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THREE FUNGOUS DISEASES OF THE CULTIVATED GINSENG.[1]

By Howard S. Reed, Assistant Botanist.

The ginseng plant (**Aralia quinquefolia. D. & P.**) has been cultivated in Missouri for upwards of ten years. Within the past four years the plantations have so increased in size and number that in certain districts ginseng constitutes an important crop. The plant is cultivated for the large, fleshy root, which, when dried, forms an article of commerce. In 1904 the dried roots sold at an average price of ten dollars a pound. Practically all of the crop is exported to China and, up to the present, the demand has exceeded the supply.

From statistics furnished by the growers in 1904, there were about twelve acres of ginseng gardens in the state, representing a value of $150,000. In the same year there was produced approximately 250 pounds of seed, worth according to the growers $12,500, and about 400,000 seedling plants, worth $30,000. Most of the plantations were too young to yield many dried roots, consequently the income from that source was small. The growers estimated that there were 250 pounds of dried root which had a market value of $2,500.

I. METHOD OF CULTIVATING GINSENG.

Since the method of cultivation has a direct bearing upon the problem to be discussed later, it will be necessary to outline briefly the practice in vogue in this state.

[1] The author wishes to express his obligation to the members of the Missouri Ginseng Growers' Association for the very kind way in which they supplied him with information and material for his studies. He also wishes to thank Mr. L. F. Childers, a senior student in the College of Agriculture, for very liberal assistance in starting the pure cultures of the various fungi.

The ginseng plant is a native of deep, shady woods, and is indigenous to the United States and Canada, being found, in its wild state, as far west as Eastern Kansas. The root is perennial, the top is annual. The root is not unlike the carrot in form, and when five or six years old often weighs four or six ounces. Cultivation increases the size and weight. The wild root weighs from half an ounce to two ounces. Since the wild root has almost become extinct through the vigorous digging of the "Sang hunters," it has been found that the plant can be cultivated with great success, and hundreds are turning their attention to it.

The growers strive to duplicate the conditions of the forest, in so far as is possible. They often construct the beds entirely, or in part, of leaf-mould taken from the forest, and mulch the ground around the plants each autumn with forest leaves.

The plants are shaded artificially by erecting arbors over the beds. (Figs. 2 and 3.) In many cases the arbors consist of scaffolding six or eight feet above the ground, upon which is spread a layer of leafy boughs, until no direct light reaches the ground below. The sides of the arbor are covered with lattice work to protect the plants growing near the edge of the beds. Recently, lattices of lath have been substituted for the brush arbors and have proven more satisfactory.

After the plants are put into the ground and the mulch of leaves put on, the surface of the soil is not stirred. The ground is enriched by applications of wood-ashes, lime, or bone-meal, and these applications are frequently heavy. Each autumn, as stated above, the ground is mulched with forest-leaves.

II. APPEARANCE OF THE DISEASES.

The first notice of the occurrence of disease in the ginseng plantations was received in the early summer of 1904. At that time the cause of the disease seemed to be a leafspot fungus, and it was thought that spraying with Bordeaux mixture might keep it in check. In August of the same year word was received from the secretary of the Missouri Ginseng Growers' Association that the disease was daily becoming more serious. It was estimated that the damage to the seed crop at Houston amounted to $50,000.

The aforesaid Association asked that one of the staff be sent from the Experiment Station to investigate conditions, and, if possible, to give the ginseng growers some aid in combating the disease in their arbors. Accordingly, the writer was sent and visited the ginseng district in Texas county.

Through the courtesy of the growers, it was possible to obtain very full data of the diseases and to bring back plenty of suitable material for study. The following pages contain the results of a study of the causes of the diseases and methods of holding these diseases in check.

The summer of 1904 was marked by a very abundant rainfall. The dense shade of the arbors kept the soil beneath them moist, if not wet, for several weeks at a time. This moist soil, rich in humus and other organic substances, formed an exceedingly favorable place for the growth of fungi. Gardens under dense shade, with poor ventilation and poor drainage, suffered the greatest loss. All ages of plants were attacked and seemed to suffer alike, if the conditions were favorable for the growth of fungi.

The diseases found were caused by entirely different fungi from those producing diseases in the ginseng gardens of New York;[1] moreover, they had never been known to attack ginseng plants before. A somewhat detailed account is therefore given of the studies which the writer has made, in order that the characters of the disease-producing fungi may be known as fully as possible.

[1]See Bull. 219 Cornell Agr. Exp. Sta. 1904.

III. CAUSES OF THE DISEASE AND DEATH OF THE GINSENG PLANTS.

The investigations carried out have shown that each of the three diseases is caused by a fungus which lives in the tissues of the ginseng plants. Their presence in these tissues brings about conditions which cause the disease or death of the plant. Before their host-plant dies, however, they usually succeed in producing immense numbers of spores, or seed-like bodies, which may be carried to other plants by the wind or rain. If the soil and plants are continually moist and are protected from the direct rays of the sun, these spores will germinate very quickly and infect the new plants with the same disease. It is needless to repeat that the ginseng arbors furnish very favorable conditions for the germination and growth of fungi during rainy weather. This fact will no doubt explain the widespread belief that the diseases were in some way caused by the wet season.

IV. THE DISEASES AND THEIR EFFECTS UPON THE GINSENG PLANTS.

Stem Anthracnose (Vermicularia Dematium).—Between the first and fifteenth of May (i. e., about thirty days after the plants had appeared above the ground), an anthracnose appeared which produced numerous black scars on the stems of the plants. These scars gradually spread, sometimes encircling the stems. The first indication of injury was seen when one leaflet after another turned brown; from them the disease spread down the petiole to the main stalk. Other stalks were attacked so badly that they broke off and fell over before the upper portions had even become wilted. Mr. Millard remarks that in some cases the severed stalks remained green for some days after connection with the root had been broken. This, however, may have been due to the exceedingly moist atmosphere under the arbor. After the loss of the top from this anthracnose, the "crown" of the root was liable to be attacked by fungi or bacteria, causing decay. Although many plants were attacked by this anthracnose, only a small portion were directly killed by it. The greatest loss caused by this disease lies in the destruction of the seed crop.

