TAXONOMY AND PATHOGENICITY OF ECTOTROPHIC FUNGI WITH *PHIALOPHORA* ANAMORPHS FROM TURGRASSES

BY

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ABSTRACT

Smiley and Craven Fowler (1984) implicated the fungus Phialophora graminicola (Deacon) Walker as the incitant of summer patch disease of Kentucky bluegrass (Poa pratensis L.) turf. Paradoxically, P. graminicola was reported previously as a non-pathogenic fungus with potential as a biological control agent against take-all of cereals and take-all patch of bentgrasses (Agrostis spp.) (Deacon, 1973; Deacon, 1974; and Wong, 1981). In consequence, this study was undertaken to reexamine the etiology of summer patch disease and establish the identity of the incitant.

The specific objectives of this study were to isolate and identify the ectotrophic Phialophora species from the roots and crowns of turfgrasses exhibiting symptoms of summer patch disease and determine the relative pathogenicity of the individual species. In addition, efforts would be made to determine the host range of the most aggressive pathogen and attempt to reproduce typical summer patch symptoms by inoculating mature (22-month-old) Kentucky bluegrass sod with the fungus.

The three species found during this investigation included the anamorphs of Magnaporthe poae (Phialophora) sp. nov., Gaeumannomyces incrustans (Phialophora) sp. nov., and Gaeumannomyces cylindrosporus Hornby, Slope, Gutteridge, and Sivanesan (anamorph P. graminicola). Identification of M. poae and G. incrustans were based on ascomata produced by

pairing opposing mating types on either side of sterile wheat stems in Sach's agar (Luttrell, 1958). The identity of the P. graminicola isolates was established by comparing them with single-ascospore cultures obtained from G. cylindrosporus.

Isolates designated as P. graminicola (ATCC 64413, ATCC 56773, NY-258, NE-179, and ATCC 60239) and cited as the cause of summer patch were obtained from Smiley and associates. A comparison of these isolates with single-ascospore cultures of G. cylindrosporus revealed morphological and physiological differences in vitro. Subsequent production of the teleomorphs by these isolates proved that the fungus reported as the incitant of summer patch had been incorrectly identified and that the fungus was actually M. poae.

Of the three species studied, M. poae was the most pathogenic on both 8-week-old and 7-year-old Kentucky bluegrass turf at 28°C. Gaeumannomyces incrustans was mildly pathogenic while G. cylindrosporus was not pathogenic on Kentucky bluegrass. Magnaporthe poae was also pathogenic on other species of turfgrasses and cereals.

When inoculated into mature (22-month-old) Kentucky bluegrass turf, M. poae produced patches which were identical to field symptoms of summer patch. M. poae was reisolated from the roots and crowns of tillers from diseased plants. These results indicate a causal relationship between M. poae and summer patch disease.

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INTRODUCTION

Patch diseases caused by root and crown-infecting fungi are among the most destructive diseases of turfgrasses in the United States (Couch, 1985; Dernoeden, 1986; Endo and Colbaugh, 1974; Smiley and Craven Fowler, 1985a; and Snow and Watschke, 1986). Symptoms appear as ribbons or roughly circular patches of dying or dead turf, often ring-shaped, with a tuft of healthy grass in the center. Roots and crowns of affected tillers are often rotted, appearing brown or black in color. Patch diseases severely disfigure turf and influence the uniformity of sports turf playing surfaces. Since root and crown tissues are destroyed, recovery growth is slow, and affected areas are often filled in by undesirable grasses or broadleaf weeds.

As a group, patch diseases are among the most intensively studied, but least understood diseases of turfgrasses. The latter is primarily due to difficulties in identifying the primary causal agents involved (or lack of acceptance of proposed causal agents) as well as inconsistencies reported for cultural and chemical control measures (Smiley, 1987b). One group of fungi has recently received a considerable amount of attention for its role in the development of patch diseases. Included are the ectotrophic, root-inhabiting fungi with Phialophora anamorphs and their teleomorphs in the genera Magnaporthe and Gaeumannomyces (Couch, 1985; Smiley and Craven Fowler,

1984; and Smiley, 1987b). Smiley (1984) proposed the name summer patch for a serious disease of Kentucky bluegrass (Poa pratensis L.) turf in New York State and implicated the fungus Phialophora graminicola as the causal agent. Summer patch is one component of what was a poorly defined complex of diseases previously referred to as Fusarium blight syndrome (Smiley, 1984).

Although Smiley and Craven Fowler (1984) reported that P. graminicola was the primary incitant of summer patch, previous studies had shown that P. graminicola is a non-pathogenic, ectotrophic, root-inhabiting fungus with potential as a biological control agent against take-all of wheat (Triticum aestivum L.) and take-all patch of bentgrasses (Agrostis spp.) (Scott, 1970; Balis, 1970; Deacon, 1973; Deacon, 1974; and Wong, 1981). Smiley et al. (1985b) suggested that this apparent contradiction could be explained by the cool temperatures under which all previous pathogenicity studies were conducted. While European and Australian studies with P. graminicola were conducted at 22°C or below, summer patch symptoms on closely mowed grasses only developed at temperatures above 25°C. Smiley et al. (1985b) also suggested that isolates of P. graminicola in the United States conceivably were more virulent than those from Europe or Australia. Another explanation is that their causal fungus was misidentified as P. graminicola.

The purpose of this study was to isolate and identify

the ectotrophic Phialophora species from turfgrasses exhibiting symptoms of summer patch disease and to determine the relative pathogenicity of the species involved. In addition, an attempt was made to induce patch symptoms on mature turf and to determine the host range of the most aggressive pathogen.

LITERATURE REVIEW

This review of literature is presented in two parts. Part one concerns patch diseases of Kentucky bluegrass (Poa pratensis L.) turf caused by root and crown-infecting fungi. Emphasis has been placed on the search for the primary causal agents involved in patch diseases as well as the influence of environment, cultural practices, and fungicides on disease development. Part two of this review concerns the taxonomy of Phialophora, Magnaporthe, and Gaeumannomyces species associated with grass roots.

1. Patch diseases of Kentucky bluegrass turf.

In the late 1950s and early 1960s a serious disease of Kentucky bluegrass turf appeared in the Northeastern and Mid-Atlantic regions of the United States (Couch, 1964; Couch and Bedford, 1966; Smiley, 1980; and Sanders and Cole, 1981). Couch and Bedford (1966) reported that the disease had become wide-spread in stands of 'Merion' Kentucky bluegrass during the 1960, 1961, and 1963 growing seasons in South Central and Eastern Pennsylvania. It was also observed in Ohio, New York, New Jersey, Delaware, Maryland, and Washington, D.C. (Couch and Bedford, 1966).

Symptoms of this disease initially appeared as scattered, light green patches 5-15 cm in diameter. As

conditions favored disease development, the patches changed to a dull reddish brown, then to tan, and finally to a light straw color. In shape, these patches were elongate streaks, crescents, or circular patches. The most distinctive feature of this disease was circular patches of blighted turfgrass 0.6 - 1 meter in diameter with tufts of apparently unaffected grass in the center (this feature was referred to as the 'frog-eye' effect). Leaf lesions were visible both at the cut tip and at random over the entire leaf. The lesions progressed from dark green blotches to a light green, then a reddish brown hue, and finally, a dull tan color. Lesions often extended over the entire width of the leaf (Couch and Bedford, 1966).

Couch and Bedford (1966) implicated two species of Fusarium as the causal agents of this disease, Fusarium roseum (Lk.) Snyd. and Hans. f. sp. cerealis 'Culmorum' and F. tricinctum (Cda.) Snyd. and Hans. f. sp. poae (Pk.). They based their assumptions on (1) consistency of isolation from diseased turfgrass plants over a broad geographic area for several growing seasons, (2) the general lack of pathogenicity or the inconsistency of continued association of other microorganisms, or both, and (3) the high degree of pathogenicity shown by the two Fusarium spp. on 6-8 week-old turf. Hence, they named the disease Fusarium blight.

Although Couch and Bedford (1966) did not mention a root and/or crown rot stage for this disease in their original paper, Bean (1966) found both a leaf blight and

crown rot stage on turfgrasses exhibiting Fusarium blight symptoms in Washington D.C.

A similar disease was reported in 1967 on Zoysia japonica Steud. 'Meyer' in Arkansas and on centipedegrass (Eremochloa ophiuroides (Monro.) Hack.) in Alabama in 1972 (Dale, 1967 and Subirats and Self, 1972). Fusarium spp. were implicated as the causal agents in both cases. Endo et al. (1973) reported the occurrence of Fusarium blight on Kentucky bluegrass in Central and Southern California. They noted that the disease occurred primarily as a foot rot and secondarily as a foliage blight. The primary pathogen was reported as F. roseum f. sp. cerealis 'Culmorum'. Fusarium blight has also been reported in a number of midwestern states (Endo et al., 1973 and Smiley, 1980).

Environmental conditions strongly influence incidence and development of Fusarium blight. Fusarium blight usually occurs in midsummer under conditions of high temperatures and humidity (Couch and Bedford, 1966). Couch (1976) claimed that optimum conditions for disease development included high atmospheric humidity with daytime air temperatures of 27-35 °C and night temperatures of 21 °C or above. Cole (1976) also noted the influence of high temperatures on the development of Fusarium blight, stating that the most severe problems occurred on the southern range of Kentucky bluegrass adaption where high midsummer temperatures were obtained. In California, Fusarium blight most often occurred during periods of hot, dry weather (Endo

et al., 1973). Smiley (1980) found that incidence of Fusarium blight coincided with areas where Kentucky bluegrass was grown and average daily summer temperatures exceeded 21 °C. However, Smiley (1980) noted that information regarding precise cardinal temperatures was unavailable.

Soil moisture is also considered to be important in the development of Fusarium blight, although reports differ as to its specific role. Couch and Bedford (1966) found no significant differences in Fusarium blight incidence among three different soil moisture regimes in pot studies. However, later studies showed disease severity was closely correlated with available soil moisture, the most severe infection occurring on drought-stressed turf (Bean, 1969 and Cutright and Harrison, 1970b). Field studies in Pennsylvania indicated that Fusarium blight was almost always preceeded by a period of warm, wet weather followed by dry weather (Fulton et al., 1974). Smiley (1980) found that major outbreaks of Fusarium blight were associated with periods of abundant moisture or with alternating periods of wetness and drought.

Light intensity is another factor influencing the development of Fusarium blight. Bean (1966) noted that areas receiving direct sunlight (i.e. south-facing slopes) were the first to develop Fusarium blight whereas the disease seldom occurred in shaded areas.

A number of cultural practices have been reported to

influence the development of Fusarium blight. Among the practices generally favoring an increase in disease severity are high levels of soil nitrogen (N) (Couch and Bedford, 1966; Cutright and Harrison, 1970b; Endo and Colbaugh, 1974; and Turgeon, 1976); low calcium nutrition (Couch and Bedford, 1966); application of hydrated lime (Sanders and Cole, 1981); and a low mowing height (Funk, 1976; Smiley and Craven, 1977; Turgeon and Meyer, 1974; and Turgeon, 1976). In some cases, however, no relationships were found between soil N, calcium nutrition, and application of lime and the severity of Fusarium blight (Bean, 1966; Bean, 1969; and Sanders and Cole, 1981). Bean (1969) suggested heavy irrigation during summer months as a means of reducing Fusarium blight incidence.

Substantial variation in Fusarium blight resistance occurs among Kentucky bluegrass cultivars (Funk, 1976; Turgeon, 1976; Meyer and Berns, 1976; and Smiley et al. 1981). Funk (1976) noted that varieties better able to tolerate environmental and cultural stresses were less likely to be damaged by Fusarium blight. Smiley et al. (1981) found that Kentucky bluegrass cultivars selected from areas of highest summer temperatures or of lowest latitudes tended to be least susceptible to Fusarium blight.

Fusarium blight resistance in other turfgrass species has been explored to a limited extent. Gibeault et al. (1980) reported that mixes of perennial ryegrass (Lolium perenne L.) and Kentucky bluegrass significantly reduced

incidence of Fusarium blight in California. Smiley (1980) obtained control of Fusarium blight by overseeding perennial ryegrass into bluegrass fairways that had been previously affected by Fusarium blight. Although Couch and Bedford (1966) claimed that Fusarium blight was prevalent on bentgrasses (Agrostis spp.), Sanders and Cole (1981) noted that it was absent from most bentgrass areas. Smiley (1980) claimed that overseeding creeping bentgrass (Agrostis palustris L.) into bluegrass fairways curtailed the development of Fusarium blight. Funk (1976) found an increase in Fusarium blight incidence on mixtures of fine-leaf fescues (Festuca rubra L.) (cv. 'Pennlawn' and 'Jamestown') and 'Fylking' Kentucky bluegrass over 'Fylking' alone.

Fusarium blight can be effectively controlled with a number of commercially available fungicides (although variable results have been obtained depending on timing and method of application). Couch (1964) found mancozeb (Dithane M-45) effectively controlled Fusarium blight in a preventative program. In a later test, Bean et al. (1967) reported that thiramorganic mercury (Tersan OM) was the most effective fungicide tested for control of Fusarium blight, while chlorothalonil (Daconil 2787) and mancozeb were ineffective. Cutright and Harrison (1970a) found that the systemic fungicide benomyl controlled Fusarium blight as a preventative spray. Thiramorganic mercury provided some control, while treatments of maneb and chloroneb had no

effect on Fusarium blight development. Vargas and Laughlin (1971) also reported the control of Fusarium blight with biweekly applications of benomyl as a drench. However, foliar sprays of benomyl were reported to be ineffective (Vargas and Laughlin, 1971). Failure to control Fusarium blight with benzimidazole-derivative fungicides was noted in the 1970s (Smiley and Howard, 1976 and Burpee et al., 1977). Isolates of Fusarium spp. from areas where control failures occurred proved to be highly tolerant and grew in vitro on media amended with 1000 ug benomyl/ml (Smiley and Howard, 1976). More recently, field studies demonstrated control of Fusarium blight with the systemic fungicides triadimefon (Sanders et al., 1978a; Smiley and Craven, 1977; Burpee et al., 1976; Nutter et al., 1979; and Loughner et al., 1979); ecticonizole (CGA 64251) (Nutter et al., 1979; Loughner et al., 1979; and Smiley and Craven, 1979a); fenarimol (EL222) (Brown, 1976; and Smiley and Craven, 1977) and iprodione (RP 26019) (Smiley and Craven, 1979a and Smiley and Craven, 1979b). Smiley (1980) reported that the relative efficacy of systemic fungicides for controlling Fusarium blight is triadimefon = ecticonizole = fenarimol › benomyl › methyl thiophanate › ethyl thiophanate = iprodione.

Several lines of evidence suggest inconsistencies in assigning F. roseum and F. tricinctum as the primary causal agents of all 'Fusarium blight' symptoms. Smiley (1980) claimed that many investigators, including himself, were unable to show that the Fusarium spp. pathogenic to

seedlings were also pathogenic to mature Kentucky bluegrass in the field. Bean (1969) noted that Couch and Bedford (1966) had demonstrated pathogenicity of F. roseum and F. tricinctum on 6-8 week-old turf but that the characteristic symptoms of Fusarium blight include the destruction of crowns in established (2 years or older) turf. The author attempted to reproduce field symptoms of this disease on established turf in the greenhouse using high spore concentrations (8.0×10^6 conidia/ml) but found that no leaf or crown infection occurred (Bean, 1969). Smiley (1980) insisted he could not induce leaf or crown necrosis in the field by 'painting' spore suspensions of 72 isolates of 10 species of Fusarium on mature, highly susceptible 'Fylking' Kentucky bluegrass. In addition, Fusarium-colonized food bases, such as cereal grains, applied as inoculum at various times of the year to highly susceptible grasses in the field, failed to induce disease symptoms (Smiley, 1980). Fulton et al. (1974) provided the only evidence that field symptoms could be approximated by inoculating mature Kentucky bluegrass sod with F. roseum and F. tricinctum in the greenhouse, yet they failed to induce the typical, serpentine or 'frog-eye' patch disease patterns.

