

Atypical Disease Symptoms Associated with *Phymatotrichum* Root Rot of Cotton

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ABSTRACT

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Cotton plants showing atypical symptoms of *Phymatotrichum* root rot were observed during the 1983 growing season. Aboveground symptoms consisted of gradual wilting followed by leaf chlorosis and defoliation. Mature bolls and young leaves remained attached to the stem and the plant remained alive. In contrast, foliar wilting normally occurs very rapidly and the plant dies, but the leaves remain attached. Externally, roots displayed discrete discolored sunken lesions 10–20 cm below the soil surface as opposed to typical lesion formation at the soil surface. Basipetal to this lesion, typical *Phymatotrichum* root rot symptoms had developed, the periderm and phloem were destroyed, and strands of *Phymatotrichum omnivorum* were visible. However, acropetal to the lesion, tissues appeared healthy with no evidence of the fungus. Internally and acropetal to the lesion, the root xylem showed extreme discoloration that usually extended to the transition zone between root and stem. Attempts to isolate the fungus from this region failed, and microscopic examination confirmed that the fungus had not advanced acropetal to the external lesion. The atypical symptoms were observed only during periods of low soil moisture availability. When soil moisture was replenished, strands of the fungus resumed growth on the root surface up to the soil line and plants previously showing atypical symptoms died.

wood Medical Industries, St. Louis, MO). Specimens were sectioned (15 μ) for light microscopy with a rotary microtome, mounted on slides with Haupt's adhesive, and stained with Triarch's quadruple stain. Standard isolation techniques were also conducted with roots of atypical plants. Roots were washed in distilled water, cut into 2-cm sections, surface-sterilized for 1 min in 0.5% sodium hypochlorite, and plated onto potato-dextrose agar amended with 200 μ g/ml of streptomycin. The plates were incubated at 28 C. Root segments were checked daily for fungal growth over a 5-day period to determine the extent of root infected by *P. omnivorum* and also to check for other organisms that differed from the usual soil microflora.

RESULTS

Plants began to die from *Phymatotrichum* root rot about 42 DAE, and the first disease count was made 56 DAE. Disease developed rapidly early in the season, increasing from 3% at 56 DAE to 22% at 71 DAE, a rate of 1.3% per day (Fig. 1); however, during the next 36 days (72–108 DAE), disease increased only 0.44% per day. During this period, the mean soil temperature at 30 cm was 26.5 C, but soil moisture levels at 30 cm dropped to a low of –22 bars. Plants showing atypical symptoms were observed at this time in all fields on the research farm. Initially, few of the plants showed atypical symptoms, but toward the end of the dry period (about 100 DAE), plants with atypical symptoms made up an estimated 10–15% of the total plant population. As the percentage of plants dying from *Phymatotrichum* root rot decreased, the percentage of plants showing atypical symptoms increased, although actual percentages were not determined.

Disease symptoms atypical of *Phymatotrichum* root rot were observed on aboveground and belowground portions of affected plants. Mature, fully expanded leaves wilted during the day but regained partial turgor in the evening. These leaves eventually became chlorotic and dropped from the plant. The stem and young leaves at the apical meristem remained alive. This is in contrast to the rapid wilt and plant death with leaf retention normally observed (Fig. 2).

Atypical root symptoms consisted of discrete sunken lesions coalescing and girdling the root 10–20 cm below the soil

purpose of this paper is to report atypical root and foliar symptoms associated with *Phymatotrichum* root rot and to discuss possible reasons for their appearance.

MATERIALS AND METHODS

Four fields at the Blackland Research Center in Temple, TX, were planted to cotton (GP-3774) on 6–12 April. The average date of plant emergence was 25 April. Extensive temperature, moisture, and disease measurements were taken in only one field, but because of the proximity of the four fields, climatic and soil conditions were similar. Soil temperatures were measured using copper-constantan thermocouples and soil moisture content was determined by the neutron-scattering method (4). Temperature and moisture measurements were recorded at weekly intervals at six locations within the field and related to disease progression.