This disease, which we may call Stem-anthracnose, is caused by a fungus belonging to the genus Vermicularia. It occurs quite commonly upon a number of our native plants. Among others which it infests may be mentioned the Indian turnip, the pokeroot, the honeysuckle, the maple, and the willow. Microscopical examination shows that the long, slender filaments which make up the vegetative body of the fungus, infest those tissues of the plant lying just beneath the epidermis. They live upon the nourishment which those tissues contain, and, after attaining to a certain stage of development, come to the surface of the stem and produce the black spots previously mentioned. These black spots are the fruiting bodies of the fungus; when mature they discharge a large number of spores. (See Fig. 4. A, B, and C.)

In some plants the growth of the fungus encircled the stems and, by killing a ring of cells, caused the stems to break off and fall over.

The direct injury which this fungus brings about is due to the presence of its filamentous threads in the cells of the cortical and sclerenchyma layers. It lives on the contents of these cells, and eventually disintegrates them. About the time this is accomplished, the fungus breaks through the epidermis and forms the black spore-producing bodies shown in Fig 4, A, B, and C.

Treatment. Fortunately, this disease can be effectually checked by the use of Bordeaux spraying mixture. The plants should be sprayed about three weeks after they appear in the spring, and every three weeks thereafter until

August 1st. If the season should be very wet, it might be necessary to spray once in two weeks instead of three.

Inasmuch as many ripe spores are left in the scars on the ginseng stalks in the fall, and as these spores may infect the arbor the next spring, all dead stalks and leaves should be raked up and burned in the fall before the winter mulch is applied. Since this disease attacks many of our native plants, one should not use forest leaves as a mulch; because in so doing, he will run great risk of introducing the spores of the disease.

Leaf Anthracnose (Pestalozzia funerea).—About the first week in July, the plants were attacked by another "stem-spot," or anthracnose disease. It appeared as a black, velvety growth at the bases of the leaves and flower stalks, causing the leaves and seed stalks to die and fall off. This disease was extremely destructive to young plants, but did not seriously damage the older ones.

This disease, which may be called "Leaf-anthracnose," was due to a fungus belonging to the genus Pestalozzia. The life-history of the fungus was not investigated as thoroughly as that of the others, but there is little doubt that it affects the ginseng plant in practically the same manner as the Vermicularia fungus.

This Leaf-anthracnose caused the death of thousands of young plants, because it girdled the leaf stalks and cut them off at the very time when they were most needed.

Treatment. It has been shown that this fungus may also be kept in check by spraying with Bordeaux mixture.

The Wilt Disease (Neocosmospora vasinfecta).—The wilt disease, which made its appearance about the first of July, was by far the most dangerous one found in the ginseng gardens; and it was this one which caused such wholesale destruction. Whole plantations were often destroyed in a single week. Mr. H. S. Millard says that neither the Bordeaux spraying mixture nor lime-dust seemed to have any effect in checking its ravages.

The accompanying illustration (Fig. 1.) shows the progressive stages of the wilt.

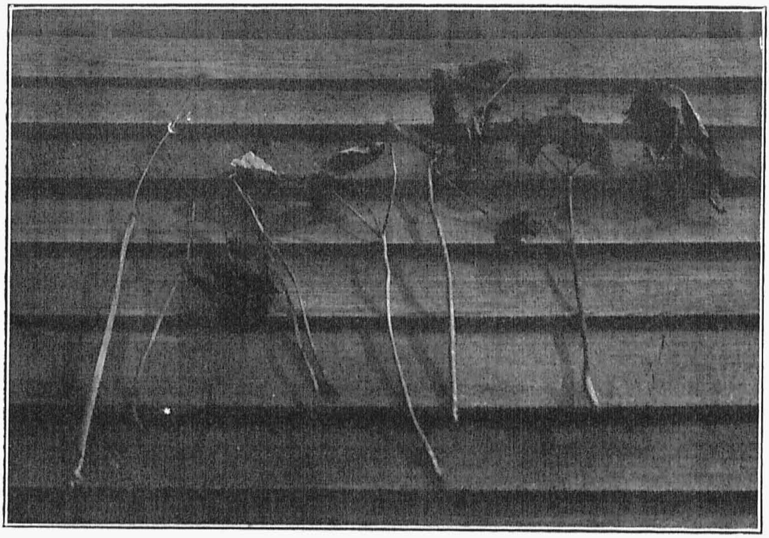

Fig. 1. A photograph showing the progressive stages of the Wilt-Disease.

The leaflets first droop as if the plant were suffering from lack of water; one after another they turn yellow and die. Gradually the entire leaf dies back to the base, dries up, and falls off, as shown by the plants in the left of the illustration. In the meantime the flower and flower stalk are also affected. Here the disease first manifests itself in the immature seeds; they shrivel up, die, and fall to the ground before reaching maturity.

By the first of August the entire stem is dead back to the surface of the ground, but remains erect for some time afterwards.

The roots of the plants are not directly killed by the wilt fungus, but, naturally, the death of the top must put an end to the growth of the root for that season. Another result is, that the buds formed on the "crowns" of the roots and from which the next season's tops are produced, are dwarfed. There were some cases of soft rot in the roots, which were also due indirectly to the wilt disease. When the tops died they allowed the organisms of decay to enter at the base of the stems and to work their way downward into the roots, producing a soft rot.