Another inconsistency involved reports that the fungicide triadimefon totally suppressed field symptoms of Fusarium blight at various test locations (Burpee et al., 1977 and Smiley and Craven, 1977), yet showed no significant inhibition of F. tricinctum or F. roseum in vitro (Sanders

et al., 1978a). In addition, Sanders et al. (1978b) reported that iprodione had no significant activity against F. tricinctum or biotypes of F. roseum in vitro., yet field studies showed a 50-70 % reduction of symptoms of Fusarium blight. Smiley and Craven (1979a) found that iprodione significantly increased the number of Fusarium propagules in the crowns of turf and in soil over that of the controls. Smiley (1980) suggested that iprodione may act to suppress symptoms indirectly by altering host metabolism or the microbial balance in the soil and thatch. Another explanation he offered was that Fusarium spp. are not the primary causal agents of this disease.

A third line of evidence casting doubt on the role of Fusarium spp. in the development of Fusarium blight symptoms involved a study in which Fusarium blight severity was not correlated ($P = 0.10$) with the total no. of Fusarium propagules (including F. roseum and F. tricinctum). In addition, the relationship between the percentages of Fusarium infected crowns and of blighted turf indicated that the disease symptoms were least severe where crown infection was greatest (Smiley et al., 1980).

Several attempts have been made to determine the role of other causal agents (whether primary or in association as a complex) in the development of Fusarium blight.

Vargas and Laughlin (1972) initiated a survey to determine the role of nematodes in the development of Fusarium blight. They found that Tylenchorynchus dubius

(Butschli) Filipjev alone or in combination with F. roseum significantly reduced topgrowth of seedling turf when compared with nontreated controls. No significant differences were observed between plants inoculated with Fusarium alone and the controls. The same relationships were found when root weights were compared. The authors concluded that F. roseum by itself was not sufficient to explain the severe stunting of top growth and root growth and that the nematode may be predisposing bluegrass to attack by F. roseum. They went on to say that the nematode is the dominant pathogen in the disease interaction involving T. dubious and F. roseum (Vargas and Laughlin, 1972).

Smiley (1980) proposed a hypothesis whereby the gross symptomology of Fusarium blight could be a reflection of the synchronous death of most shoots of an entire bluegrass plant (rather than the independent death of individual shoots). The hypothesis takes into consideration the growth habit of Kentucky bluegrass. The shoots which arise from rhizomes connected to a single parent are new individuals of identical genotype. If these are subjected to a severe abiotic stress, the shoots respond in unison (as a synchronous unit rather than independently). Assuming this occurs, death of a small patch could be expressed if death is concentrated into a short time interval. In addition, stress-weakened plants of a given genotype could then be easily killed by antagonistic facultative symbionts (Smiley,

1980). This hypothesis has not been validated experimentally.

Pennypacker et al. (1982) conducted a study of Kentucky bluegrass plants exhibiting Fusarium blight symptoms in an effort to determine the fungi present at the onset of visible symptoms. Thirty-five percent (19/54) of the samples contained an unidentified basidiomycete and histological examination revealed abundant mycelium, with clamp connections, external to the epidermis and intracellular in the cortical cells of the crown and lower stem. Limited hyphal penetration and colonization of the vessel elements and tracheids of the xylem were noted. The authors, however, acknowledged they had no evidence that the cortical-infecting basidiomycete actually caused death of the plants (Pennypacker et al., 1982).

Smiley and Craven Fowler (1984) reported that two fungi with growth habits similar to Gaeumannomyces graminis (Sacc.) Arx and Olivier were associated with a patch disease of Kentucky bluegrass in New York State. The fungi, Leptosphaeria korrae Walker and Smith and Phialophora graminicola (Deacon) Walker were found to produce disease symptoms indistinguishable from those reported for Fusarium blight syndrome (Smiley and Craven Fowler, 1984 and Smiley 1983). Smiley, 1984 proposed the name summer patch for the disease favored by warm to hot, wet conditions caused by P. graminicola and necrotic ring spot (NRS) for the disease favored by mild, dry to wet environments caused by L.

korrae. A full description of symptoms for both these diseases was not provided, instead; reference was made to the symptoms of Fusarium blight syndrome (Smiley, 1983).

Jackson (1984), Smiley et al. (1984), and Chastagner et al. (1984) reported isolation of L. korrae from Kentucky bluegrass exhibiting patch disease symptoms in Rhode Island, New York State, and Washington, respectively. Chastagner and Byther (1985) reported the occurrence of NRS in Idaho, Oregon, and British Columbia. A L. korrae-like fungus was also isolated from Kentucky bluegrass exhibiting patch disease symptoms in Michigan (Otto and Vargas, 1985).

Leptosphaeria korrae was first reported as a turfgrass pathogen by Walker and Smith (1972) in Australia. Leptosphaeria korrae (as well as L. namari) was found to cause a patch disease of Bermudagrass (Cynodon dactylon L.) referred to as spring dead spot (SDS) (Walker and Smith 1972). Endo et al. (1985) have since implicated L. korrae as the causal agent of SDS in California and Dernoeden (personal communication, 1986) has found L. korrae in association with SDS symptoms in Maryland.

Leptosphaeria korrae has been isolated from P. pratensis, Poa annua L., Festuca rubra L., Agrostis spp., Axonopus sp., Cynodon sp., and Erenochloa sp. and is pathogenic to Avena sativa L., Oryza sativa L. and Triticum aestivum L. (Jackson, 1984; Walker and Smith, 1972).

NRS is generally favored by mild, dry to wet environments (Smiley and Craven Fowler, 1984). Worf et al.

(1983) found that 2-8 year-old turf was most likely to show symptoms of NRS and that heavy rains may interrupt symptom development until drier weather occures. Chastagner and Byther (1985) claimed NRS is most commonly observed on turf that has been overwatered and overfertilized. Worf and Stewart (1986) reported that symptoms are most commonly observed in midsummer but occur throughout the growing season. Chastagner and Byther (1985) found NRS occurring most frequently in late spring-early summer and/or late summer-early fall.

Chastagner and Byther (1985) recommended employing cultural practices which encourage deep rooting as a means of reducing the severity of NRS. The effect of specific cultural practices on disease severity, however, has not been determined. The most effective control of NRS has been achieved with the use of fungicides. Fenarimol and propiconizole have provided excellent control of NRS in Washington, while triadimefon, iprodione, and benomyl failed to provide adequate protection (Chastagner and Byther, 1985). In Wisconsin, drenches of iprodione and benomyl controlled NRS while triadimefon was ineffective (Worf and Stewart, 1985).

Smiley and Craven Fowler (1984) implicated P. graminicola as the causal agent of summer patch disease and noted its occurrence is favored by warm to hot, wet conditions. While there is no comprehensive survey of the geographical distribution of this disease, Snow and Watschke

(1986) noted it was widespread on golf courses in the Northeastern United States.

Although Smiley and Craven Fowler (1984) reported that P. graminicola was the primary incitant of summer patch, previous studies had shown that P. graminicola is a non-pathogenic, root-inhabiting fungus with potential as a biocontrol agent against take-all disease of wheat and take-all patch of bentgrasses (Scott, 1970; Balis, 1970; Deacon, 1973; Deacon, 1974; and Wong, 1981). Smiley et al. (1985b) suggested that this apparent contradiction could be explained by the cool temperatures under which all previous pathogenicity studies were conducted. While European and Australian studies with P. graminicola were conducted at 22°C or below summer patch symptoms on closely mowed grasses only developed at temperatures above 25°C. Smiley et al. (1985b) also suggested that isolates of P. graminicola in the United States conceivably were more virulent than those from Europe or Australia. Smiley and Craven Fowler (1984) described their isolates of P. graminicola as appressed colonies, initially colorless to light gray, darkening from the colony origin to an olive brown with growth rates of 4.7 mm/day at temperatures of 20-25°C. Other taxonomic features included slight curling back of hyphae at the periphery of colonies, germinating phialoconidia (5-13 x 2-4 um), subglobose hyphopodia occurring on rotting rhizomes and tiller bases of plants. The authors reported that numerous attempts to induce fruiting over a two year period failed

(Smiley and Craven Fowler, 1984). In a later study, Smiley et al. (1985b) found considerable differences in temperature optima and osmotic potential tolerences between North American isolates of P. graminicola and those from Europe and Australia (although no direct comparisons were made). Consequently, the authors suggested that North American isolates of P. graminicola may have different adaptions to edaphic conditions than those from other continents (Smiley et al., 1985b).

Inoculation of the fungus reported as P. graminicola into mature Kentucky bluegrass resulted in the development of patch symptoms in the field and under semicontrolled conditions (Smiley and Craven Fowler, 1984 and Smiley and Craven Fowler, 1985b). The authors reported reisolation of P. graminicola from diseased roots and rhizomes of Kentucky bluegrass from the individual patches (Smiley and Craven Fowler, 1984).

The growth and pathogenicity of P. graminicola (sensu Smiley) on Kentucky bluegrass is strongly influenced by temperature. Smiley et al. (1985b) demonstrated that P. graminicola did not produce ectotrophic growth after 2 weeks at 14 °C, but grew 2 cm/week at 24 °and 29 °C. The effect of temperature on pathogenicity of P. graminicola was examined on 7-week-old 'Merion' Kentucky bluegrass grown in pots and on mature (7-year-old), field-grown 'Merion' sod incubated at 14°, 21°, and 29°C (Smiley et al., 1985b). Disease symptoms were not apparent at 14 °or 21 °C, but all plants

were killed within 3-4 weeks at 29°C. Inoculated sod segments incubated at 14°C did not exhibit a decline in foliar quality over controls. Incubation at 21°C influenced foliar quality and growth but did not generate severe disease symptoms. Inoculated sod incubated at 29°C for 3-4 weeks, showed typical disease with greatly reduced growth and foliar quality, indicating that relatively high air temperatures promoted symptom development (Smiley et al., 1985b). Temperature studies of P. graminicola in culture revealed that maximum growth occurred at 28-31°C.

Smiley et al. (1986) reported that certain management practices influenced the severity of summer patch on Kentucky bluegrass. Summer patch was more prevalent in turf established on fumigated rather than non-fumigated soil. On fumigated soil, patches were larger in irrigated turf than in non-irrigated turf. Patches were more numerous but smaller in irrigated turf established on non-fumigated soil than patches in non-fumigated, non-irrigated plots. Fewer patches occurred in plots receiving applications of phosphorus and sulfur while the number of patches were variable in plots receiving N treatments (Smiley et al., 1986). Turfgrasses treated with moderate to high rates of arsenical herbicides showed increased susceptibility to summer patch in New York State (Smiley et al., 1985c).

Cultivars of seedling cool-season turgrasses inoculated with P. graminicola (sensu Smiley) revealed varying degrees of susceptibility (Smiley and Craven Fowler, 1986a-f).

Smiley (1987a) listed colonial bentgrass and fine-leaf fescue as extremely susceptible to root infection by P. graminicola followed by Kentucky bluegrass and creeping bentgrass (very susceptible) and perennial ryegrass and tall fescue (moderately susceptible).

2. Taxonomy of Phialophora, Magnaporthe, and Gaeumannomyces species from grass roots.

The genus Phialophora was established by Medlar (1915) for a fungus pathogenic on humans, Phialophora verrucosa Medlar, isolated from a skin lesion on the buttocks of a 22 year-old man. Medlar (1915) reported 2 types of conidial formation associated with P. verrucosa. The first, termed semi-endogenous, included conidia formed from a sporogenous cell (phialide) containing a shallow, rounded cup (collarette) at the distal end. Due to successive proliferation of the sporogenous cell, the conidia were pushed into the collarette and accumulated in a slimy mass. The second type appeared as a result of a budding process from sclerotic cells and at the ends of terminal and lateral branches. Schol-Schwarz (1970) suggested the second type were probably moniliform hyphae and did not ordinarily separate into individual conidia as Medlar (1915) suggested.

Medlar's original generic concept was revised by Beyma (1943), Schol-Schwarz (1970), and Cole and Kendrick (1973).

Although there is disagreement among the authors on several taxonomic details, the basic criteria for species within this genus includes the presence of single-celled, ovoid to curved conidia formed on flask-shaped phialides with a collarette (Schol-Schwarz, 1970 and Cole and Kendrick, 1973). In addition to the above criteria, Cole and Kendrick (1973) proposed that the genus Phialophora include only those fungi possessing an enteroblastic, phialidic ontogeny. However, Sivasithamparam (1973) listed reasons for which the enteroblastic ontogeny concept was not a reliable taxonomic criterion.

Several authors have discussed the instability of Phialophora as a genus and noted difficulties in distinguishing among species within the genus. Hughes (1951) expressed doubts about assigning a generic name to a species which produces only phialides as conidiogenous cells. Cole and Kendrick (1973) referred to Phialophora as a form genus since one cannot distinguish between anamorphs of species belonging to Pyrenomycetes and those of Discomycetes. Schol-Schwarz (1970) stated that delimitation of Phialophora species was difficult because of the extensive variability and morphological differentiation of the conidial states. As a consequence, she used the term 'species group' to delimit Phialophora fungi of similar form.

Several monographs on the genus Phialophora are available, but these deal primarily with wood-inhabiting

species (Beyma, 1943; Moreau, 1963; Schol-Schwarz, 1970; and Cole and Kendrick, 1973). The genus currently contains a number of vaguely defined species that are anamorphs of Ascomycetes in the Helotiales (e.g. Pyrenopeziza, Mollisia, Ascocoryne) and Sphaeriales (e.g. Coniochaeta and Gaeumannomyces) (Schol-Schwarz, 1970 and Walker, 1981). Species for which the teleomorph has not been found are considered dematiaceous hyphomycetes in the Deuteromycetes (Barron, 1972).

Fungi of the genus Phialophora are found in soils (especially forest soils); wood; wood pulp; water; food (apples, butter, margarine); the rhizosphere of various grasses; some herbaceous dicots; and on humans and animals as causal agents of chromoblastomycosis and other skin infections (Schol-Schwarz, 1970; Barron, 1972; and Walker, 1981).

Walker (1980 and 1981) reviewed the taxonomy of Phialophora species from the roots of Gramineae. The taxonomic criteria used for these fungi differ somewhat from Phialophora species found in other habitats. In addition to the taxonomic criteria used for other species, taxonomy of Phialophora species on grasses is based on mycelial growth rates, hyphopodial shape, and the shape and germinability of conidia (Walker, 1981 and Wong and Walker, 1975).

McKeen (1952) first isolated a Phialophora species from the roots of Gramineae (Zea mays L.) in Ontario, Canada. He stated that the fungus was 'quite aggressive, but not very

pathogenic' and noted the morphological similarity of this species with Ophiobolus graminis (Sacc.) Sacc. (now referred to as Gaeumannomyces graminis (Sacc.) Arx and Olivier). The fungus was described as Phialophora radicicola by Cain (1952). Lemaire and Ponchet (1963) used the name P. radicicola for the conidial state of O. graminis. The authors, however, did not report comparing their isolates with Cain's fungus.

Scott (1970) isolated a Phialophora species from grass roots in Britain and suggested it was similar but not identical to P. radicicola. Deacon (1974) described Scott's fungus and proposed the name P. radicicola var. graminicola Deacon, based on morphological differences between the fungus found by Scott and that described by Cain. In making this distinction, he automatically established Phialophora radicicola (Cain) var. radicicola to accomodate the fungus described by Cain (Int. Code Bot. Nomencl. Art. 26, Stafleu et al., 1972). But Deacon (1974) used the name P. radicicola var. radicicola for a fungus producing lobed hyphopodia on grass roots in Britain. Walker (1981) pointed out that use of this name by Deacon was a violation (according to Int. Code Bot. Nomencl. Art. 26, Stafleu et al, 1972) since the fungus described by Cain did not produce lobed hyphopodia. Walker (1981) subsequently referred to Deacon's P. radicicola var. radicicola as Phialophora sp. (lobed hyphopodia).