Rate of disease development was measured as the percentage of plants that died each week in six two-row plots 30 m long. First disease counts were made on 20 June, 2 wk after initial appearance of root rot symptoms, and were continued until harvest. Plants showing atypical symptoms of *Phymatotrichum* root rot were first noticed about 68 days after emergence (DAE). Representative samples were harvested and taken to the laboratory for further study. Taproots from these plants were cut into 50-mm sections and fixed in formalin-propionic acid-propanol (FPP) for 2 wk. Sections were dehydrated 2 wk in an isopropyl alcohol series, then vacuum-infiltrated and embedded in Paraplast Plus (Sher-

Phymatotrichum root rot is the most economically serious disease of cotton in the Blackland regions of Texas (1). Foliar symptoms of the disease normally develop with extreme rapidity. Beginning at time of flowering, apparently healthy plants wilt and die within 2–3 days. The first symptom of *Phymatotrichum* root rot is a slight bronzing of the leaves followed by a rapid wilt (3,7). Leaves become flaccid and later desiccated but do not fall from the plant. Roots of wilted plants are typically covered by numerous fungal strands extending to the soil surface. Near the soil line, the fungus changes from strand development to a floccose growth. It is at this point that discolored sunken lesions coalesce, reportedly causing plant death (5). At times, slight xylem discoloration extends into the stem, but the fungus cannot be isolated above the soil line (2).

During the growing season of 1983, cotton plants began to show foliar symptoms similar to those caused by excessive moisture stress. Upon excavation of these plants, strands of *Phymatotrichum omnivorum* were always visible but restricted to the lower portion of the tap root (> 10 cm below the soil surface). The

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surface instead of at the soil surface (Fig. 3). Basipetal to this lesion, typical root rot symptoms were observed both internally and externally; however, acropetal to the lesion, variations from the norm were observed. The external portion of the root appeared healthy with no evidence of *P. omnivorum*, but internal xylem tissue was discolored a deep burgundy extending from the lesion to the root stem transition zone. Discoloration was primarily restricted to the xylem. Removal of the periderm from root segments of plants showing atypical symptoms revealed apparently healthy phloem tissue (Fig. 4).

Although xylem tissue was obviously affected by the fungus as indicated by the extensive discoloration, all attempts to isolate *P. omnivorum* acropetal to the lesion failed. Conversely, *P. omnivorum* was always recovered from isolations basipetal to the lesion. Microscopic examination of xylem tissue acropetal to the lesion also failed to detect any presence of *P. omnivorum* (Fig. 5). In every instance where root systems of plants showing atypical symptoms were inspected, *P. omnivorum* was always observed on the lower portions of the taproot. We found no evidence that any other organisms were involved in the development of atypical root rot symptoms in cotton.

As stated, plants with atypical symptoms were first noted about 68 DAE and reached a peak about 100 DAE. Soil moisture levels increased after this period with a concomitant increase in disease. A correlation between rainfall and disease development resulted in a correlation coefficient of 0.94. After soil moisture levels increased, plants with atypical symptoms began to die. When excavated, the old lesion could still be detected 10–15 cm below the soil surface, but strands of *P. omnivorum* had resumed growth and reached the soil surface where the typical floccose growth and lesion development occurred.

DISCUSSION

Atypical symptom development of *Phymatotrichum* root rot on cotton was closely associated with reduced soil moisture levels and resembled symptoms caused by severe water stress. The resemblance to water stress can explain why these symptoms have not been reported previously since *Phymatotrichum* normally kills its host. On most of the roots showing atypical symptoms, one or two lateral roots above the sunken lesion were usually present (Fig. 3). Olsen et al (5) reported that resistance to water flow in infected cotton root tissue was 40 to infinite times greater than in healthy tissue. This could adequately explain the observed foliar symptoms. Most of the root system was, in terms of water uptake, cut off from the plant, and only the portion above the lesion was able to

supply the plant with water. With the reduced capacity for water uptake, the plant response was defoliation. Identical plant responses can be seen when roots are pruned during cultivation.