It is a remarkable fact that the wilt disease never appeared alone, but was always found in plants which were more or less affected with anthracnose.

The Wilt-fungus lives from one season to the next in the soil. It has been known to exist there for a number of years without losing its vitality. In the heavily mulched ginseng gardens it would find conditions extremely favorable for living through the winter.

The fungus is identical with the one which causes the destructive wilt of cotton, watermelon, and cowpeas.

The writer was at first inclined to believe that the Wilt-fungus gained entrance through the roots of the ginseng plant, but more careful examination of the roots failed to show any signs of fungus infection. There is evidence, however, for believing that the fungus enters the ginseng plant at the base of the stem through the lesions, or open scars, caused by the stem-anthracnose. The facts which support such a belief are as follows: (1) Careful microscopical examination fails to show the presence of any fungus in the young parts of the roots; (2) in gardens where the anthracnose fungus was held in check by spraying, there was no Wilt-fungus; (3) the Wilt-disease did not appear until two months after the Stem-anthracnose had made its appearance.

The Wilt-fungus possibly enters the roots in some cases through the scar left at the place where the stem of the previous year broke off.

Once inside of the ginseng plant, the fungus spreads through the vascular bundles. The vascular bundles are a system of ducts which runs through the entire plant. They are woody, thick-walled cells, whose function it is to convey water upward from the roots to the leaves. The filaments of the Wilt-fungus fill these ducts with a compact mass of vegetative filaments and stop the ascending current of water. This explains why the plants wilt and die, although growing in a very moist soil. Figure 6 shows the appearance of one of the vascular bundles infested with fungus filaments, as seen under the microscope.

The fungus is not found outside of the vascular bundles while the plant continues alive. It lives entirely within the host plant, except for patches of spores which are produced on the surface of the older stems. It is very common to find these clusters of spores in the interior of dead, hollow stems.

TREATMENT.

It will be seen from this brief description of the fungus that it is an exceedingly difficult disease to combat. Living from year to year in the soil, it enters the plants through the lesions left by other fungi and spreads upward through the water-conducting channels. It does not once appear upon the surface until the plant is beyond recovery. Obviously, we cannot directly apply any substance to kill the fungus without first killing the plant it infests. It can be successfully checked, however, by indirect means.

Extensive experiments have shown beyond all question that the Wilt-fungus cannot be killed by the application of fungicides to the soil.[2]

Fortunately for the ginseng grower, there is an indirect method of attacking the Wilt-disease which is eminently successful. The entire history of the Wilt-fungus indicates that it is weak in its ability to attack healthy ginseng plants and infect them. It follows in the path of other diseases and attacks plants which are already somewhat

[2]See Bull. 27. U. S. Dept. Agr., Div. of Vegetable Physiol. and Pathology. 1900.

weakened.[3] So constant is the association of the Stem-anthracnose and the Wilt, that the men who cultivate ginseng regard the former as an early stage of the latter disease.

These facts point to but one conclusion; viz., **the Wilt-disease can be controlled by keeping the plants free from anthracnoses and other fungi which produce open lesions on the plant.**

That this conclusion is a correct one is shown by the following instance. At Houston, two of the largest gardens are those owned by the Millard Ginseng Company and Mr. R. E. Barnard. Their gardens are only a few rods apart on the Piney River and have exactly the same conditions except that the Millard gardens were thoroughly sub-drained and the plants were sprayed with Bordeaux mixture. In 1904, Mr. Barnard's plants were completely killed down by the Wilt-disease, while not a single plant was lost from that disease in the Millard gardens.

V. GENERAL REMEDIES.

The most effective of all remedies for these fungous diseases which have been tried is Bordeaux mixture, sprayed on the plants once in three weeks. The applications should begin as soon as the leaves of the ginseng plant have unfolded and continue until August first. If the season should be excessively rainy, it might be necessary to apply the spray once every two weeks.

If the spraying is not done early enough to prevent the anthracnoses it will be lost, since no amount of spraying will prevent the growth of the wilt-fungus when it has once entered a plant.

In addition to the practice of spraying, the growers of ginseng should take all possible preventive measures, for even the best of remedies sometimes fail. A few suggestions are herewith given in the hope that they may be found useful.

[3]See Bull. 41. Alabama Agr. Exp. Sta. 1892. Also, Bull. 79. Massachusetts Exp. Sta. 1902.

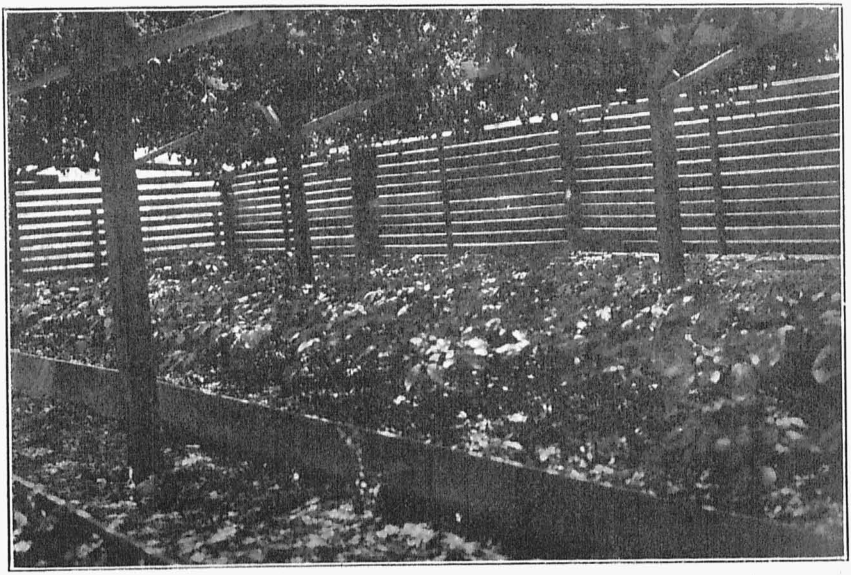

Fig. 2. A raised bed in the garden of D. L. Max. Note the absence of diseased plants.