Walker (1980) reexamined the original collection of P.

radicicola deposited in the University of Toronto Herbarium (TRTC), a subculture from the living type culture in the Centraal bureau voor Schimmelcultures (CBS), and a culture of the original isolate from McKeen. Following a thorough examination of the material, Walker (1980) concluded that "neither the type culture nor the descriptions given by Cain (1952) and McKeen (1952) show characters necessary for accurate comparisons to be made between P. radicicola and recent Phialophora isolates from Gramineae". Thus, Walker (1980) recommended that the name P. radicicola Cain be rejected for any isolates of Phialophora spp. other than the type specimen and the type isolate. Walker (1980) transferred P. radicicola var. graminicola to P. graminicola so that it would not be confused with P. radicicola. The full description of P. graminicola is provided by Walker (1981). P. graminicola differs from P. radicicola, Phialophora sp. (lobed hyphopodia), and the conidial states of G. graminis varieties in that it has slower growth rates and in lacking curved, non germinating conidia (Wong and Walker, 1975 and Walker, 1981).

Hornby et al. (1977) described Gaeumannomyces cylindrosporus Hornby, Slope, Gutteridge, and Sivanesan from wheat roots inoculated with P. graminicola and noted that it was probably the teleomorph of this fungus.

Sivasithamparam (1975) isolated several other Phialophora species from the rhizosphere of wheat in Australia. Individual species included P. mutabilis (Beyma)

Schol-Schwarz, P. cyclaminis Beyma, P. fastigiata (Lagerb. and Melin) Conant, P. hoffmannii (Beyma) Schol-Schwarz, P. lignicola (Nannf.) Goidanich, P. malorum (Kidd and Beaumont) McColloch, P. verrucosa Medlar, and some isolates which could not be matched with any previously described species of Phialophora.

Deacon and Scott (1983) reported the occurrence of two types of Phialophora species on maize roots from France and South Africa. One type was described as Phialophora zeicola Deacon and Scott, the other remained unnamed. In addition to taxonomic features described for other Phialophora species, Deacon and Scott (1983) emphasized darkly pigmented, vesicle-like, swollen cells in roots as taxonomic features. Deacon and Scott (1983) claimed they could be used to distinguish between the two groups of Phialophora fungi (other references to these structures have been made by Scott, 1970; Deacon, 1973; Deacon, 1974; and Deacon, 1981). Walker (1980) noted the presence of these structures but stressed the need to examine a wide range of isolates on a number of hosts before considering them as reliable taxonomic feature.

Genus Magnaporthe

Krause and Webster (1972) established the genus Magnaporthe to accommodate M. salvinii (formerly Leptosphaeria salvinii Cattaneo) the causal agent of stem

rot of rice.

There is some disagreement among mycologists concerning the classification of Magnaporthe. Krause and Webster (1972) placed Magnaporthe in the Diaporthales based on the endothia-type ascus, the pseudoparenchymatous centrum, and the wide paraphyses-like bands which partially gelatinize at maturity. Barr (1977) classified Magnaporthe in the Phyllachorales (Physosporellaceae) based on the similarity of its anamorphic state to Beltraniella portoricensis (Stevens) Pirozynski and Patil and Buergenerula biseptata (Rostrup) Sydow. Each possesses sympodial, geniculate, indeterminate conidiogenous cells with blastic conidia borne on denticles. Barr (1977) also noted the formation of hyphopodia on rice culms by M. salvinii and their similarity to those of Buergenerula spartinae Kohlm. and Gessner. Finally, Barr (1977) suggested that Magnaporthe should be inserted in place of Monographella Petrak in her synopsis of the Physosporellaceae (Barr, 1976), based on the narrow elongate ascospores and globose-conic, scarcely clypeate perithecia found in Magnaporthe.

Arx (1979) cited evidence for using anamorphic characters as a basis for classifying Ascomycetes in preference to the traditional approach based on ascus morphology. He referred to difficulties in distinguishing between unitunicate and bitunicate asci and suggested there may be a number of species intermediate between the two. Arx (1979) assigned Magnaporthe to the Pleosporaceae because

of the similarity of its Nakataea anamorph to species of Pleospora with Stemphylium conidia; Cochliobolus with Curvularia or Bipolaris conidia; and Pyrenophora with Drechslera conidia. He also stated that the asci of Magnaporthe are bitunicate although they frequently do not function as such because the ascospores are not violently discharged, but are extruded in a slimy mass (as with fungi in the Diaporthaceae). If this classification system were adapted, M. salvinii, (Nakataea anamorph) would be classified under the Pleosporaceae (Dothideales) while M. rhizophila (Phialophora anamorph) would probably be placed in Diaporthaceae (Sphaeriales), even though their respective teleomorphs are very similar.

Monod (1983) reduced Magnaporthe to synonomy under the genus Phragmoporthe Petrak in the Diaporthaceae. The type species, Pragmoporthe conformis (Berk. and Br.) Petrak, contains 4-celled hyaline ascospores that appear similar to those of Gnomonia spp. Walker (personal communication, 1986) does not agree with reducing Magnaporthe to synonomy under Phragmoporthe because (1) the ascospores of Phragmoporthe do not show darker central cells but are uniformly hyaline and (2) Magnaporthe is a hyphopodiate genus parasitic on Gramineae whereas Phragmoporthe appears to be a nonhyphopodiate genus occurring on dead leaves and branches of Alnus spp. with no superficial mycelium. Although more detailed taxonomic work is required to determine the correct classification of Magnaporthe, Walker

(personal communication, 1986) regarded it as a distinct genus, parasitic on Gramineae, and a member of the Diaporthales.

There are currently three species in the genus Magnaporthe. Magnaporthe salvinii (Cattaneo) Krause and Webster (Syn.: Leptosphaeria salvinii Cattaneo = Phragmoporthe salvinii (Cattaneo) Monod) is the causal agent of stem rot of rice (Krause and Webster, 1972 and Tullis, 1933). Its anamorph is Nakataea sigmoidea Hara (Syn: Helminthosporium sigmoideum Cav. = Vakrabeeja sigmoidea (Cav.) Subramanian = Curvularia sigmoidea (Cav.) Hara). This fungus occurs in rice-growing areas of the United States, Europe, and Asia and is pathogenic on all varieties of rice as well as Zizaniopsis miliacea (Tullis, 1933 and Tsuda and Ueyama, 1978).

Magnaporthe grisea (Herbert) Barr (Bas.: Ceratosphaeria grisea (Herbert) Monod) is the teleomorph of Pyricularia grisea (Cooke) Sacc. (Syn.: Trichothecium griseum Cooke). P. grisea is a parasite of crabgrass (Digitaria sanguinalis L.) in North America and is morphologically indistinguishable from Pyricularia oryzae Cav., the causal agent of rice blast (Herbert, 1971). Apparently, the only characteristic that distinguishes P. oryzae from P. grisea is its pathogenicity to rice and host range (although some P. grisea isolates can infect rice and some P. oryzae isolates can infect wild grasses) (Asuyama, 1965 and Herbert, 1971). Herbert (1971) produced the perfect state

of P. grisea by mating isolates from crabgrass in all possible combinations on barley grains and rice straw partially embedded in Sach's agar. Attempts to mate isolates of P. oryzae (from rice) together or in combination with P. grisea failed. Ueyama and Tsuda (1975), however, reported the formation of perithecia similar to M. grisea from matings of P. oryzae and a species of Pyricularia from Eleusine. In addition to crabgrass, P. grisea has been found on St. Augustinegrass (Stenotaphrum secundatum (Walt.) Kuntze) (Malca and Owen, 1957) as well as other grasses (Sprague, 1950).

Magnaporthe rhizophila Scott and Deacon is an ectotrophic, root-inhabiting fungus with a Phialophora anamorph. It was isolated from wheat roots grown in soil from a millet field in South Africa (Scott and Deacon, 1983). Although the authors noted vascular discoloration in wheat seedlings inoculated with this fungus, it has not been implicated as a causal agent for any known disease. M. rhizophila conforms to the teleomorphic concept of Magnaporthe as defined by Krause and Webster (1972) although it differs from other species of Magnaporthe spp. in that it appears to be homothallic and produces a Phialophora anamorph (Scott and Deacon, 1983).

Genus Gaeumannomyces:

Saccardo (1875) described Rhaphidophora graminis Sacc.

from a rotted culm base of Cynodon or Agropyron in Italy. R. graminis was later transferred to the genus Ophiobolus and renamed Ophiobolus graminis (Sacc.) Sacc. by Saccardo in Roumeguere and Saccardo (1881).

Prillieux and Delacroix (1890) reported that O. graminis was the causal agent of Maladie du Pied (disease of foot) or what is now known as take-all disease of wheat. Cobb (1892) first reported the occurrence of take-all on oats in New South Wales. Saccardo (1916) described Ophiobolus oryzinus Sacc. from decaying rice culms in the Philippines. Tullis (1933) reported O. oryzinus was the causal agent of black sheath rot of rice (also called brown sheath rot) in the United States.

Prior to 1940, no distinction was made between isolates of O. graminis causing take-all of oats and those causing take-all of wheat. However, Turner (1940) suggested a distinct biological strain of O. graminis was responsible for take-all of oats in England. Because of its pathogenicity to oats and longer ascospores (85-140 um for the oat take-all fungus and 60-110 um for the wheat take-all fungus)., Turner (1940) proposed the name Ophiobolus graminis var. avenae for the strain of O. graminis affecting oats. In doing this she automatically established the variety O. graminis var. graminis for the nomenclatural type of O. graminis (Sacc.) Sacc. (Int. Code Bot. Nomencl. Art. 25, Stafleu et al., 1972).

Arx and Olivier (1952) examined perithecia, asci, and

ascospores of the wheat take-all fungus, O. graminis and found several features which suggested it did not belong in the genus Ophiobolus. Among the distinguishing features were (1) perithecia of Ophiobolus were sunk deep in the substratum whereas the take-all fungus had a perithecial neck which formed a papillate projection above the surface of the substatum; (2) the take-all fungus had unitunicate asci that exhibited a highly refractive ring; and (3) the perithecium of the take-all fungus had a centrum resembling fungi in the family Diaporthaceae (Arx and Olivier, 1952). Arx and Olivier (1952) rejected the classification of the take-all fungus as O. graminis and erected the genus Gaeumannomyces for this fungus.

Muller and Arx (1973) included Gaeumannomyces in the Pyrenomycetes based on the presence of an ascomata entirely surrounded by a peridial wall, unitunicate asci arranged in a hymenial layer, and an opening (ostiole) at the terminal portion of the ascomata that is covered by hyphae-like periphyses. Gaeumannomyces was placed in the order Sphaeriales by Muller and Arx (1973) and in the family Diaporthaceae. In this family, the apical pore of the ascus is surrounded by a refractive ring-like structure.

Walker (1972) examined the type specimens of Gaeumannomyces graminis (Sacc.) Arx and Olivier and of O. oryzinus and found they were identical. Both fungi produced mycelium with strongly lobed brown hyphopodia in association with the perithecia. Since Turner (1940) established the

variety O. graminis var. avenae (renamed Gaeumannomyces graminis var. avenae (Turner) Dennis in Dennis (1960)), the type specimen (nomenclatural type) of O. graminis automatically assumed the name Gaeumannomyces graminis var. graminis. Walker (1972) found that no lobed hyphopodia (as seen on the type specimen of G. graminis) occurred on any specimens of the wheat take-all fungus. Therefore, he concluded that the epithet var. graminis could not be applied to the wheat take-all fungus. However, perithecia, asci and ascospores of the wheat take-all fungus were indistinguishable from G. graminis var. graminis. Walker (1972) thus proposed a new variety G. graminis (Sacc.) Arx and Olivier var. tritici Walker for the wheat take-all fungus.

There are currently three varieties of G. graminis: G. graminis var. graminis, G. graminis var. avenae, and G. graminis var. tritici. Of the three varieties, G. graminis var. graminis is the least pathogenic on Gramineae. Brown sheath rot of rice rarely causes damage and some isolates of G. graminis var. graminis have been used as biological control agents against take-all of wheat (Walker, 1972 and Wong, 1981). G. graminis var. avenae and G. graminis var. tritici are more pathogenic than G. graminis var. graminis on Gramineae. While G. graminis var. tritici causes take-all of wheat and barley, G. graminis var. avenae can cause take-all on oats, wheat, barley and take-all patch on Agrostis turf (Walker, 1975). Hornby et al. (1977)

described G. cylindrosporus from rotting wheat and barley roots in test tubes inoculated with P. graminicola. The authors noted that single-ascospore isolates formed colonies resembling P. graminicola, however, they did not reproduce the teleomorph from single or combined ascospore isolates and concluded there was no direct proof the perithecia developed from P. graminicola. Jackson (personal communication, 1987) recently produced perithecia of G. cylindrosporus from single-ascospore cultures on sterile wheat stems in Sach's agar. Perithecia of G. cylindrosporus have been found in the field on annual bluegrass and perennial ryegrass (Jackson and Landschoot, 1984).

One species and two taxonomic groups of Gaeumannomyces have been found on species in the family Cyperaceae. Walker (1980) redescribed Gaeumannomyces caricis Walker from a collection found on Caricis paniculatae in Denmark. G. caricis was originally referred to as Gaeumannomyces sp. by Munk in Larson (1952), but later redescribed under the name Linocarpon eucrytum (Berk. and Br.) Petrak (Munk, 1957). Walker (1972) rejected the epithet 'eucryptum' and the generic differences between Linocarpon and Gaeumannomyces are discussed by Walker (1980).

Walker (1980) discussed the Gaeumannomyces spp. occurring on Cyperaceae and placed them in two groups based on ascospore size. The two groups are referred to as taxonomic sp. 1 and 3 (taxonomic sp. 2 is G. caricis).

MATERIALS AND METHODS

A. Survey of ectotrophic *Phialophora* species from turfgrasses.

Turf exhibiting symptoms of summer patch disease was surveyed for the presence of ectotrophic *Phialophora* species on the roots and crowns of affected plants. Several sites were surveyed in Rhode Island in 1985 and 1986 and in Maryland in 1987. In addition, plant pathologists from other locations in the U.S. were solicited for isolates of ectotrophic *Phialophora* species from turf showing patch disease symptoms. Finally, some isolates were obtained from turfgrass swards exhibiting an overall decline in quality, but not necessarily showing patch symptoms. Information relevant to each isolate was tabulated and included the fungus species, isolate designation, host, origin of isolate, and date of isolation.

Characteristic symptoms in diseased turf included circular areas of dying or dead plants with a tuft of apparently unaffected grass in the center (Fig 1.). Some patches were devoid of living grass plants, but often were colonized by broadleaf weeds (Fig. 2). Diseased tillers were initially yellow or reddish brown, becoming tan as the plant died. Roots and crowns of affected tillers appeared brown or black, often exhibiting pronounced vascular discoloration.

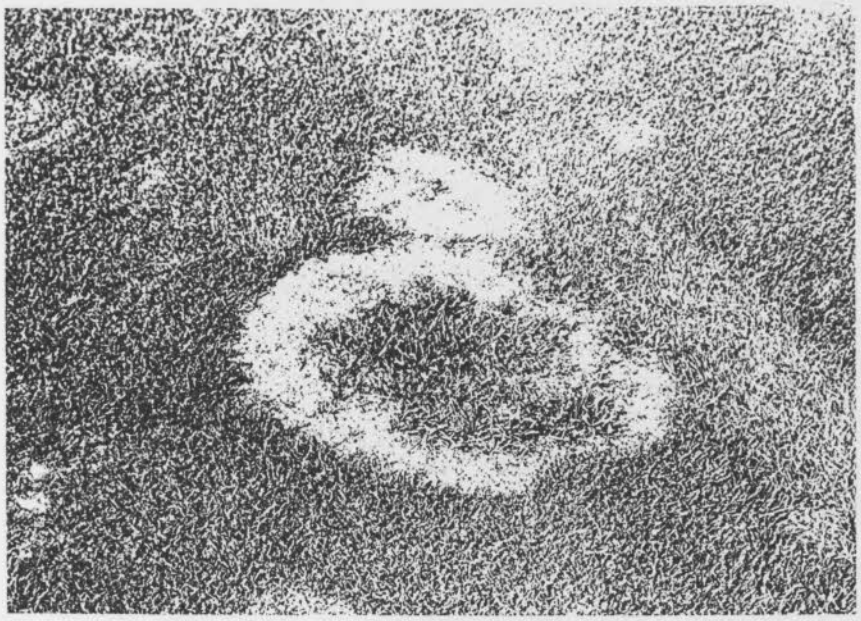


Fig. 1. Symptoms of summer patch disease on annual bluegrass fairway in Providence, RI.