Although the observed foliar symptoms can be adequately explained by reduced root capacity for water uptake, the reason for cessation of strand growth is unknown. The heavy clay soils of the Blackland regions of Texas are composed

primarily of the clay mineral montmorillonite, which is characterized by a high shrink-swell capacity. As these soils dry, they shrink, often pulling away from plant roots. *P. omnivorum* does not grow above the soil surface, although stem tissue offers no resistance to invasion when placed under conditions favorable for fungal growth (2). Whether fungal growth acropetal to that point is prevented by higher temperatures,

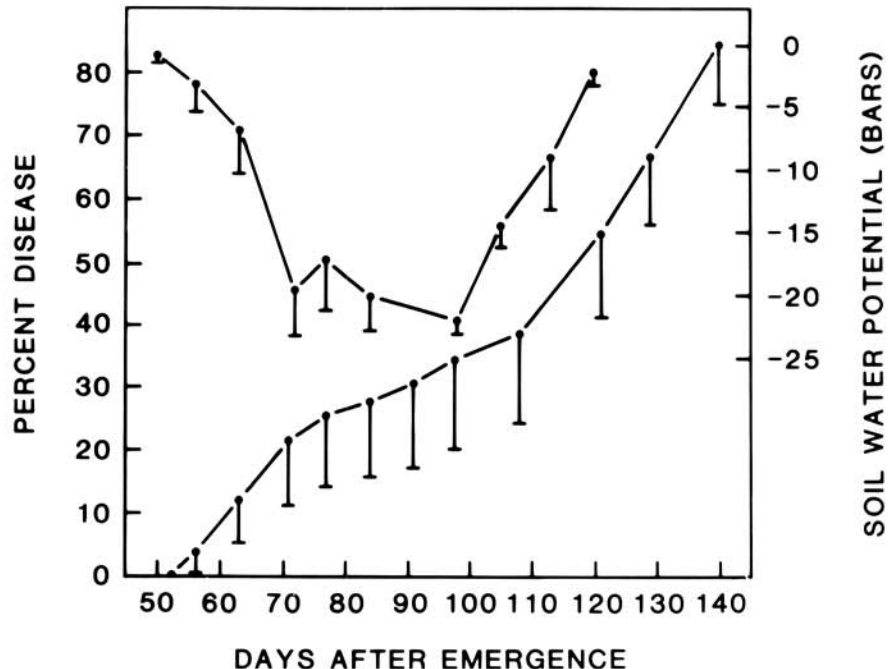


Fig. 1. Soil water potential and root rot development during the 1983 growing season (6). The upper curve shows changes in soil water potential at a depth of 30 cm. The lower curve is for disease progression. High soil moisture correlated highly ($r = 0.94$) with an increased rate of disease development.