The soils employed in making the beds should not be taken from the forest, as it is likely to contain spores of the fungus causing the wilt, because, as I have found, this fungus also infects some of our native plants. Soil may be made by mixing rich river silt and sand with well rotted stable manure, or by enriching sandy or gravelly soil. The beds may be fertilized with stable manure or with bone-meal, wood ashes, air-slaked lime, or muriate of potash. It should always be borne in mind that anything which adds to the vigor or thrift of the plant enables it to withstand better the attacks of parasitic fungi.

Gardens should be small and located some little distance apart, then if one becomes infected with the disease, it can be taken up before the disease infests a large territory.

Proper drainage is very necessary for a successful ginseng garden. It is advisable to locate the garden on a gentle slope, if possible. In all cases the ground should be well under-drained by tile or stone drains. The drainage should be further aided by making raised beds.

The condition of the gardens at Houston at the time of my visit in August shows how important is this matter of drainage. The garden of Mr. Sandidge, which showed almost no signs of the disease, was provided with ditches one foot deep, filled with rock, between the beds. The garden of Mr. D. L. Max afforded an interesting bit of evidence in favor of raised beds. He had four raised beds, in which there was very little disease, but adjoining them were a number of flat beds which were badly infested with the disease. (See Figs. 2 and 3.)

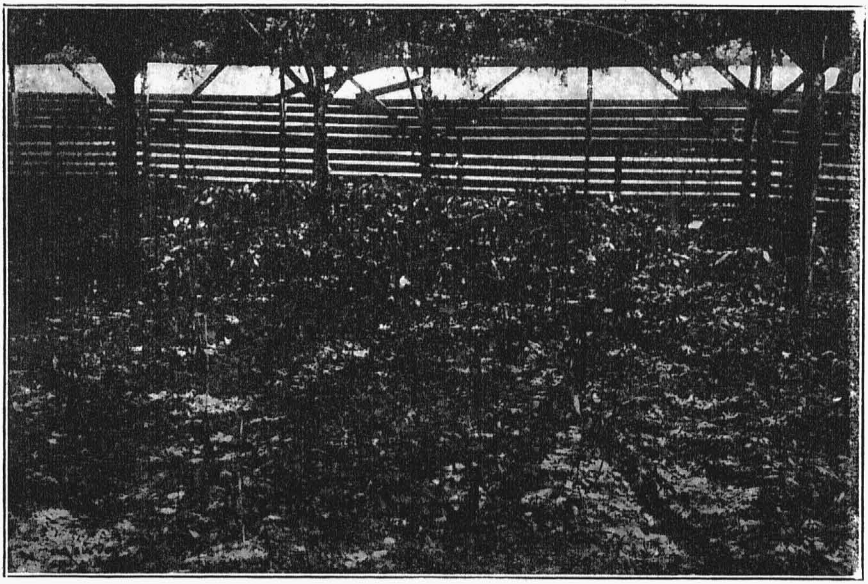

Fig. 3. A flat bed in the same arbor as that shown in Fig. 2. Note the large number of plants killed by the Wilt-disease.

Ample ventilation must also be provided in building the arbor. Many of the arbors are enclosed at the sides too tightly, thus hindering a free circulation of air over the beds and favoring the growth of fungi. The plan of construction employed by Mr. Sandidge, of Houston, seems to me to be nearly ideal. He has a strip of wire-netting three or four feet wide nailed to the posts next to the ground, and boards or lattice-work from that upwards. This style of wall prevents the hot sunlight from striking the plants and at the same time allows a good circulation of air over the beds.

Seeds from diseased plants may be used in planting, but it is safer to sterilize them first. For this purpose one may dissolve one ounce of corrosive sublimate in seven and one-half gallons of water, and place the seeds in it for fifteen to twenty-five minutes. Another method is to dissolve one ounce of potassium bichromate in two and one-half gallons of water, and place the seeds in it for three hours.

Gardens may be contaminated with the disease from the tools used, therefore one should clean the tools of all adhering soil and wash them occasionally with one per cent formalin or one per cent carbolic acid.

Diseased plants should be removed and burned as soon as they are affected. A prompt destruction of one diseased plant may save a number of healthy plants from becoming infected.

The material used for mulching should be of a sort which will not contaminate the garden with disease. It is safer not to use forest leaves, but rather to use clean straw or marsh hay. Some fungi will be killed if the ground is allowed to freeze before putting on the mulch.

Another, and perhaps the most promising, mode of procedure lies in propagating a variety of ginseng which will be resistant to the wilt disease. In every garden, no matter how badly diseased, there are certain plants which live through the attacks of the disease and ripen seeds. These seeds should be saved and planted separately, the hardiest of their offspring should be used to propagate seeds for future planting. By thus selecting the hardiest individuals year after year, it will be possible, in time, to originate a variety which will be quite resistant to the attacks of parasitic fungi.

VI. A MORE TECHNICAL ACCOUNT OF THE DISEASES.

1. **Vermicularia Dematium** (Pers.) Fr. (The Stem-anthracnose).

The parasitism of the genus Vermicularia is well established. In the United States it has been noted as a disease-producing fungus on the following cultivated plants—Onion[4], Carnation[5], and Blue-grass[6].