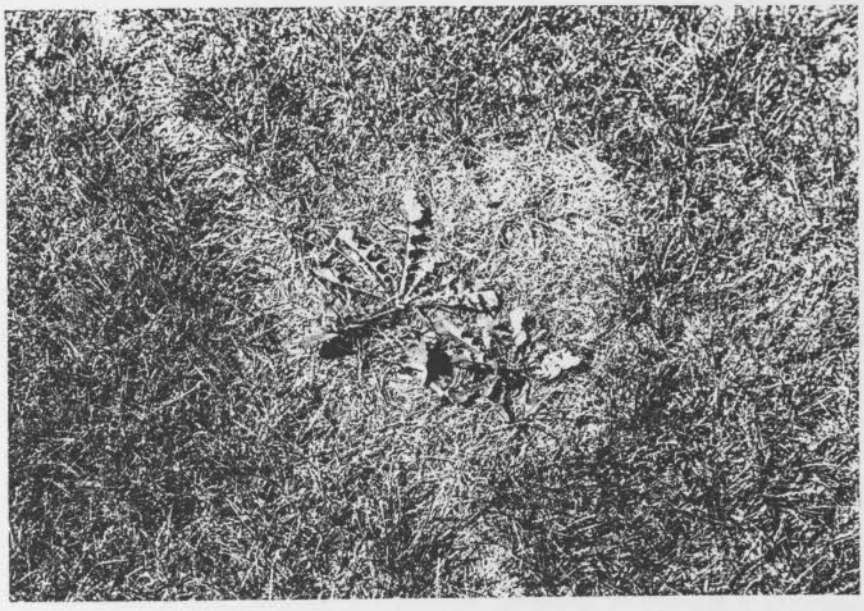


Fig. 2. Symptoms of summer patch disease on Kentucky bluegrass turf in Fairland, MD.

Small segments (3-5 mm) of roots or crowns from infected plants were removed for isolation of fungi. The root and crown segments were surface sterilized by immersion in 1% silver nitrate ($AgNO_3$) for 30-60 seconds, followed by a transfer to 5% sodium chloride (NaCl) for 10 seconds. They were then rinsed in sterile distilled water and blotted on a clean paper towel. The sterile segments were transferred to a Gaeumannomyces-selective medium (GSM) (Juhnke et al., 1984) or potato dextrose agar (PDA) (Appendix I).

Single-ascospore cultures were derived by immersing mature perithecia into 10% clorox for 20-30 seconds, followed by a rinse in sterile distilled water. The perithecia were then pipetted with 2-3 ml sterile distilled water onto PDA fortified with antibiotics (Appendix I). The perithecia were crushed with sterile forceps on the agar surface and the contents dispersed over the entire petri dish with a sterile glass rod. After 48-72 hours, individual germinating ascospores were transferred to PDA medium using a sterile, fine-tipped glass rod.

All cultures were maintained on 15 ml PDA in 100 ml, screw-capped medicine bottles at 5-10^{o}C and were transferred every 3-4 months to fresh PDA. Cultures used for temperature and/or pathogenicity studies were recycled at least once each year using a method described by Speakman (1982) in which wheat seeds (cv. redcoat) were surface sterilized with $AgNO_3$, placed on water agar (Appendix I),

and inoculated with agar discs containing the fungus. After the roots were colonized, the fungi were reisolated from root tissues.

B. Taxonomic studies.

1. Morphological characteristics of Phialophora anamorphs.

All isolates were cultured on sterile wheat seedlings growing in water agar (Speakman, 1982). Wheat was chosen as the host plant because of its fast germination, large roots, and its close similarity in reaction as a host for ectotrophic Phialophora, Gaeumannomyces, and Magnaporthe species. Vegetative structures of taxonomic significance that were examined (on the roots or in the agar itself) included phialospores, phialides, hyphopodia, runner hyphae, and sclerotium-like structures. Appropriate photographs and measurements were obtained. Phialospores, phialides, and sclerotium-like bodies have only been seen in axenic culture. Growth rates on 1/2 strength PDA of the different isolates at various temperatures were also determined for taxonomic comparisons.

2. Teleomorph morphology and development.

Perithecia of Magnaporthe poae sp. nov. were produced initially on wheat (cv. redcoat) growing in pots of quartz

sand during a 12 week pathogenicity study. Wheat seedlings growing in 8 cm plastic pots containing quartz sand were inoculated by placing 10-12 oat grains colonized by M. poae in the center of each pot. The plants were grown in the greenhouse for 10 weeks and then transferred to a growth chamber at 29°C with a 12 hour photoperiod. Mature perithecia were noticed on roots 2 weeks after the plants were placed in the growth chamber.

The perithecia of M. poae and Gaeumannomyces incrustans sp. nov., were produced in culture by placing 6 mm diameter plugs from colonies of opposing mating types on opposite sides of propylene oxide-sterilized wheat stems embedded in Sach's agar in petri dishes (Luttrell, 1958). The dishes were sealed with parafilm and incubated on a laboratory bench at room temperatures under alternating light and dark from a west facing window. Perithecia of M. poae developed in 6-8 weeks and perithecia of G. incrustans developed in 8-10 weeks.

Perithecia of G. cylindrosporus were found on partially decomposed root and stem tissue in the thatch of fairway turf. Attempts to produce perithecia from single-ascospore isolates under axenic conditions failed.

Photographs and measurements were made of teleomorphic structures of taxonomic significance (i.e. perithecia, asci, ascospores, etc.).

3. Sexuality of M. poae, M. rhizophila, and G. incrustans.

Mature perithecia of M. poae (ATCC 64413), M. rhizophila (PREM 45952) and G. incrustans (ATCC 64416 x ATCC 64417) were produced in culture using the procedures described in section B-2. Fifteen single-ascospore isolates were obtained from 3 perithecia from each of these species using the procedure described in section A. The 15 isolates from each species were paired in all possible combinations by placing 6 mm diameter plugs from colonies growing on PDA on opposite sides of wheat stems in Sach's agar as described in section B-2. All plates were examined for the presence or absence of mature perithecia 6-8 weeks after inoculation with M. poae and M. rhizophila and 8-10 weeks following the inoculation of G. incrustans.

4. Variation in ascospore size between M. poae and M. rhizophila.

Mature perithecia of M. poae and M. rhizophila were produced on wheat stems in Sach's agar as described in section B-2. Perithecia of M. poae used for this study were derived from selected matings (ATCC 64412 x ATCC 64411; ATCC 64412 x ATCC 64131; ATCC 64412 x MD-1; ATCC 64412 x ATCC 60239; and ATCC 64412 x NE-197). Perithecia of M. rhizophila were derived from isolate PREM 45952. All perithecia were collected 8 weeks following inoculation.

The mean, standard deviation, and range of ascospore length and width for 5 crosses of M. poae and the type culture of M. rhizophila were determined. Paired comparisons of sample means of 100 ascospores were made to determine if there was a significant difference in ascospore size between M. poae and M. rhizophila (Snedecor and Cochran, 1980).

C. Effects of temperature on radial growth.

The isolates of M. poae, G. incrustare, P. graminicola, and G. cylindrosporus used for temperature studies are listed in Table 10. All isolates were transferred from medicine bottles to 90 mm plastic petri dishes containing 1/2 PDA. The cultures were then incubated in darkness for 8 days at 25°C. Five mm agar plugs were cut from the actively growing margins of each colony and transferred to the center of 90 mm plastic petri dishes containing approximately 20 ml of 1/2 PDA after incubation. Four replicate plates were used for each isolate and the plates were incubated at 5°, 10°, 15°, 20°, 25°, 30°, 35°, and 40°C. The test was repeated once. Growth rates were determined by measuring radial growth every 24 hrs. for 8 days or until the colony margin reached the edge of the plate. Two measurements of colony radius were taken at right angles to each other on each plate and the values were averaged. Growth measurements used for temperature comparisons were from the 24 hr. period showing maximum growth for each isolate.

D. Pathogenicity studies.

1. Relative pathogenicity of ectotrophic *Phialophora* species from turgrasses.

The relative pathogenicity of *M. poae*, *G. incrustans*, *P. graminicola*, and *G. cylindrosporus* was evaluated on 8-week-old and 7-year-old 'Merion' Kentucky bluegrass. The fungus species and isolate designations are listed in Tables 11 and 12. Because some pathogenic fungi tend to become less virulent when maintained for long periods on artificial media, all isolates were passed through wheat seedlings one one month before the start of this experiment (Speakman, 1982). The recycled isolates were grown on 1/2 PDA for 10 days at 25°C, several 5 mm plugs of each isolate were then transferred from the margins of actively growing colonies to 125 ml erlenmyer flasks containing 1.5 g perennial ryegrass grains and 5 ml distilled water that had been autoclaved twice for 60 minutes. The flasks were incubated at 25°C for four weeks following inoculation.

Clay pots (10 cm in diameter) were filled with washed Terra-Green® calcined clay (nutrient status and pH are provided in Appendix I). Pots were seeded with 0.12 g 'Merion' Kentucky bluegrass and were placed in a mist house until seeds had germinated (approximately 2 weeks). The pots of seedling grass were then transferred to a greenhouse

for 8 weeks.

A 7.6 cm diameter cup cutter was used to remove plugs of 7-year-old 'Merion' Kentucky bluegrass from plots at the University of Rhode Island Turfgrass Research Farm. All plugs were cut to a depth of 3 cm, planted in 10 cm pots containing calcined clay, and placed in the greenhouse for 10 days prior to inoculation. Both the seedling and the 7-year-old Kentucky bluegrass swards were maintained at a height of 3.0 cm. All pots were watered daily with approximately 150 ml water and fertilized once every 2 weeks with a 20-10-20 fertilizer at 0.5 g/150 ml H_2O/pot. Pots of 8-week-old Kentucky bluegrass were inoculated with 1.0 gram of air-dried inoculum (colonized perennial ryegrass grains) by placing the inoculum at the surface in 2, 10 cm strips on two diameters at right angles. Inoculation of 7-year-old Kentucky bluegrass plugs was identical to 8-week-old Kentucky bluegrass seedling turf with the exception that the inoculum was placed in 1.0 cm slits in order to facilitate contact with the roots. Four replicate pots were inoculated with each isolate. Controls were seeded with 1.0 gram of uncolonized, autoclaved, air-dried perennial ryegrass grains.

Following inoculation, all pots of Kentucky bluegrass were immediately placed in a growth chamber at $28^{o}C$ +/- $2^{o}C$ with a 12 hour photoperiod (620 uE $m^{-2}s^{-1}$), and 60-100% relative humidity. Temperature and relative humidity were recorded with a hygrothermograph. Plants were watered daily

with approximately 150 ml and the medium was kept moist by placing water absorbant pads beneath the pots. All pots were fertilized once every 2 weeks with equal amounts of a 20-10-20 fertilizer (0.5 g/150 ml H_2O/pot). Weekly applications of resmethrin (applied as a mist in growth chambers) were necessary to control insects. The experimental design was a randomized design. The experiment was terminated after 6 weeks and was repeated once.

The criteria for evaluating pathogenicity included weekly quality ratings, clipping yields (dry weights), and estimates of root colonization/discoloration. Weekly quality ratings were based on a scale of 1 to 9, 1 = dead plants (straw-yellow color and no growth apparent) and 9 = dense, dark-green and actively growing turf. Clipping yields were expressed on a dry weight basis and served as a quanti tative, non-destructive measure of plant growth. Leaf growth above 3.0 cm height was harvested once each week, air-dried at 60°C, and weighed. Root colonization/ discoloration was assessed visually on a scale of 1-5: 1 = no colonization, 2 = colonization and light discoloration, 3 = colonization and moderate discoloration, 4 = colonization and moderately-severe discoloration, 5 = colonization and severe discoloration. All data were subjected to analysis of variance and means were compared using Duncan's Multiple Range Test (P = 0.05).

2. Host Range of *M. poae*.

One isolate of *M. poae* (ATCC 64413) was used to inoculate several species and cultivars of turfgrasses and cereals. The species and cultivars of turfgrasses used in this test are listed in Table 13. The cereal species and cultivars are listed in Tables 14 and 15. Isolate ATCC 64413 was recycled on Kentucky bluegrass roots one month prior to initiation of this study and inoculum was prepared as described in section D-1. Paired comparisons (t-test) of parameters used to assess pathogenicity were made between the inoculated and control plants for each species/cultivar (Snedecor and Cochran, 1980).

Clay pots (10 cm) were filled with calcined clay and seeded with turfgrasses at the following rates: velvet bentgrass (*Agrostis canina* L.) 0.07 g/pot; creeping bentgrass (*Agrostis palustris* Huds.) 0.07 g/pot; chewings fescue (*Festuca rubra* var. *commutata* Gaud.) 0.20 g/pot; tall fescue (*Festuca arundinacea* Schreb.) 0.20 g/pot; perennial ryegrass (*Lolium perenne* L.) 0.20 g/pot; and annual bluegrass 0.12 g/pot. All pots were placed in a mist house until seeds had germinated. The pots of seedling turfgrasses were then transferred to a greenhouse and maintained at a height of 3.0 cm. All turf was watered and fertilized as described in Section D-1. After 8 weeks all pots of turfgrasses were inoculated as described for 8-week-old Kentucky bluegrass and placed in growth chambers

under the same conditions as described in Section D. Four replicate pots of inoculated and control plants were included for each species/cultivar. The experimental design was a randomized design. The criteria for evaluating pathogenicity are described in section D-1.

Cereal seeds were surface sterilized with 10% clorox for 1-2 minutes and placed on moistened paper towels in glass dishes and allowed to germinate (24-48 hours). Five pregerminated seeds were placed over 1.0 g of inoculum in 10 cm pots filled with calcined clay and covered with an additional 2 cm of this medium. Controls were planted over 1.0 g sterile, air-dried perennial ryegrass grains. Following inoculation, all pots of turfgrasses were placed in growth chambers under the same conditions as described in Section D-1. Four replicate pots of inoculated and control plants were included for each species. The experimental design was a randomized design. All plants were harvested after 6 weeks and evaluated for the degree of pathogenicity. The criteria for evaluating pathogenicity included measuring the longest leaf on each plant, shoot dry weights, root dry weights, and degree of root colonization/ discoloration (using the same scale as described in Section D-1).

3. Inducement of patch symptoms on mature Kentucky bluegrass sod.

Mature (22-month-old), field grown Kentucky bluegrass

sod (50% Baron, 20% Ram I, 15% Majestic, and 15% Touchdown) was transplanted to a ground bed (9.1 m long by 0.4 m wide by 0.18 m deep) filled with a compost/sand (50/50) mix in a greenhouse. The mix was steam-sterilized and left fallow for 1 year before the sod was established. The sod was established on 23 May 1985 and inoculated on 10 June, 1985 with 3 isolates of M. poae (ATCC 64413, ATCC 56773, and MI-1); 3 isolates of G. incrustans (SAUG-1, CREST, and ATBM); 1 isolate of G. graminis var. avenae (LLGga); 1 isolate of G. cylindrosporus (SAK); 1 isolate of P. graminicola (REY) and a control. Inoculum was prepared as described in Section D-1, and consisted of 1.0 g placed at the soil-thatch interface (1.0-1.5 cm below the sod surface). The inocula for individual treatments were placed in the center of 43 X 43 cm plots. The experimental design was a randomized complete block with 3 replications per treatment. The sod was maintained at 4.5 cm height, watered approximately 3 times per week (more frequently in summer months), and fertilized every 2 weeks with a 20-10-20 fertilizer at 2.5 g/gal H_2O (1.4 g/m^2). Where symptoms occurred, efforts were made to reisolate the causal fungus. Isolation of fungi from the roots of grass plants in the control plots was also undertaken to determine the species present.