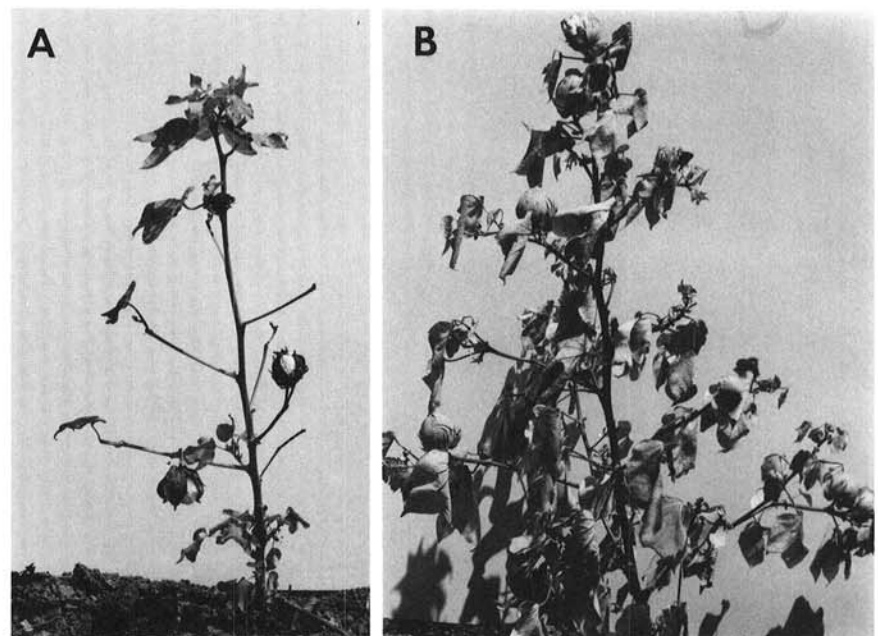


Fig. 2. Atypical and typical foliar symptoms of *Phymatotrichum* root rot. (A) Defoliation of older leaves, survival of leaves at the apex, and the main stem remaining alive were atypical symptoms that were closely associated with low soil moisture content. (B) Typical symptoms of root rot, stem death, and wilted leaves that remain attached to the petiole.



Fig. 3. External root symptoms of *Phymatotrichum* root rot. The root at left represents typical development of *Phymatotrichum omnivorum* on a cotton root. The center root, representing atypical development, shows the sunken lesion that was 15 cm below the soil surface. The taproot and laterals above the lesion appear healthy. Compare with healthy root at right.

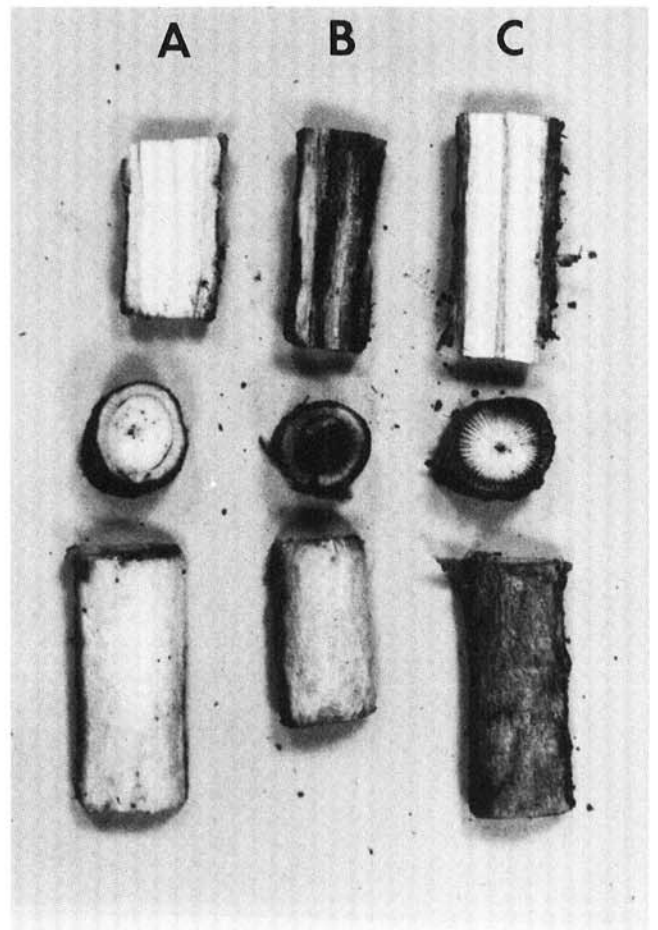


Fig. 4. Root segments taken near the soil surface. Column A = not infected. B = atypical symptoms, and C = typical symptoms. Bottom row of column B shows apparently healthy phloem associated with extremely discolored xylem in middle row. Compare with typical symptoms of roots in column C.