If one makes transverse sections of an affected ginseng stem, the hyphae of the fungus are found principally in the cells of the sclerenchyma and cortical layers. They are easily recognized by their short, barrel-shaped cells and dark brown color. After a longer or shorter life within the host-plant, the fungus makes its way to the surface and forms the erumpent perithecia-like bodies in which the conidia are produced. (Fig. 4, A, B, and C.)

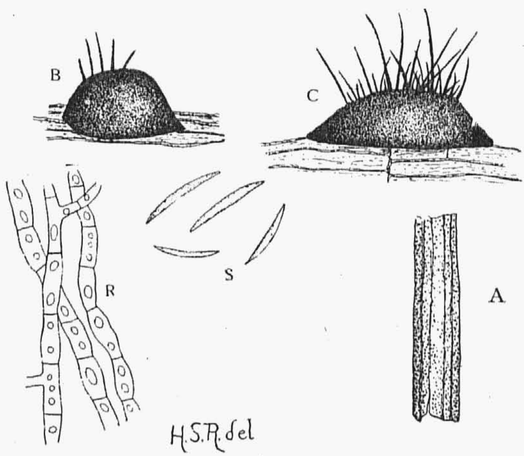

Fig. 4. Vermicularia Dematium. A.—Fragment of diseased ginseng stem, showing the black spore-producing bodies of the fungus. Natural size. B.—A young "perithecium." C.—A nearly mature "perithecium." R.—Hyphae of the fungus. S.—Spores.

[4] Annual Rept. Conn. Agr. Exp. Sta. 1889.
[4] Annual Rept. New Jersey Agr. Exp. Sta. 1890.
[5] Annual Rept. New Jersey Agr. Exp. Sta. 1890.
[6] Bull. 29. So. Dak. Agr. Exp. Sta. 1891.

Cultural Characters. Cultures of the fungus were made from affected stems in Petri dishes of ginseng agar. By using a flamed scalpel it was usually possible to cut out small bits of the stems under sterile conditions. When a pure culture was isolated from the Petri dishes, the fungus was cultivated upon a variety of media.

Cultures made on sterile potato plugs developed small patches of white mycelium on the second or third day, at the end of one week they were grayish white, and at the end of two weeks they were distinctly black in places. The addition of small quantities of lactic or malic acid seemed to accelerate slightly the growth of the mycelium. On acid media there was a greater tendency for the mycelium to spread, while on alkaline media, it formed compact tufts. When grown upon cooked rice to which a few drops of malic acid had been added, the mycelium formed a compact growth over the surface of the rice and down through the interstices between the rice grains. The best growth was obtained upon sterile plugs of sugar beet to which a few drops of dilute malic acid were added.

The branched hypae have a distinct Dematium-like appearance (Fig. 4, R). The cells have a length of 10-12 microns in young hyphae, but in mature hyphae are 30-35 microns in length. They are hyaline when young, turning brown as they become older. The contents of the cells include a large amount of fatty or oily material which exists in the form of refractive globules.

Cultures made on cornmeal and steamed rice developed the characteristic black color, but the stroma was not well developed. Tube cultures of the fungus on sterile plugs of potato and sugar beet showed some indications of stroma where the plugs came in contact with the wall of the tube. No perithecia-like bodies nor spores were produced on the media used, although they occur in great numbers on the ginseng plants. These "perithecia" are at first somewhat conical but subsequently become flattened, and have a diameter varying from 85-120 microns. (Fig. 4, B. C.) They are thickly beset with slender, black spines, among whose bases the spores are given off. The spores (Fig. 4, S) are hyaline, spindle-shaped bodies, having a length of 20-22 microns and a transverse diameter of 4 microns.

The manner of infection was not observed, but from the nature of the disease, I infer that infection takes place on the green parts of the host-plant. In all events, we know that the fungus may be kept out by spraying the tops of the plants with poisonous solutions.

In the crustaceous, perithecia-like fruiting bodies, the spores are well protected and are able to retain their vitality from one growing season to the next. I found that the mycelium does not retain its vitality in culture more than three or four months.

2. **Pestalozzia funerea** Desm. (The Leaf-anthracnose).

Characters in Cultures. Cultures of this fungus were obtained from the upper parts of dead stems. Dilution cultures were made in sugar beet juice agar by scraping off spores with a sterile needle. From these dilution cultures in Petri dishes pure cultures were obtained and the fungus was cultivated on different media with the results indicated below.

On sterile potato plugs. The growth of mycelium was very extensive in tubes to which a few drops of weak malic acid solution was added. At the end of 12 days the plugs were covered with a dense cottony growth of white mycelium. At the end of the fourth week the white mycelium was dotted with numbers of black pustules containing conidia. In tubes which had been rendered alkaline with sodium carbonate solution there was very little growth and no spore production.

On chopped onions. The fragments of onion bulbs were placed in small Erlenmeyer flasks, were rendered acid by the addition of a few cubic centimeters of weak malic acid, and sterilized in the autoclav. The fungus did not grow extensively on this medium, forming only a loose network of mycelium on the upper part of the culture. Nevertheless, at the end of four weeks it had produced a number of spore pustules.

In tubes of cooked rice+malic acid. The growth started off slowly but eventually produced a large amount of mycelium. Numerous black pustules were formed in the interstices between rice grains.

In flasks of cooked gluten meal+n|300 hydrochloric acid+grape juice. The mycelial development was here very extensive. At the end of twelve days the surface of the meal was covered with a thick white feltwork of mycelium. At the end of the fourth week it was dotted with small, black pustules containing conidia.

In flasks of cornmeal+n|300 hydrochloric acid+cane sugar+grape juice. The growth of the mycelium on this medium was very similar to that on preceding, but spore formation was much greater.

MICROSCOPICAL CHARACTERS.