E. Histology

Fungal fruiting structures (perithecia) were fixed in formalin-aceto-alcohol (FAA) for 24 hours followed by a transfer to 70% ethanol for another 24 hours. The fixed perithecia were then dehydrated in a graded tertiary butyl alcohol (TBA) series (Appendix II). Following dehydration, the perithecia were placed in vials of pure TBA to which 20% by volume xylene was added. Chips of low-grade parafin were added to the TBA : xylene mixture (2/3 solvent : 1/3 parafin) and the open vial was placed in a 45°C oven for 24 hours followed by a transfer to fresh low-grade parafin for 24 hours at 60°C. Two more transfers were made to high-grade Paraplast® parafin at 60°C, the first for 24 hours, the second for 6 hours. Perithecia were embedded in fresh Paraplast in aluminum dishes on a hot plate and transferred to ice-water as soon as the Paraplast began to solidify.

Serial sections 8-10 um thick were obtained with a rotary microtome. Sections were placed on microscope slides containing a thin layer of Haupt's adhesive and flooded with 4% formalin. The slides were then placed on a warming plate at 45°C for 24 hours. Sections were rehydrated and stained for 2 hours in 1% safranin followed by dehydration and a counter stain for 30-60 seconds with 0.5% toluidine blue in 95% ethanol (Appendix II). Sections were then mounted in diaphane and allowed to dry for 2 days.

Root and crown tissues were boiled in 3% KOH for approximately 30 seconds, rinsed in distilled water and mounted on microscope slides in 0.05% analine blue in lactophenol. The tissues were examined under a compound microscope.

RESULTS AND DISCUSSION

A. Survey of ectotrophic *Phialophora* species from turfgrasses:

Three species of ectotrophic *Phialophora* species were found on the roots and crowns of turfgrasses during this survey. The fungus species, isolate designation, host plant, origin of isolate, and date of isolation are provided in Table 1. Of the species listed, one is an undescribed species in the genus *Magnaporthe* (*Magnaporthe poae* sp. nov.); another is an undescribed species in the genus *Gaeumannomyces* (*Gaeumannomyces incrustans* sp. nov.); and the third species is *Gaeumannomyces cylindrosporus* (now established as the teleomorph of *Phialophora graminicola*). Although *G. cylindrosporus* and *P. graminicola* are considered to be different forms of the same species, the form found in the field was designated by the name appropriate for that form. If an isolate was derived from ascospores of *G. cylindrosporus*, it is referred to as *G. cylindrosporus*, if it was isolated directly from the roots and/or crowns of turfgrasses, it is referred to as *P. graminicola*.

Magnaporthe poae was isolated from the roots and crowns of *P. pratensis* and *P. annua* in Rhode Island, New York, Maryland Virginia, Nebraska, Pennsylvania, Michigan, and Colorado. *Gaeumannomyces incrustans* was isolated from *P. pratensis*, *P. annua*, *Cynodon* sp., *Agrostis* sp., *Zoysia* sp.,

Table 1. Fungus species, isolate designation, host plant, origin of isolate, and date of isolation of ectotrophic Phialophora species from turfgrasses.

Fungus Species	Isolate Designation	Host	Origin	Date of Isolation
M. poae	ATCC 64131	Poa annua	Providence, RI	8/85
M. poae	WG-14	Poa annua	Providence, RI	8/85
M. poae	RICH-1	Poa annua	Warwick, RI	8/86
M. poae	ATCC 64413	Poa pratensis	Manorville, NY	9/83
M. poae	ATCC 56773	Poa pratensis	Farmingdale, NY	----
M. poae	NY-258	Poa pratensis	Cold Spr. Har., NY	9/85
M. poae	MD-1	Poa pratensis	Fairland, MD	8/86
M. poae	PAMD	Poa pratensis	Glendale, MD	7/86
M. poae	MYS-1	Poa pratensis	Fairland, MD	7/87
M. poae	MYS-2	Poa pratensis	Fairland, MD	7/87
M. poae	D-1	Poa pratensis	Fairland, MD	7/87
M. poae	D-2	Poa pratensis	Fairland, MD	7/87
M. poae	ARL-1	Poa pratensis	Arlington, VA	7/87
M. poae	ARL-2	Poa pratensis	Arlington, VA	7/87
M. poae	NE-179	Poa pratensis	------- NE	9/84
M. poae	WF-868	Poa pratensis	Philadelphia, PA	----
M. poae	MI-1	Poa annua	------- MI	8/84
M. poae	CO-191	--------	------- CO	----
M. poae	ATCC 60239	Poa pratensis	Oyster Bay, NY	/84
G. incrustans	ATCC 64416	Poa annua	Locust Valley, NY	8/86
G. incrustans	ATCC 64417	Cynodon sp.	Witchita, KA	10/85
G. incrustans	BERM-MD	Cynodon sp.	-------, MD	5/86
G. incrustans	ATCC 64419	Poa pratensis	Kingston, RI	8/85
G. incrustans	SAUG-1	Poa pratensis	Kingston, RI	8/84
G. incrustans	CREST	Agrostis sp.	Rehoboth, MA	4/85

Table 1. Continued.

Fungus Species	Isolate Designation	Host	Origin	Date of Isolation
G. incrustans	ATBM	Cynodon sp.	Atlanta, GA	6/84
G. incrustans	S-1	Poa pratensis	Kingston, RI	7/86
G. incrustans	S-2	Poa pratensis	Kingston, RI	7/86
G. incrustans	S-3	Poa pratensis	Kingston, RI	7/86
G. incrustans	S-4	Poa pratensis	Kingston, RI	7/86
G. incrustans	S-5	Poa pratensis	Kingston, RI	7/86
G. incrustans	S-6	Poa pratensis	Kingston, RI	7/86
G. incrustans	S-7	Poa pratensis	Kingston, RI	7/86
G. incrustans	S-8	Poa pratensis	Kingston, RI	7/86
G. incrustans	S-9	Poa pratensis	Kingston, RI	7/86
G. incrustans	S-10	Poa pratensis	Kingston, RI	7/86
G. incrustans	ZOY-5	Zoysia sp.	------ IL	9/87
G. incrustase	ZOY-7	Zoysia sp.	------ IL	9/87
G. incrustans	PL-FES-B	Festuca rubra	Fairland, MD	7/87
G. cylindrosporus	SAK	Poa annua	L. Compton, RI	6/84
G. cylindrosporus	ATCC 64420	Poa pratensis	Kingston, RI	10/85
P. graminicola	REY	Poa pratensis	N. Kingstown, RI	6/84
P. graminicola	SIGN	Poa pratensis	Exeter, RI	7/85
P. graminicola	ATCC 64414	Poa pratensis	Narragansett, RI	6/85
P. graminicola	ATCC 64415	Lolium perenne	Kingston, RI	6/85
P. graminicola	ROSSI	Poa pratensis	Warwick, RI	6/85
P. graminicola	BG	Poa pratensis	Kingston, RI	7/85
P. graminicola	FCONN	Festuca sp.	------ CT	9/85
P. graminicola	LOB	Poa pratensis	Kingston, RI	9/85

and F. rubra. Isolates of this fungus were found in Rhode Island, New York, Maryland, Massachusetts, Kansas, Illinois, and Georgia. Perithecia of G. cylindrosporus were found on partially decomposed root and stem tissue of P. annua and P. pratensis in Rhode Island. Isolates of P. graminicola have been obtained from P. pratensis, L. perenne, and Festuca sp. in Rhode Island and Connecticut. The identities of G. cylindrosporus and P. graminicola were confirmed by Mr. John Walker of the Biological and Chemical Research Institute in Rydalmere, Australia and Dr. James Deacon of the Dept. of Microbiology, University of Edinburgh, U.K., respectively.

Magnaporthe poae was isolated most frequently from areas where summer patch was active (i.e. symptoms were progressing from smaller to larger sized patches). Gaeumannomyces incrustans, G. cylindrosporus, and P. graminicola were isolated most frequently from patches that were in various stages of recovery from summer patch disease. Some isolates of these fungi were obtained from turfgrass swards exhibiting a decline in quality but not necessarily showing patch symptoms, while some isolates were from turfgrasses exhibiting symptoms of other patch diseases (i.e. spring dead spot, necrotic ring spot, and yellow patch).

B. Taxonomic studies.

The taxonomic features of M. poae, G. incrustans, G. cylindrosporus, and P. graminicola are provided below. M. poae and G. incrustans are described as new species while features of G. cylindrosporus and P. graminicola are compared with those listed by Hornby et al. (1977), Deacon (1973), Walker (1980), and Wong (1980). The results of the sexuality studies for M. poae, M. rhizophila, and G. incrustans are included in this section, as is a comparison of ascospore length and width between M. poae and M. rhizophila.

1. Magnaporthe poae sp. nov. (Fig. 3-10)

Perithecia gregarious or single, immersed, sometimes superficial, black, body globose, 252-556 um diameter, neck cylindrical, 357-756 um long and 95-179 um at widest point. Perithecial wall up to 47 um thick, composed of several layers of brown, radially compressed isodiametric cells. External cells epidermoidal. Neck canal up to 40 um diameter, lined with hyaline upwardly pointed periphyses in canal. Asci numerous, clavate, 63-108 um long and 7-15 um at widest point, cylindrical, short stalked, straight or slightly curved, 8-spored, unitunicate, apex tapering but rounded, apical pore with non-amyloid refractive ring. Ascospores fusoid, 23-42 um long and 4-6 um diameter, 3

Fig. 3. Longitudinal section of a mature perithecium of *M. poae*. X 200

Fig. 4. Longitudinal section of perithecium wall of *M. poae*, ascospores in cavity. X 380

Fig. 5. Perithecia of *M. poae* immersed in wheat root. X 25

Fig. 6. *M. poae* ascus with refractive ring at apex. X 800

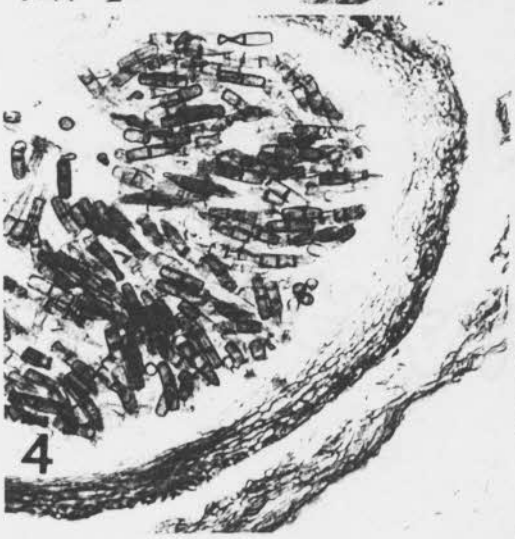
Fig. 7. *M. poae* ascospores showing darkly pigmented inner cells, one germinating from hyaline end cell. X 1360

Fig. 8. Phialide and phialoconidia of *M. poae*. X 1295

Fig. 9. Globose hyphopodia of *M. poae* with an infection peg (arrow) on Kentucky bluegrass roots. X 1000

Fig. 10. Clusters of swollen hyphal cells of *M. poae*, some of which function as hyphopodia, on Kentucky bluegrass stems. X 225


3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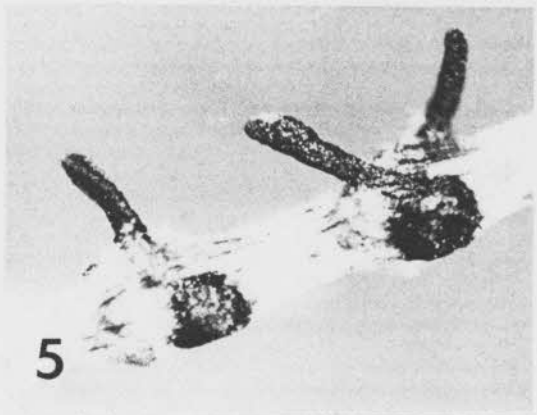
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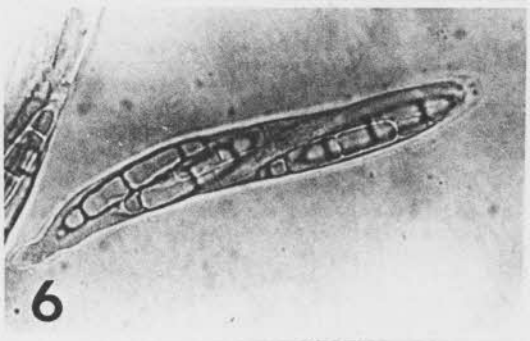
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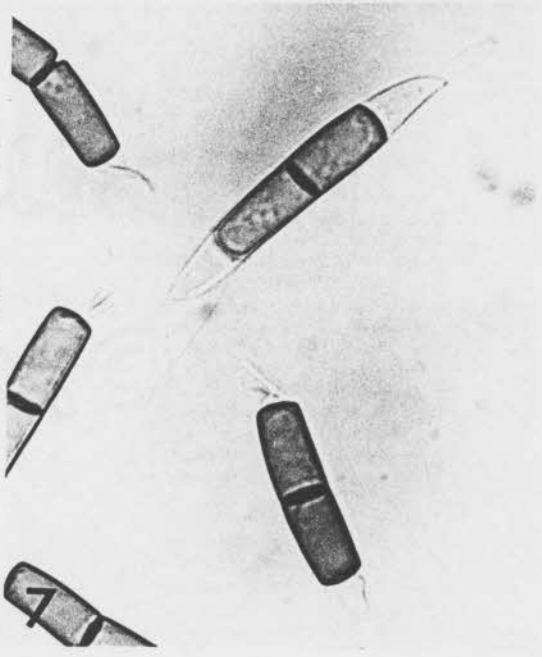
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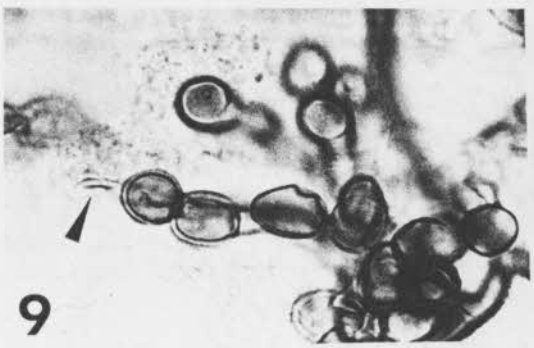

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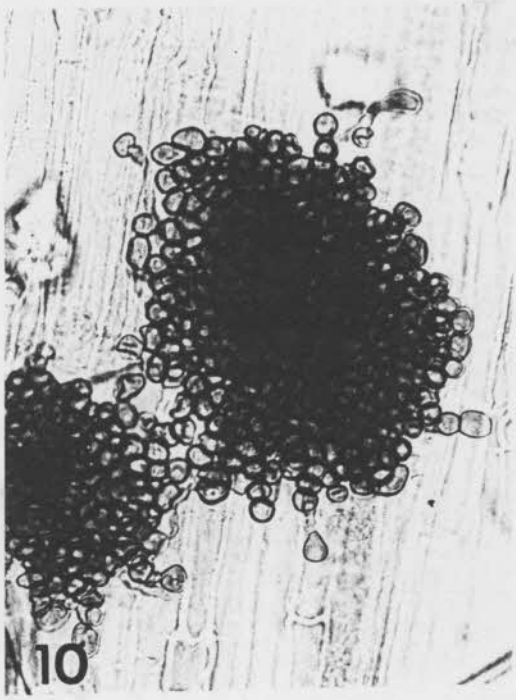


10

septate at maturity, end cells hyaline, intermediate cells thick walled and germinating from one or both hyaline cells. Paraphyses arising from hymenium between asci, hyaline, septate sometimes branched, 64-112 um long, 5-12 um diameter near base tapering to 2-3 um at tip. Superficial hyphae on the host roots sparse, brown, septate, 2-5 um diameter, often with septa delimiting the lateral branches, single or in strands of 2-3. Infection hyphae hyaline, slender, permeating root tissue. Swollen hyphal cells often fill cortical cells. Hyphopodia globose, occurring singly or in aggregates on stem bases or on roots, 6-12 um in diameter.