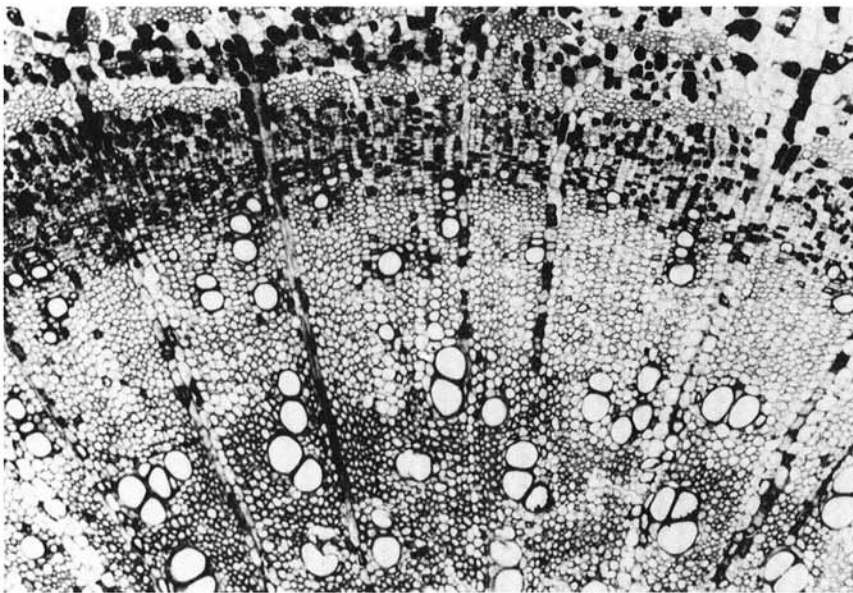


Fig. 5. Cross section of root with atypical symptoms. Section was taken above the lesion. No symptom of root rot or sign of *Phymatotrichum omnivorum* is present.

oxygen tensions, lower water potential, or some other factor is unknown. When the soil pulled away from the upper portion of the root as the soil dried, however, this possibly simulated above-

ground conditions, resulting in the cessation of strand growth and the appearance of the lesion on the lower portion of the taproot.

Growth of *P. omnivorum* inside the

root was not observed to occur as previously described by Watkins (8,9). He reported that once *P. omnivorum* reached the xylem, it could grow relatively unrestricted in the xylem vessels and that fungal mycelium could be found well in advance of any external lesion. In this study, *P. omnivorum* was never seen or isolated from xylem tissue acropetal to the external lesion but was easily recovered from xylem tissue basipetal to the lesion. This is in agreement with other reports (2,5). A possible explanation is that Watkins worked with cotton seeding in vitro as opposed to field grown plants (9-11).

The excessive xylem discoloration associated with healthy phloem tissue acropetal to the lesion was another interesting aspect of atypical symptomatology. Xylem discoloration acropetal to the lesion suggested translocation of fungal metabolites, possibly toxins. Toxins have been implicated in pathogenicity of *P. omnivorum* (8,10,11), but the exact nature of these toxins and their role in disease development is unknown. If xylem discoloration was the result of a toxin, it is significant to note that the toxin did not kill the plants. Plants were

not killed until soil moisture increased and the fungus resumed external strand growth to the soil surface with associated disruption of the phloem tissue. This supports the results of Olsen et al (5).

This study points out the inadequacy of our present understanding of the mode of parasitism and pathogenicity of *P. omnivorum*. The observations reported here suggest experiments that could help clarify certain aspects of this disease. If these symptoms could be reproduced in the laboratory, one would be able to separately determine the importance of xylem discoloration and fungal colonization to increased water resistance in root tissue infected by *P. omnivorum*. We

believe that basic studies of the effects of water potential on toxin production and the initial infection process would provide increased understanding of the pathogen and its mode of parasitism.

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