Material was obtained from the cultures for a microscopical study of the fungus. The branching hyphae are composed of cells which, when young, are quite short, but elongate as the hypha grows (Fig. 5, B, and C). The filaments are more or less constricted at the septa. The black pustules contain conidia embedded in a mucilaginous slime. The conidia have three dark median and two hyaline terminal cells, but the basal cell of the median trio is

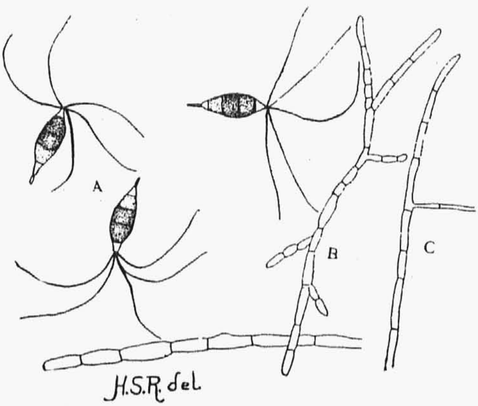

Fig. 5. Pestalozzia funerea. A.—Spores. B.—A young mycelium. C.—Hyphae of an older mycelium.

usually lighter colored than the other two. The basal hyaline cell bears a short basidium, and the apical hyaline cell bears four or five spreading ciliate appendages.

Name. The identity of the fungus has been difficult to determine. It appears to belong to the species which Desmazieres described in 1843 as *Pestalozzia funerea,* although I have been unable to make any comparison with authentic specimens. Since it answers to the meagre description transcribed in Saccardo's Sylloge, it is tolerably safe to assume that it belongs to the above mentioned species. I have given its cultural peculiarities in some detail in order that its identity may rest on more characters than those shown by the spores.

3. **Neocosmospora vasinfecta var. nivea** (Atk.) Smith.

Historical. In 1892 Atkinson described a Wilt-disease[7] of cotton and okra caused by a fungus which infested the fibro-vascular bundles of the plants. The fungus was found only in the fibro-vascular bundles and caused their walls to turn yellowish-brown. After growing it in pure cultures and obtaining the conidia, he gave it the name *Fusarium vasinfectum.*

Nothing further was known until 1899, when Erwin F. Smith described the Wilt-disease of cotton, watermelon, and cowpeas[8]. He found that spores obtained by Atkinson belonged to one of three conidial stages and that there was, in addition, an ascospore stage. Smith established a new genus, Neocosmospora, based upon the characters he found. The Wilt-fungi of cowpea and watermelon

[7] Bull. 41. Ala. Agr. Exp. Sta. 1892.

[8] Bull. 17. Div. of Vegetable Physiol. and Pathology, U. S. Dept. Agr., Washington 1899.

were varieties of the species which infested the cotton, and cross-inoculations failed. Cowpeas, for example, could not be infected with cultures of the melon or cotton fungus.

Orton has followed the studies of Smith on the Wilt-fungus of cotton[9] and cowpea[10] in the field. He reports that it is actively parasitic, and retains its vitality from year to year in the soil.

Wilt-fungi have also been discovered in tobacco[11] and China asters[12], which are probably caused by Neocosmospora.

Presence of the Wilt-fungus in Ginseng. The first indication of the Wilt-fungus is the drooping and fading of the ginseng leaves. If one takes up such a plant and examines a section from the upper part of the root, he will notice that the fibro-vascular bundles are decidedly yellow. I have never found a root so badly infested

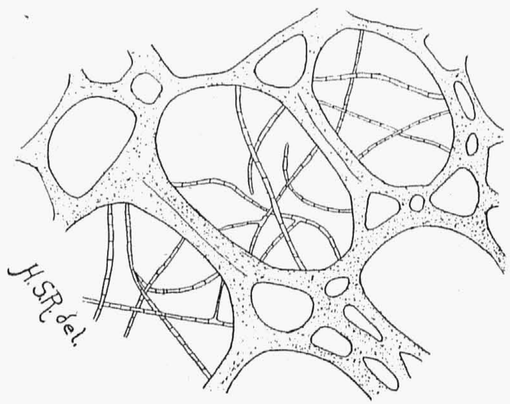

Fig. 6. Transverse section of part of a fibro-vascular bundle of a stem, showing the filaments of the Wilt Fungus (Neocosmospora).

that the walls were brown, although they may become so. When a thin section of the stem or root is examined under the microscope, the presence of fungus hyphae in the vascular tissue is not hard to make out (Fig. 6). I have found the conidal tufts which produce the spores frequent on the interior of hollow stems.

The mycelium of the fungus is quite richly branched and hyaline (Fig. 7, B). According to the nature of the substratum, it may or may not form a stroma. Three kinds of spores were produced by the mycelium.

[9] Bull. 27. Div. of Vegetable Physiol. and Pathology, U. S. Dept. Agr., Washington. 1900.

[10] Bull. 17. Bureau Plant Industry, U. S. Dept. Agr., Washington. 1902.

[11] Bull. 51. Bureau of Plant Industry, U. S. Dept. Agr., Washington. 1902.

[12] Bull. 79. Massachusetts Agr. Exp. Sta. 1902.

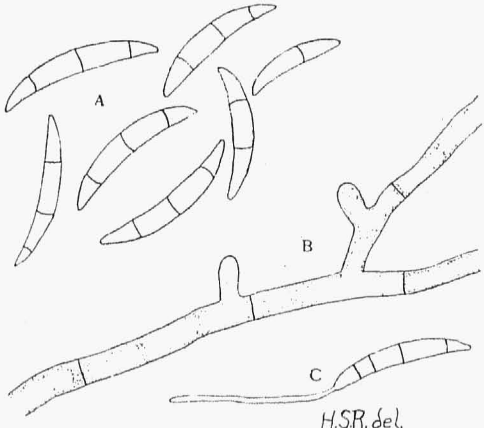

Fig. 7. Neocosmospora vasinfecta. A.—Macroconidia from culture on a bean-pod. B.—Portion of a hypha. C.—A germinating macroconidium in hanging drop culture.