Cultures derived from single-ascospores maintained on half-strength PDA attained a diameter of 76 mm in 6 days at 28-30 ^{o}C. Mycelium appressed, initially hyaline, then turning gray or olivaceous brown with dark, thick strands of mycelium radiating from the center of the colony. Leading hyphae wavy, curling back towards center of colony. Older colonies (7-8 weeks) appear olivaceous brown or black.

Phialides hyaline, 6-15 um long, 2-5 um diameter, straight or curved, tapering at tip, some with collarette, collarette not visible in most, born on lateral branches or terminally, occurring singly or in clusters. Phialoconidia hyaline, 3-8 um long and 1-3 um wide, most slightly curved, some straight, commonly rounded at both ends.

The holotype, DAR 59044 (live culture ATCC 64413), was obtained from septic, necrotic roots on wheat plants, artificially inoculated with cultures designated incorrectly

as P. graminicola by R.W. Smiley and M. Craven Fowler and originally isolated from Kentucky bluegrass turf in Manorville, N.Y.

Morphological features of M. poae are very similar to those of M. rhizophila Scott and Deacon (1983); however, M. poae differs from M. rhizophila in (1) sexuality and (2) ascospore size. Experiments were performed to confirm these differences; the results are provided in the following two sections.

Sexuality of M. poae and M. rhizophila.

Results of mating compatibility for 15 single-ascospore isolates of M. poae (DAR 59044) and M. rhizophila (PREM 45952) are provided in Table 2 and Table 3, respectively. Isolates of M. poae segregated into two mating types designated A and a. When any isolate of type A was mated with type a, mature perithecia were produced in 4-6 weeks. In cases where two isolates of the same mating type were paired, no perithecia were produced. Isolates of M. rhizophila produced mature perithecia regardless of which isolates were paired. Thus, it was concluded that M. poae is a heterothallic species, while M. rhizophila is homothallic. M. rhizophila is the only homothallic species in the genus Magnaporthe.

When paired with single-ascospore isolates of both mating types (ATCC 64412 (a) and ATCC 64411 (A)), 19 field

Table 2. Compatibility (+ = formation of mature perithecia, 0 = absence of perithecia) and mating type (A or a) of 15 single-ascospore isolates of Magnaporthe poae paired in all possible combinations.

Mating type		a							A							
	Isolate	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15
	15	+	+	+	+	+	+	+	0	0	0	0	0	0	0	0
	14	+	+	+	+	+	+	+	0	0	0	0	0	0	0	
	13	+	+	+	+	+	+	+	0	0	0	0	0	0		
A	12	+	+	+	+	+	+	+	0	0	0	0	0			
	11	+	+	+	+	+	+	+	0	0	0	0				
	10	+	+	+	+	+	+	+	0	0	0					
	9	+	+	+	+	+	+	+	0	0						
	8	+	+	+	+	+	+	+	0							
	7	0	0	0	0	0	0	0								
	6	0	0	0	0	0	0									
	5	0	0	0	0	0										
a	4	0	0	0	0											
	3	0	0	0												
	2	0	0													
	1	0														

Table 3. Compatibility (+ = formation of mature perithecia, 0 = absence of perithecia) of 15 single-ascospore isolates of Magnaporthe rhizophila (PREM 45952) paired in all possible combinations.

Isolate	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15
15	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
14	+	+	+	+	+	+	+	+	+	+	+	+	+	+	
13	+	+	+	+	+	+	+	+	+	+	+	+	+		
12	+	+	+	+	+	+	+	+	+	+	+	+			
11	+	+	+	+	+	+	+	+	+	+	+				
10	+	+	+	+	+	+	+	+	+	+					
9	+	+	+	+	+	+	+	+	+						
8	+	+	+	+	+	+	+	+							
7	+	+	+	+	+	+	+								
6	+	+	+	+	+	+									
5	+	+	+	+	+										
4	+	+	+	+											
3	+	+	+												
2	+	+													
1	+														

Table 4. Compatibility (+ = formation of mature perithecia, 0 = absence of perithecia) and mating type (A or a) of field isolates of Magnaporthe poae paired with 2 single-ascospore isolates, ATCC 64412 (a) and ATCC 64411 (A).

Isolate	Origin (State)	Host	Paired with isolate: ATCC 64412 (a)	ATCC 64411 (A)	Mating Type
ATCC 64131	RI	Poa annua	+	0	A
WG-14	RI	Poa annua	+	0	A
RICH-1	RI	Poa annua	+	0	A
ATCC 64413	NY	Poa pratensis	+	0	A
ATCC 60239	NY	Poa pratensis	+	0	A
ATCC 56773	NY	Poa pratensis	+	0	A
NY-258	NY	Poa pratensis	+	0	A
MD-1	NY	Poa pratensis	+	0	A
PAMD	MD	Poa annua	+	0	A
MYS-1	MD	Poa pratensis	0	+	a
MYS-2	MD	Poa pratensis	+	0	A
D-1	MD	Poa pratensis	+	0	A
D-2	MD	Poa pratensis	+	0	A
ARL-1	VA	Poa pratensis	0	+	a
ARL-2	VA	Poa pratensis	0	+	a
NE-179	NE	Poa pratensis	+	0	A
WF-868	PA	Poa annua	0	+	a
MI-1	MI	Poa annua	+	0	A
CO-191	CO	-----	+	0	A

isolates of M. poae were also segregated as either mating type A or a. The sexual compatibility and mating type for the 19 field isolates of M. poae are listed in Table 4.

Variation in ascospore size between M. poae and M. rhizophila.

The mean, standard deviation, and range of ascospore length and width for 5 crosses within M. poae and one isolate of M. rhizophila are listed in Table 5. Based on paired comparisons of sample means of 100 ascospores, a highly significant difference ($P = 0.01$) in ascospore size was found between M. poae and M. rhizophila. Ascospores derived from 5 crosses of M. poae were significantly longer and narrower than those of M. rhizophila. No significant differences occurred between any 2 isolates of M. poae.

Discussion: Taxonomy of M. poae.

Based on perithecium, ascus, and ascospore morphology, M. poae fits well into Krause and Webster's concept of the Magnaporthe genus. As with all other members of this genus, M. poae produces hyphopodia and is parasitic on species within Gramineae. M. poae and M. rhizophila are the only known Magnaporthe species with Phialophora conidial states.

There is no consensus concerning the correct taxonomic position (order) for the genus Magnaporthe (Scott and

Table 5. Mean and range of ascospore length and width for five crosses of M. poae compared with an isolate of M. rhizophila (PREM 45952).

Isolate	Ascospore measurements			
	Mean[1]		Range	
	Length	Width	Length	Width
	um			
ATCC 64412 x ATCC 64411	30 ± 3	5 ± 0.5	37-23	6-4
ATCC 64412 x ATCC 64131	31 ± 2	5 ± 0.5	37-26	6-4
ATCC 64412 x MD-1	30 ± 2	5 ± 0.7	36-23	6-4
ATCC 64412 x ATCC 60239	31 ± 3	5 ± 0.7	42-24	6-4
ATCC 64412 x NE-197	30 ± 3	5 ± 0.5	36-23	6-4
PREM 45952	26 ± 3	6 ± 0.3	31-22	8-5

[1]Means and ranges based on measurements of 100 ascospores from at least 5 different perithecia for each isolate.

Deacon, 1983). Krause and Webster (1972) and Monod (1983) placed the genus in the Diaporthales, Barr (1977) considered it a member of the Phyllachorales, and von Arx (1979) has suggested it be classified in the Pleosporales.

Monod (1983) recently reduced Magnaporthe to synonomy with Phragmoporthe Petrak. Walker (personal communication, 1986), disputes this arrangement because (1) the ascospores of Phragmoporthe do not show darkly pigmented central cells but are uniformly hyaline and (2) Magnaporthe is a hyphopodiate genus with all known species parasitic on Gramineae. Phragmoporthe appears to be a non-hyphopodiate genus with no superficial mycelium and inhabits dead leaves and branches of Alnus sp.

2. Gaeumannomyces incrustans sp. nov. (Fig. 11-20)

Perithecia gregarious or single, mostly immersed, some superficial, black, body globose, 179-420 um diameter, neck cylindrical 252-672 um long and 84-147 um at widest point. Perithecial wall 20-38 um thick, composed of brown, radially compressed isodiametric cells. External cells epidermoidal. Neck canal up to 40 um diameter containing upwardly pointed, hyaline periphyses. Asci clavate, cylindrical, short-stalked, straight or slightly curved, 8-spored, unitunicate, 65-107 um long and 6-11 um diameter. Apex tapering, rounded, containing apical pore with non-amyloid refractive ring. Ascospores fusoid, 3-5 septa, hyaline,

Fig. 11. Longitudinal section of mature perithecium of *G. incrustans*. X 130

Fig. 12. Longitudinal section of perithecium wall of of *G. incrustans*. X 440

Fig. 13. *G. incrustans* perithecia on wheat roots. X 20

Fig. 14. *G. incrustans* ascus with refractive ring at apex. X 600

Fig. 15. Germinating ascospore of *G. incrustans*. X 900

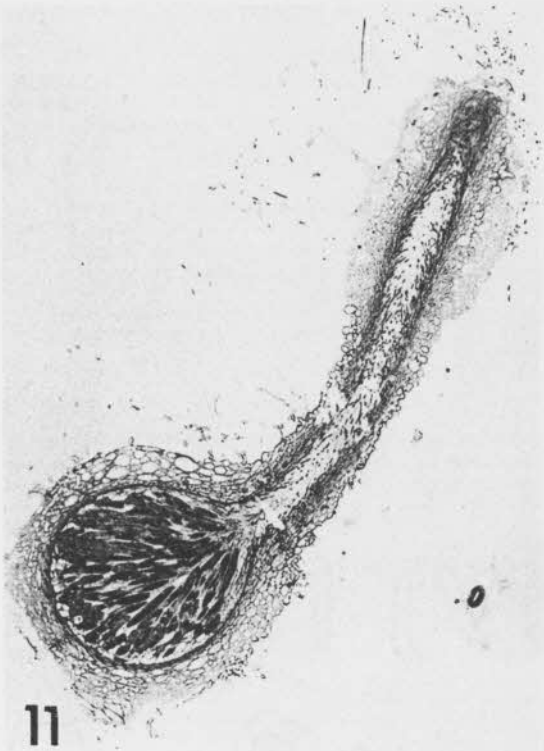
Fig. 16. Ascospore of *G. incrustans*. X 930

Fig. 17. Phialides and phialoconidia of *G. incrustans*. X 1100

Fig. 18. Hyphopodia of *G. incrustans* on Kentucky bluegrass root. X 930

Fig. 19. Crusts of *G. incrustans* mycelium on wheat stem surface. X 7

Fig. 20. Ectotrophic hyphae of *G. incrustans* on wheat root surface. X 370



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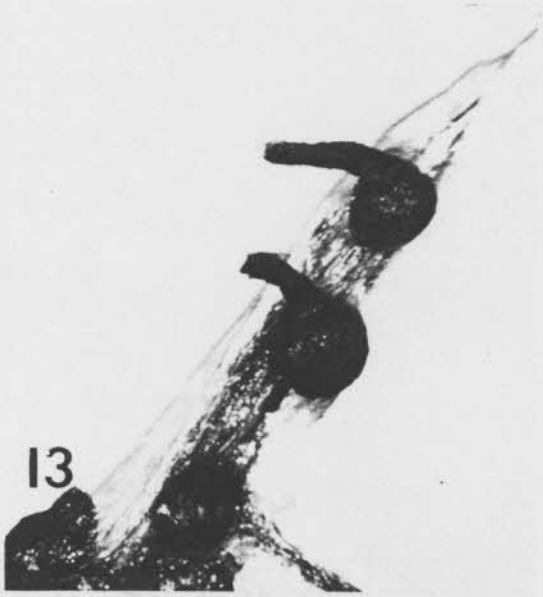

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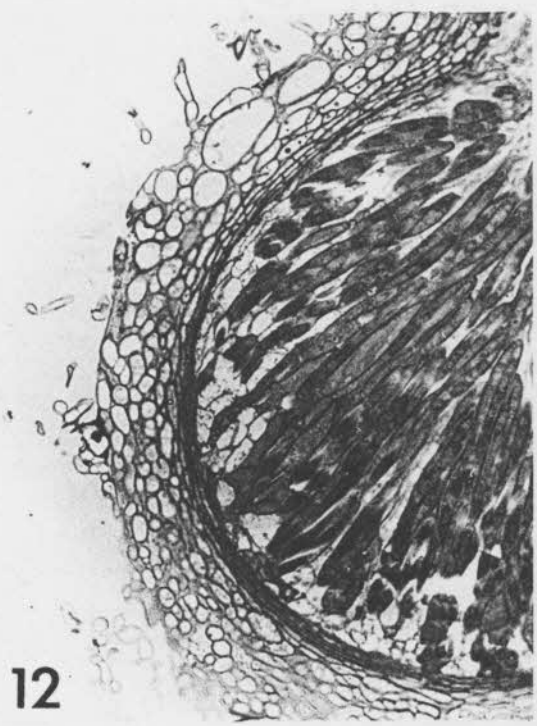

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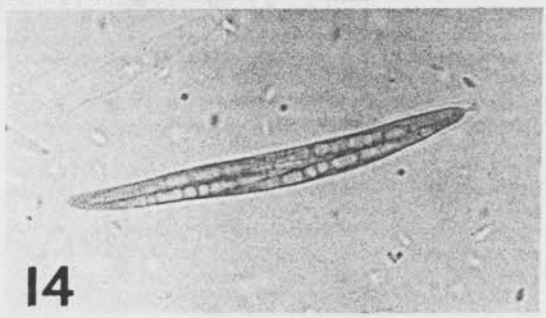

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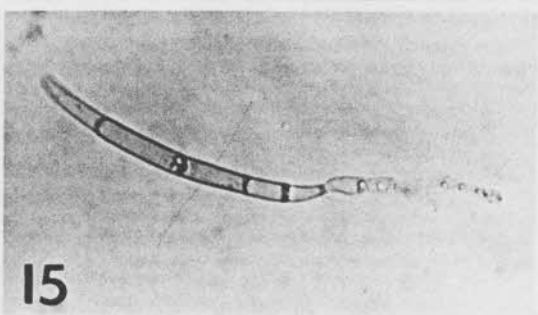

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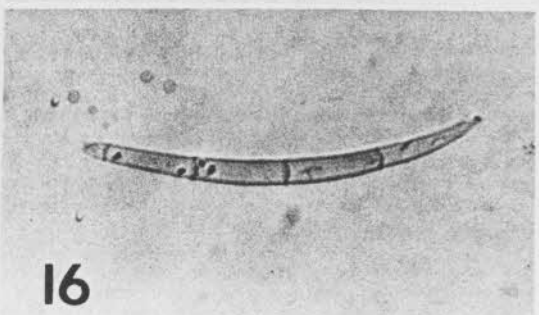

