma, black, carbonaceous, plurilocular, ostiolate. Spores subfusoid, containing two oil drops, continuous, hyaline, measuring 7 to 10×3 to 4μ . Basidia awlshaped, measuring $20 \times 2.5 \mu$. Secondary spores threadlike, hooked, or S-shaped, attenuate, measuring 20 to $36 \times 1.5 \mu$.]

The vegetative portion of the fungus grows in the living part of the bark next to the wood, only its fruiting bodies appearing as minute blackish elevations, which break through the bark in diseased areas, giving them a pimplelike appearance. In these are contained the spores or reproductive bodies, which on being carried to wounds or small abrasions are under favorable conditions capable of germinating and producing the disease.

Many attempts have been made to find an ascogenous form of this species, but so far without success. As most of the known species of the genus have proved to be imperfect forms of *Diaporthe*, the writer has every reason to expect that eventually a *Diaporthe* will be found to be the ascogenous form of this fungus.

GROWTH ON CULTURE MEDIA.

On beef-agar slants.—White cottony, with numerous flocculi, which later turn black, becoming pycnidia, containing spores only in rare cases. On this medium the pycnidia do not occur in a stroma.

In corn-meal flasks.—Surface at first is covered with cobwebby mycelium, with numerous irregular raised places brownish in color. From these stromalike places spores are extruded in long, ropelike masses after one month's time. Pycnidia are also produced singly.

On corn-meal agar slants.—Mycelium forms a delicate film over surface of the medium. After 45 days long ropelike masses of spores are produced from pycnidia, which are usually separate but are sometimes in a stroma.

On potato cylinders.—Growth at first consisting of fine, flocculent, pure-white mycelium, which later becomes dark. Finally pycnidia are formed, which produce spores in slender, threadlike masses. Pycnidia are usually separate but may be in a stroma.

On prune-agar plates.—Mycelial growth cottony; pycnidia often formed in a stroma, as on corn meal. Spore masses appear after 20 days.

¹ Roberts, J. W. A new fungus on the apple. *Phytopathology*, v. 2., no. 6, p. 263-264, Dec., 1912.

On oxalic-acid agar slants.—After 12 days the surface is covered with powderylike growth, white with a tendency toward brown in some places. Neither pycnidia nor spores are formed on this medium.

On sterile apple wood.—Growth white and scant. Pycnidia and spores are formed abundantly in 30 days.

On synthetic agar slants.—After 15 days the medium is covered with a luxuriant growth of downlike mycelium, which later becomes dark at the surface of the slant.

On beef bouillon.—Delicate white hyphæ permeate the liquid in five days. Later the surface is covered with a crust of hyphæ having a white powdery surface. Pycnidiumlike bodies are produced on this crust next to the wall of the tube, but no spores are to be found.

PREVENTIVE MEASURES.

While definite spraying experiments for the control of this disease have never been carried on, observations in orchards which have been well sprayed both in growing and dormant seasons tend to show that such orchards may be as subject to the disease as those in which there has been little or no spraying, other conditions being the same. One of the orchards showing the disease the worst of any which the writer has seen had for several years past been sprayed in the winter with a strong solution of lime sulphur for scale, with dilute lime-sulphur solution once or twice in the early spring, and has besides received three or four applications of Bordeaux mixture during July and August. It is possible, of course, that spraying with bluestone or strong Bordeaux mixture before the buds open in the spring, with subsequent applications of weaker Bordeaux mixture during the early part of the growing season, might serve to check the disease, but it hardly seems probable. From the fact that fall inoculations have not been successful, spraying at that time of the year or in early winter for the control of this disease would seem of little value. When it is considered that the spores are protected by their pycnidial coverings under ordinary circumstances, oozing out only under moist conditions, and that the disease is probably initiated through the germination of spores which have lodged in small abrasions, hail marks, insect punctures, etc., the probability of successful spraying for its control becomes very remote. Spraying, therefore, probably is of value only in so far as in helping to maintain the general health of the tree it gives an increased resistance to disease.

From a commercial standpoint, careful pruning, proper cultivation, and, when necessary, fertilization seem quite successful as control measures, for as long as the trees are in good growing condition the disease will not be very serious.

SUMMARY.

The “rough-bark” disease of the Yellow Newtown apple in Virginia is caused by a new species of fungus, to which the writer has given the name *Phomopsis mali*.

As trees which are in good growing condition are usually exempt from attacks of the fungus, fertilization and cultivation, together with proper pruning, should act as preventives, and as checks where the disease has already gained a start. Spraying is apparently effective only in so far as it contributes to the general health of the tree.

The fungus will grow on leaves in spots previously diseased, often following *Sphaeropsis malorum* very closely in early spring, but is probably not a serious leaf disease.

A rot of the apple may be produced by this organism, but is probably of rare occurrence in nature.

DESCRIPTION OF PLATES.

- PLATE I. (*Frontispiece*.) The "rough-bark" disease on apple branches and leaves. Fig. 1.—A branch of a Yellow Newtown tree, showing the earlier stages of the disease. Fig. 2.—A Yellow Newtown apple branch which shows the disease in its later stages. Fig. 3.—Apple leaves showing pycnidia of the fungus on areas which were first burned with a hot needle and then sprayed with spores.
- PLATE II. Fig. 1.—Branches of Yellow Newtown apple trees, showing various stages of the "rough-bark" disease. Fig. 2.—Yellow Newtown apple twigs, showing the "rough-bark" disease produced by artificial inoculation with spores of *Phomopsis mali*. Both the incipient sunken stage and the later roughened stage are shown.
- PLATE III. Fig. 1.—York Imperial apple leaves inoculated with *Phomopsis mali* through areas which had previously been burned with a hot needle. The fungus is fruiting on the spots. Fig. 2.—Growth of *Phomopsis mali* on corn meal, showing ropelike masses of spores after 60 days. Fig. 3.—York Imperial apple, about one-third grown, showing rot produced by artificial inoculation with the fungus, which is fruiting on the decayed spots. Note the ring of pycnidia on the largest spot.

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FIG. 1.—BRANCHES OF YELLOW NEWTOWN APPLE TREES, SHOWING VARIOUS STAGES OF THE "ROUGH-BARK" DISEASE.

FIG. 2.—YELLOW NEWTOWN APPLE TWIGS, SHOWING THE "ROUGH-BARK" DISEASE PRODUCED BY ARTIFICIAL INOCULATION WITH SPORES OF *PHOMOPSIS MALI*.

FIG. 1.—YORK IMPERIAL APPLE LEAVES INOCULATED WITH PHOMOPSIS MALI THROUGH SCORCHED AREAS.

FIG. 2.—GROWTH OF PHOMOPSIS MALI ON CORN MEAL, SHOWING ROPELIKE MASSES OF SPORES.

FIG. 3.—YORK IMPERIAL APPLE, SHOWING ROT PRODUCED BY ARTIFICIAL INOCULATION.