Macroconidia (Fig. 7, A). These spores were found in greater abundance than any other. They were large, three to five septate, lunulate bodies having a length of 25-40 microns. When mounted in water they are hyaline, but in mass, are pale salmon color. They germinate by sending out a process at one end (Fig. 7, C), which grows into a hypha.

Microconidia (Fig. 8, D). These spores were not observed within the host-plant, but were obtained many times in pure cultures. They have the form of slightly flattened ellipsoids, ranging from 10-16 microns in length. The contents are perfectly hyaline and are very rarely divided by a septum.

Chlamydospores (Fig. 8, E). These spores were found in greatest abundance in cultures on sterile horse-dung. They appear as

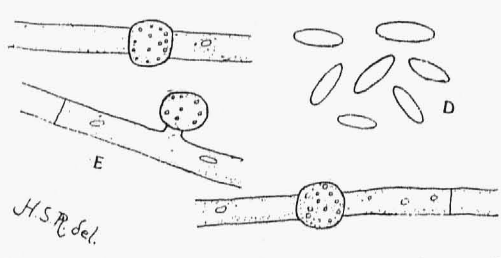

Fig. 8. Neoscosmospora vasinfecta. D.—Microconidia. E.—Chlamydospores. Both from a culture two weeks old on sterilized horse dung.

terminal or intercalary bodies, 10-14 microns in diameter, having coarsely granular contents. When mounted in water, the walls of the spores are light brown in color.

OBSERVATIONS BASED ON PURE CULTURES.

Mode of making Cultures. The following methods were employed in starting pure cultures of the fungus. Roots were thoroughly washed with water and scrubbed with a 1-10 per cent solution of mercuric bichloride. With a scalpel, which was sterilized in a flame, pieces of diseased roots and stems were cut out and transferred to sterile media. Several substances served as media, chiefly the following: cooked bean pods; cooked plugs of sugar beet; cooked stems and leaves of the ginseng plant; and nutrient agar made up with a decoction of ginseng plants.

Other cultures were inoculated with spores from the interior of dead stems. The spores were transferred by means of a sterile needle to plates of bean juice agar, beet juice agar, and ginseng agar. The best results were obtained from the use of bean pods, and ginseng agar.

From the cultures thus obtained, transfers were made to a variety of media, and the characters of the fungus were studied in a variety of conditions.

Germination. On acid beet and potato plugs, germination usu ally occurred in a very short time. At the end of twelve to fifteen hours (room temperature 19 -23 degrees C.) conspicuous tufts of mycelium could be seen. On cooked rice the period of germination was about twenty-four hours; and on grape-juice, sixty hours.

Characters in Culture. Since the cultural characters of the fungus agree so closely with those described by Smith, I will condense the description into tabular form.

	MEDIUM	CHARACTER OF MYCELIUM	SPORE FORMATION
1a	Slices of steamed potato in Petri dish (acid)	Growth good. Color grayish-blue. Stroma not generally formed.	Large quantities of macroconidia produced in two weeks.
1b.	Slices of steamed potato in Petri dish (alkaline)	Large handsome patches of flesh-colored mycelium. Gelatinous stroma formed where growth was small.	Macroconidia formed, but not so abundantly as in 1a.
2a.	Plugs of steamed potato in test tubes (acid)	Gray-blue covering the substratum. Aerial portions have a tendency to become spinose.	Macroconidia produced in enormous numbers.
2b.	Plugs of steamed potato in test tubes (alkaline)	Growth good but less than 2a.	Macroconidia present in smaller numbers than in 2a.
3a.	Plugs of steamed sugar beet in test tubes (acid)	Growth good, developing red-violet color.	Proceeded slowly, but eventually produced a large number of macroconidia.
3b.	Plugs of steamed sugar beet in test tubes (alkaline)	Growth somewhat less than 3a, developing same color.	Proceeded slowly, but eventually produced a large number of macroconidia.
4.	Chopped onion (acid)	Poor growth.	Macroconidia developed in part of the culture.
5.	Sterilized grape-juice	Poor growth.	No spores formed.
6.	Sterilized plum juice	Very slight growth.	No spores formed.
7a.	Cooked rice	Good growth. Somewhat red-violet.	Microconidia abundant at expiration of 4 weeks
7b.	Rice + lactic acid	Good growth of red-violet mycelium.	

MEDIUM	CHARACTER OF MYCELIUM	SPORE FORMATION
7c. Rice + sodium carbonate	Fair growth of bluish mycelium.	
7d. Rice + sodium carbonate + cane sugar	Mycelium all blue.	
8a. Cooked corn-meal	Fair growth.	
8b. Cooked corn-meal (acid)	Slight growth of white mycelium.	Large numbers of microconidia, few macroconidia.
8c. Cooked corn-meal, + grape juice + hydrochloric acid + cane sugar	Fair growth.	
9. Nutrient agar agar + 12 % cane sugar	Good growth of intensely red-violet mycelium and stroma.	
10. Sterilized horse dung	Moderate growth of pinkish mycelium.	Large quantities of micro- and macroconidia, together with chlamydo spores.

Inoculation Experiments. The writer very much regrets that it was impossible to carry out a full series of inoculation experiments in connection with his studies. Such as were performed are here reported.