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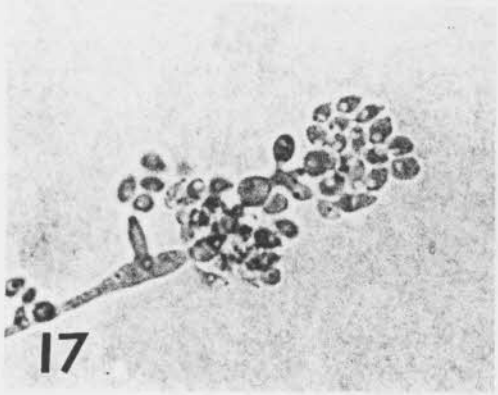

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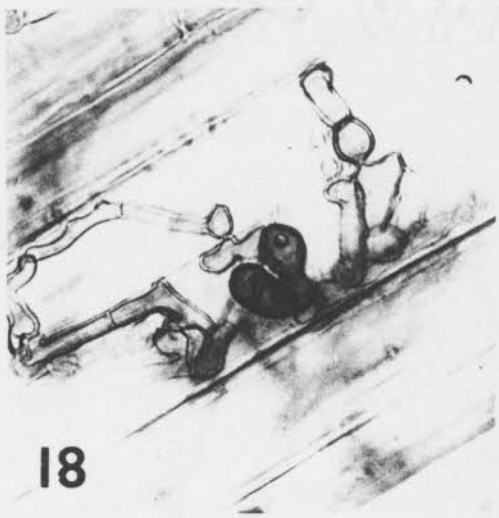

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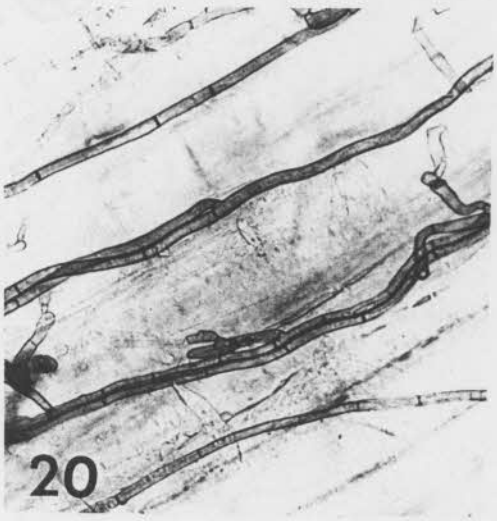


20

33-53 um long and 2-4 um diameter. Paraphyses arising from hymenial layer between asci, hyaline, septate, branching, 2-5 um diameter. Ectotrophic hyphae on host roots brown, septate, branched, 2-5 um diameter. Swollen hyphal cells fill cortical cells of roots and occur in large aggregates or as bulbils on wheat root surfaces in axenic culture. Infection hyphae hyaline, slender, permeating root tissue. Hyphopodia sparce, slightly lobed, occurring singly, in groups, or intercalary on root surfaces, 6-14 um in length, 4-6 um in diameter.

Single-ascospore colonies maintained on 1/2 PDA attained a diameter of 70 mm in 5 days at 28-30°C. Mycelium appressed, hyaline at first then turning gray or olivaceous black, with crusts of black mycelium appearing on the surface of older colonies, small aggregates of dark-colored cells often embedded in the medium. Leading hyphae wavy, curling back towards the center of the colony.

Phialides hyaline, 9-15 um long, 3-5 um diameter, slightly curved or straight, tapering at tip, collarette not visible in most, born on lateral branches or terminally, occurring singly or in clusters. Phialoconidia hyaline, 3-6 um long and 2-3 um diameter, round to oblong, straight or slightly curved, rounded at both ends, or pointed at one end and rounded at the other.

The holotype, DAR 59042, (live cultures ATCC 64418 and ATCC 64416) was obtained from necrotic wheat plants artificially inoculated with isolates ATCC 64418 from P.

pratensis roots in Kingston, Rhode Island and ATCC 64416 from P. annua roots in Locust Valley, New York. This fungus invests the root surface with darkly pigmented, robust, ectotrophic mycelium (Fig. 20). Extensive crust-like proliferations of mycelium form on the roots and stems of sterile wheat seedlings growing on water agar when inoculated with G. incrustans (Fig. 19). Since these structures are not formed by other ectotrophic, root-infecting fungi, they represent a distinguishing feature of this fungus, hence the epithet, 'incrustans'.

Sexuality of G. incrustans.

Results of mating compatibility for the 15 single-ascospore isolates of G. incrustans are provided in Table 6. Isolates segregated into two mating types designated A and a. When any isolate of A was paired with an isolate desigated a, mature perithecia were formed in 8-10 weeks. When two isolates of the same mating type were paired, no perithecia were produced. Thus, it was concluded that G. incrustans is a heterothallic species.

When paired with isolates of opposing mating types (ATCC 64416 (a) and ATCC 64417 (A)), 20 field isolates of G. incrustans segregated into either mating type A or a. Results of pairing 20 field isolates with known mating types of G. incrustans are listed in Table 7.

Table 6. Compatibility (+ = formation of mature perithecia, 0 = absence of perithecia) and mating type (A or a) of 15 single-ascospore isolates of Gaeumannomyces incrustans paired in all possible combinations.

Mating type		A				a									
	Isolate	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15
	15	+	+	+	+	0	0	0	0	0	0	0	0	0	0	0
	14	+	+	+	+	0	0	0	0	0	0	0	0	0	0	
	13	+	+	+	+	0	0	0	0	0	0	0	0	0		
	12	+	+	+	+	0	0	0	0	0	0	0	0			
	11	+	+	+	+	0	0	0	0	0	0	0				
a	10	+	+	+	+	0	0	0	0	0	0					
	9	+	+	+	+	0	0	0	0	0						
	8	+	+	+	+	0	0	0	0							
	7	+	+	+	+	0	0	0								
	6	+	+	+	+	0	0									
	5	+	+	+	+	0										
	4	0	0	0	0											
	3	0	0	0												
A	2	0	0													
	1	0														

Table 7. Compatibility (+ = formation of mature perithecia, 0 = absence of perithecia) and mating type (A or a) of field isolates of Gaeumannomyces incrustans paired with 2 isolates of opposing mating types, ATCC 64416 (a) and ATCC 64417 (A).

			Paired with Isolate:		
Isolate	Origin (State)	Host	ATCC 64416 (a)	ATCC 64417 (A)	Mating Type
ATCC 64416	NY	Poa annua	0	+	a
ATCC 64417	KA	Cynodon sp.	+	0	A
BERM-MD	MD	Cynodon sp.	0	+	a
ATCC 64418	RI	Poa pratensis	+	0	A
SAUG-1	RI	Poa pratensis	+	0	A
CREST	MA	Agrostis sp.	+	0	A
ATBM	GA	Cynodon sp.	0	+	a
S-1	RI	Poa pratensis	0	+	a
S-2	RI	Poa pratensis	0	+	a
S-3	RI	Poa pratensis	0	+	a
S-4	RI	Poa pratensis	0	+	a
S-5	RI	Poa pratensis	0	+	a
S-6	RI	Poa pratensis	0	+	a
S-7	RI	Poa pratensis	0	+	a
S-8	RI	Poa pratensis	0	+	a
S-9	RI	Poa pratensis	+	0	A
S-10	RI	Poa pratensis	+	0	A
ZOY-5	IL	Zoysia sp.	+	0	A
ZOY-7	IL	Zoysia sp.	+	0	A
PL-FES-B	MD	Festuca rubra	+	0	A

Discussion: Taxonomy of G. incrustans.

Arx and Olivier (1952) established the genus Gaeumannomyces to accommodate the take-all fungus, formerly designated Ophiobolus graminis (Sacc.) Sacc. In their description, the authors noted various features of the teleomorph, including the thin-walled ascus with a refractive ring in the apical wall and the vermiculate, hyaline ascospores. Walker (1972) noted the importance of superficial mycelium and hyphopodia in distinguishing among varieties of Gaeumannomyces graminis. Since mycelium and hyphopodia were not mentioned in von Arx and Olivier's original generic description, Walker (1980) emended the description to include production of hyphopodia on superficial mycelium. Gaeumannomyces incrustans fits well into Arx and Olivier's generic description as emended by Walker. Gaeumannomyces incrustans is the first heterothallic species reported for the genus Gaeumannomyces.

3. G. cylindrosporus and P. graminicola.

Teleomorphic and anamorphic characteristics of G. cylindrosporus and P. graminicola are presented in Tables 8 and 9, respectively. Teleomorphic characteristics of G. cylindrosporus isolates from Rhode Island were similar to those reported by Hornby et al., 1977. While the size range

Table 8. Comparison of morphological characteristics of a Rhode Island isolate of G. cylindrosporus (SAK), with those reported by Hornby et al. (1977)

	Perithecia			Asci		Ascospores		
Specimen	Body (diam.)	Neck L	Neck W	L	W	L	W	Septa
	um							
G. cylindrosporus (SAK)	380-880	40-140	---	82-112	11-17	29-52	4-7	2-6
G. cylindrosporus (sensu Hornby et. al.)	280-565	‹242	48-116	65-135	9-16	37-69	3-6	3-8

Table 9. Comparison of the morphological characteristics of *Phialophora graminicola* from turfgrasses in the United States compared with those reported by Deacon (1973), Walker (1980), and Wong (1980) and with the anamorph of *Gaeumannomyces cylindrosporus*.

	Radial Growth			Phialides		Phialoconidia		Hyphopodia			Culture Characteristics	
Isolate	20°C	25°C	30°C	L	W	L	W	Slightly Lobed	L	W	Aerial Mycelium	Color
	---mm/24 hr--			---------- um ----------					---- um ----			
ATCC 64414	4.1	5.3	4.4	8-20	2-4	3-8	2-3	+	10-20	7-14	+	gray
ATCC 64415	3.0	4.1	3.5	7-25	2-3	4-8	1-2	+	8-22	6-15	+	tan
SIGN	3.0	3.4	3.3	----	---	---	---	+	9-28	5-13	+	tan
REY	2.6	4.1	2.5	8-13	2-4	3-7	1-3	+	10-20	7-15	+	gray
ROSSI	2.9	4.2	4.0	8-15	2-4	5-9	1-3	+	12-24	10-15	+	tan
BG	3.3	3.3	2.8	9-25	2-4	3-7	2-4	+	10-24	10-19	+	gray
FCONN	3.3	2.6	1.6	7-20	2-3	3-8	1-3	+	7-18	5-14	+	gray
LOB	2.9	3.0	2.6	6-14	3-5	4-10	1-5	+	8-21	7-16	+	gray
P. graminicola sensu Deacon, Walker, and Wong.	2.8 1.5 2.3	4.5 2.3 4.3	2.0 2.6 1.5	7-18	2-3	2.5-7	2-3.5	+	9-18	6-15	+	pale gray, creamy white, or greenish-gray.
G. cylindrosporus (SAK)	2.9	4.3	3.4	8-23	2-4	4-7	1-3	+	11-19	7-12	+	gray

of perithecia, asci, and ascospore measurements varied among specimens, all measurements overlapped to some extent. Although growth rates varied among isolates, most anamorphic characteristics of P. graminicola conformed to those reported by various authors (Deacon, 1973; Walker, 1980; Walker, 1981; and Wong, 1980). No differences in anamorphic characteristics were detected between isolates of G. cylindrosporus and P. graminicola, thus confirming previous reports by Hornby et al. (1977); Walker (1980); and Walker (1981).

Smiley and Craven Fowler (1984) described North American isolates of P. graminicola as appressed colonies, initially colorless to light gray, darkening from the colony origin to an olive brown with growth rates of 4.7 mm/day at temperatures of 20-25 °C. Other taxonomic features included curling of hyphae at colony margins, some germinating phialoconidia, and subglobose hyphopodia. Smiley et al. (1985b) found considerable differences in temperature optima and osmotic potential tolerences between North American isolates of P. graminicola and those from Europe and Australia. The authors noted that North American isolates of P. graminicola may have different adaptions to edaphic conditions than those from other continents.

Isolates solicited from Smiley and associates and designated P. graminicola (ATCC 64413, ATCC 56773, NY-258, NE-179, and ATCC 60239) were all identified as M. poae in this study (Table 4). Statements made by Smiley and

associates concerning the taxonomic differences between North American isolates and isolates from other continents therefore should be disregarded.

C. Effects of temperature on radial growth.

Radial growth rates of 5 isolates of M. poae, 2 isolates of G. incrustans, 2 isolates of P. graminicola, and 1 isolate of G. cylindrosporus are listed in Table 10. Comparisons of selected isolates are provided in Fig. 21-23.

Growth rates varied among species and in some cases, among isolates within the same species. Optimum growth for M. poae isolates ATCC 64413, ATCC 64411, and ATCC 60239 occurred at 30 °C while optimum growth for ATCC 64412 and ATCC 64131 occurred at 35 °C. Growth of all M. poae isolates ceased at 10 °C and 40 °C. Optimum growth for isolate ATCC 64418 of G. incrustans occurred at 25 °C, while optimum growth of the other isolate (Crest) was at 30 °C. Neither isolate grew at 10 °C or 40 °C. Optimum growth of both isolates of P. graminicola (ATCC 64414 and ATCC 64415) and G. cylindrosporus (ATCC 64420) occurred at 25 °C. In contrast to M. poae and G. incrustans, P. graminicola and G. cylindrosporus grew at 10 °C but not at 35 °C (or above).

Both isolates of P. graminicola used in this study had slightly faster growth rates than isolates of P. graminicola from the United Kingdom and Australia (Wong, 1980). The

Table 10. Mean radial growth (mm/24 hrs.) of 10 isolates of ectotrophic *Phialophora* species at 7 temperatures.

Species	Isolate	Mean Radial Growth (mm/24 hrs.)						
		10°C	15°C	20°C	25°C	30°C	35°C	40°C
M. poae	ATCC 64413	0	2.3	4.8	7.3	8.8	6.8	0
M. poae	ATCC 64412	0	0.8	4.9	7.1	7.1	7.9	0
M. poae	ATCC 64411	0	2.9	4.9	7.5	7.9	7.5	0
M. poae	ATCC 60239	0	1.8	4.3	7.1	7.5	6.6	0
M. poae	ATCC 64131	0	0	3.1	6.6	6.6	7.8	0
G. incrustans	ATCC 64418	0	1.8	4.4	8.4	7.8	5.1	0
G. incrustans	CREST	0	1.4	5.1	6.4	5.1	3.8	0
P. graminicola	ATCC 64414	1.1	2.5	4.1	5.3	3.8	0	0
P. graminicola	ATCC 64415	1.4	2.1	3.0	4.1	0	0	0
G. cylindrosporus	ATCC 64420	1.9	2.0	2.3	4.1	0	0	0

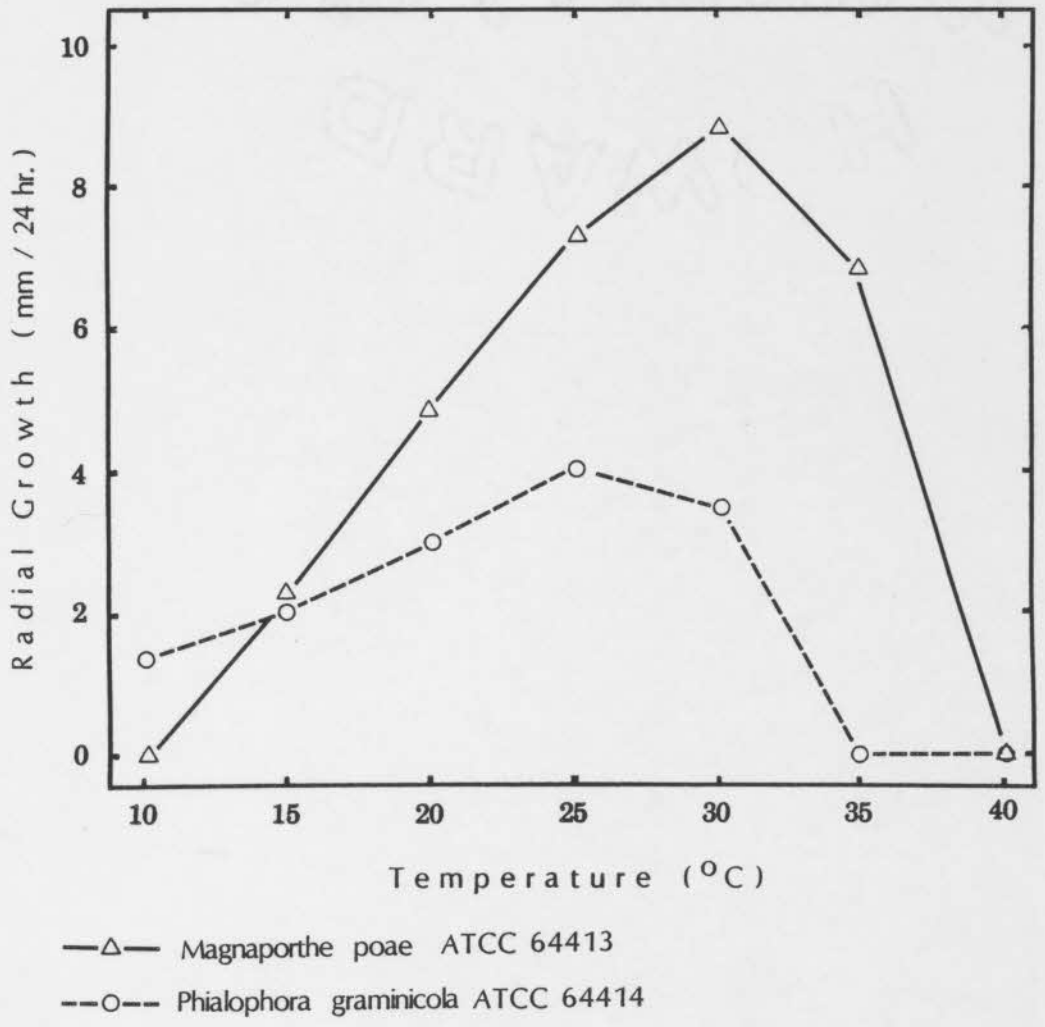


Fig. 21. Effect of temperature on radial growth (mm/24 hrs.) of M. poae (ATCC 64413) and P. graminicola (ATCC 64418).