Six crocks were filled with moist earth and sterilized in the autoclav for 45 minutes, at a temperature of 115 degrees C. When cool they were planted with seeds of cowpeas which had been sterilized in M-200 potassium bichromate solution for 4 hours. The crocks were carried to the plant house and set on glazed china plates. From day to day they were watered with sterilized water. Four weeks after planting, the plants were inoculated with spores of Neocosmospora in the following manner:

Jar 1. Control.

Jar 2. Inoculated by digging a hole in the soil (which broke numerous roots) and pouring into it distilled water containing macroconidia.

Jar 3. Inoculated by uncovering roots (without breaking them) and pouring on them distilled water containing spores.

Jar 4. Inoculated by introducing a quantity of soil taken from a bed of diseased ginseng plants.

The plants lived and grew for over a month after inoculation but showed no signs of wilting. (They were eventually killed by aphids.)

In describing his inoculation experiments, Smith does not state that sterilized soil was used. Another experiment was performed in which the writer used unsterilized soil.

On December first cultures of Neocosmospora on sterile horse-dung were placed in each of six crocks of good soil. Three crocks were planted with cowpeas and three with watermelons. Two uninoculated crocks served as controls. Three weeks later all 3 crocks of watermelon seedlings showed the presence of the wilt fungus, while the seedlings in an uninoculated crock were healthy. When sections of the wilted seedlings were examined under the microscope, they showed a pinkish mass of fungus hyphae and spores in the xylem of the fibrovascular bundles. The cowpeas showed no infection with the wilt fungus.

Identity. From its behavior in attacking the watermelon, there can be little doubt but that this fungus is the variety *nivea* of Smith, since he found that cotton and cowpeas could not be cross inoculated.

It will be noted that I have observed no perithecia either on diseased plants or in cultures. Since, however, the other three forms of spores were observed and found to agree closely in all respects to those described by Smith for *Neocosmospora vasinfecta*, I do not hesitate to place the fungus in that species, and refer it to the variety *nivea*.

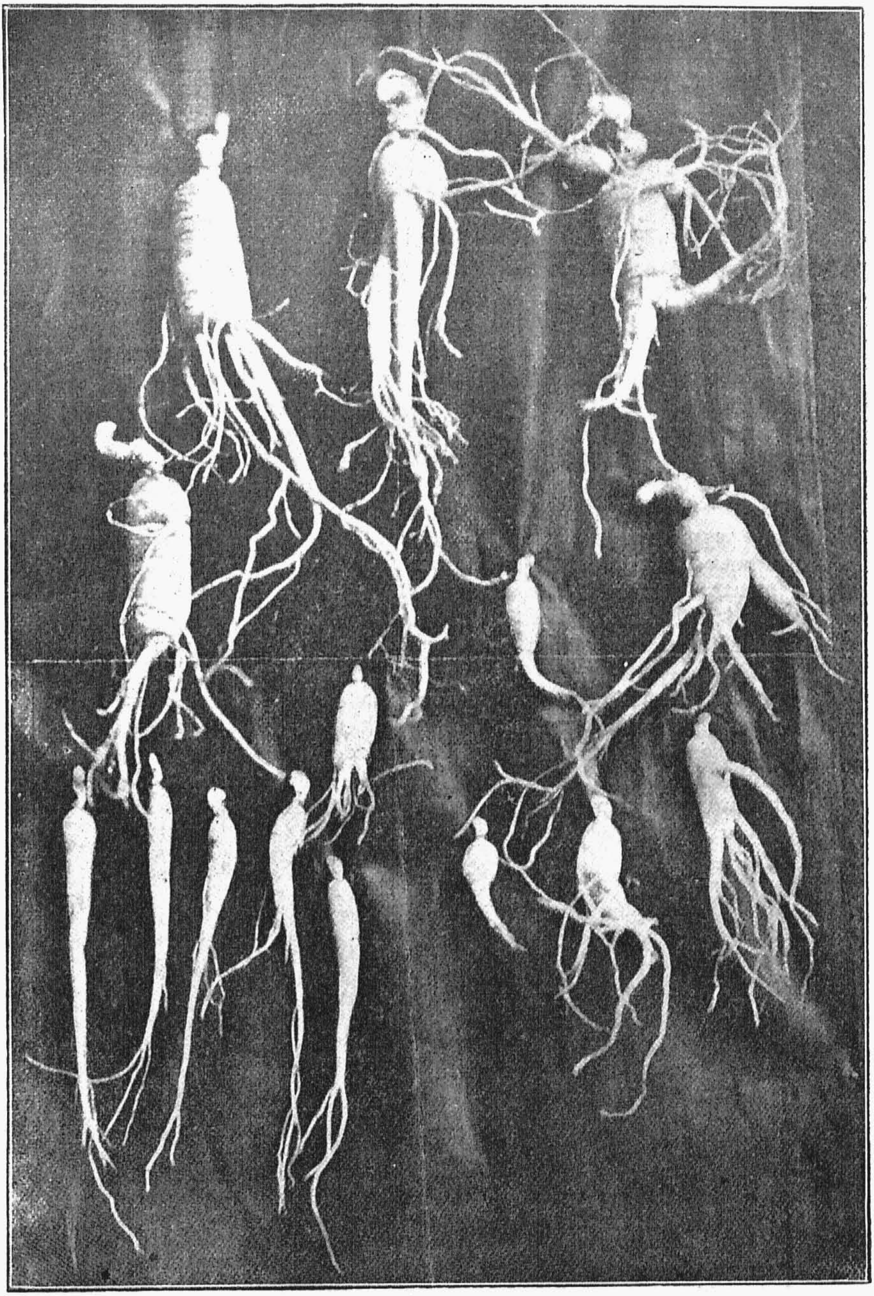

Fig. 9. Ginseng roots, one, two, three and four years old. (Cut used by courtesy of the Ozark Ginseng Co., Joplin, Mo.)