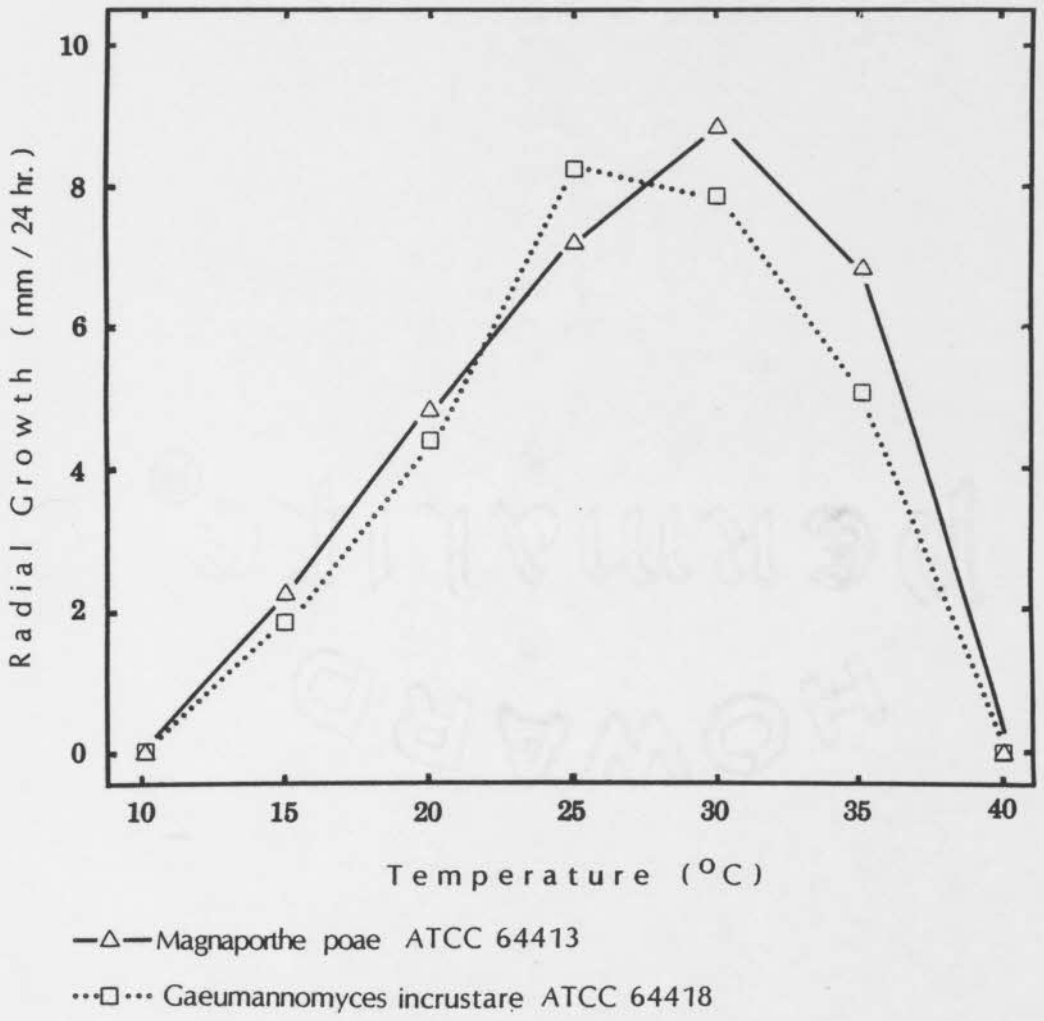


Fig. 22. Effect of temperature on radial growth (mm/24 hrs.) of M. poae (ATCC 64420) and G. incrustans (ATCC 64418).

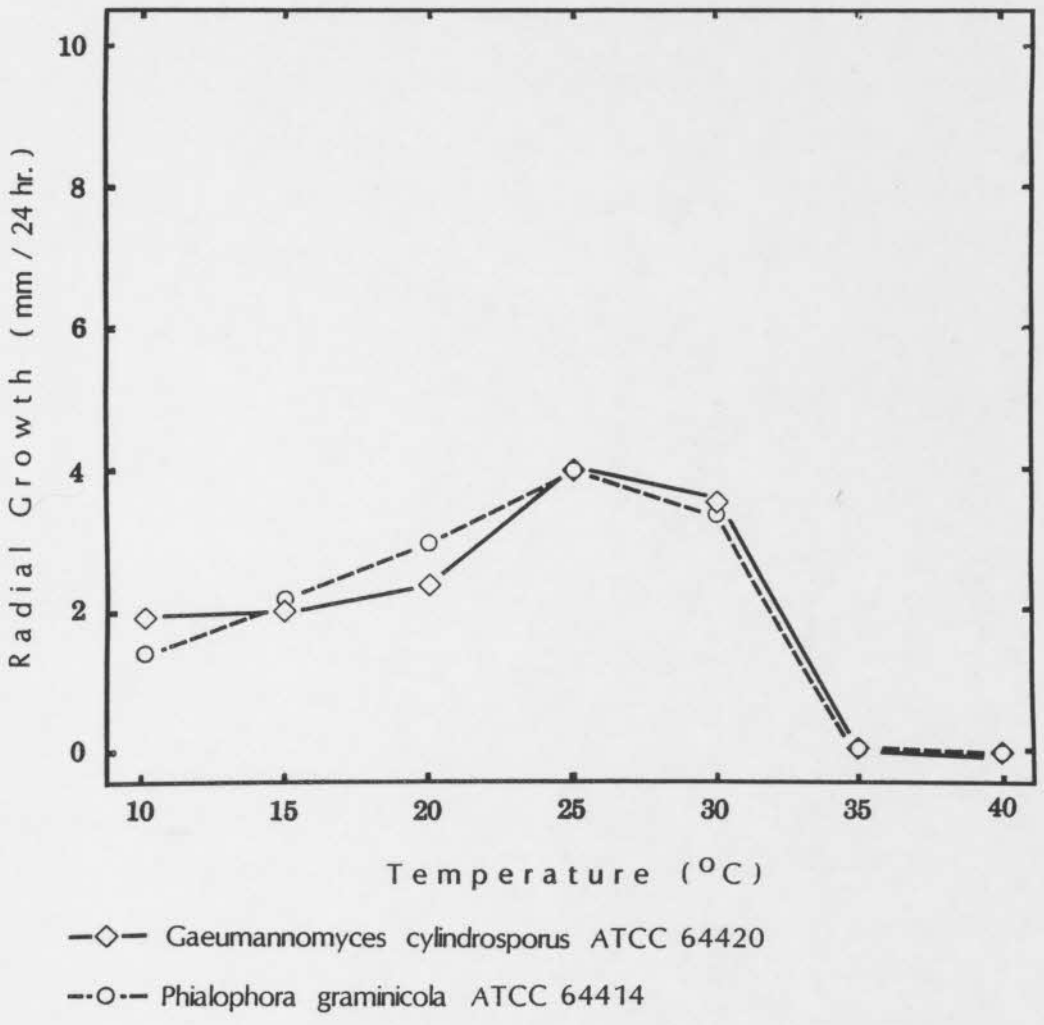


Fig. 23. Effect of temperature on radial growth of G. cylindrosporus (ATCC 64420) and P. graminicola (ATCC 64414).

slightly faster growth rates may have been due to the weaker (1/4 strength) concentration of PDA used by Wong. Although Smiley et al. (1985b) reported that American isolates of P. graminicola had considerably faster growth rates than European and Australian isolates at higher temperatures; it is now apparent that the American isolates were misidentified and were actually M. poae.

D. Pathogenicity:

1. Relative pathogenicity of ectotrophic Phialophora species from turfgrasses.

Quality ratings of 8-week-old and 7-year-old 'Merion' Kentucky bluegrass turfs inoculated with M. poae, G. incrustans, G. cylindrosporus, and P. graminicola are shown in Fig. 21-31 and Tables 1-4 in Appendix III. Clipping yields and estimates of root colonization/discoloration are provided in Tables 11 and 12.

Magnaporthe poae was the most pathogenic of the three species tested on both 8-week-old and 7-year-old Kentucky bluegrass. A significant decline in quality of 8-week-old Kentucky bluegrass was apparent after 2-3 weeks at 28°C with all isolates of M. poae when compared with controls. A similar trend was found on 7-year-old Kentucky bluegrass, however, significantly lower quality ratings did not occur until the the third or fourth week of the test. Clipping

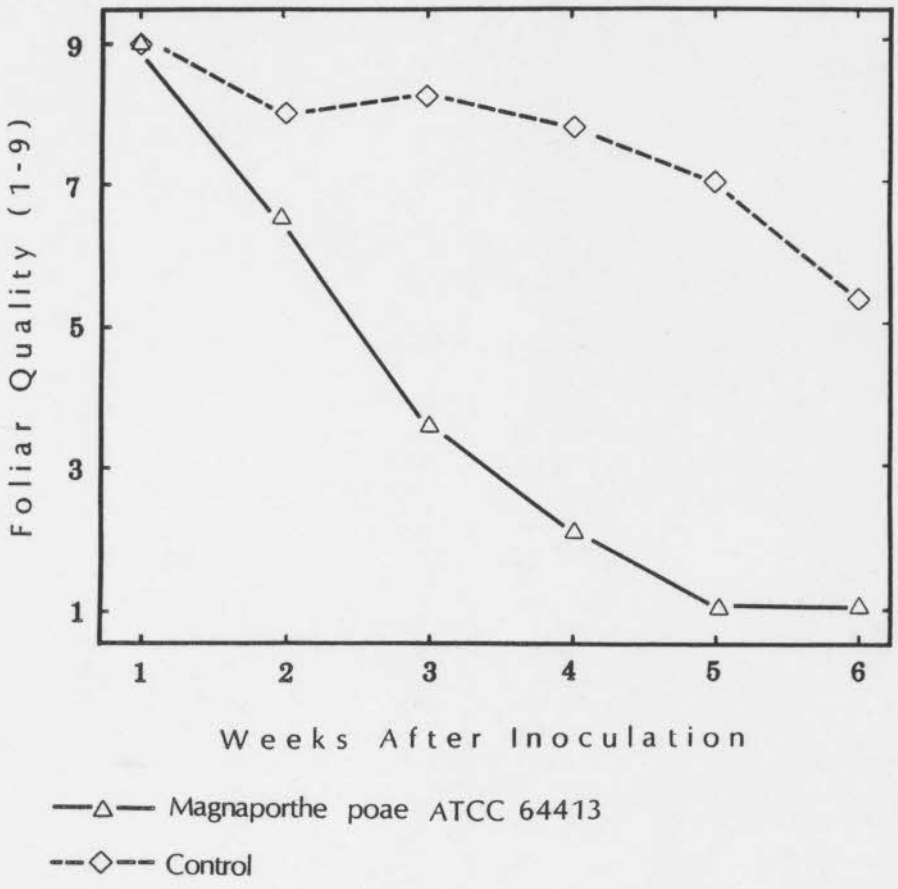


Fig. 24. Comparison of foliar quality between 8-week-old 'Merion' Kentucky bluegrass inoculated with M. poae (ATCC 64413) and a control.

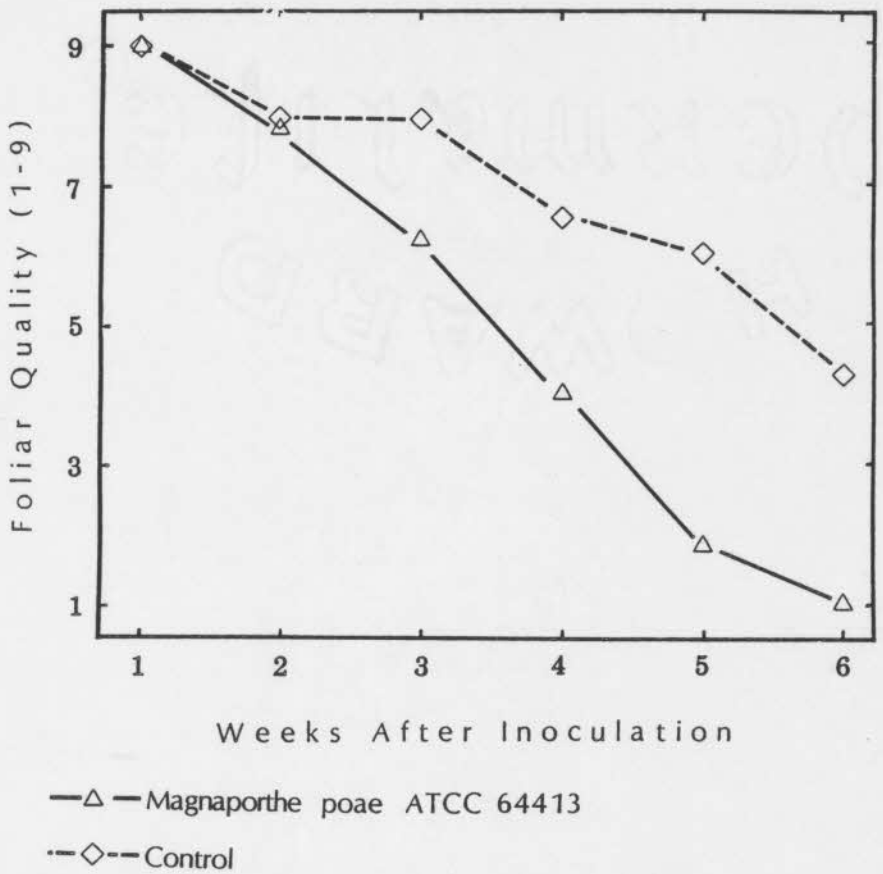


Fig. 25. Comparison of foliar quality between 'Merion' Kentucky bluegrass inoculated with M. poae (ATCC 64413) and a control.

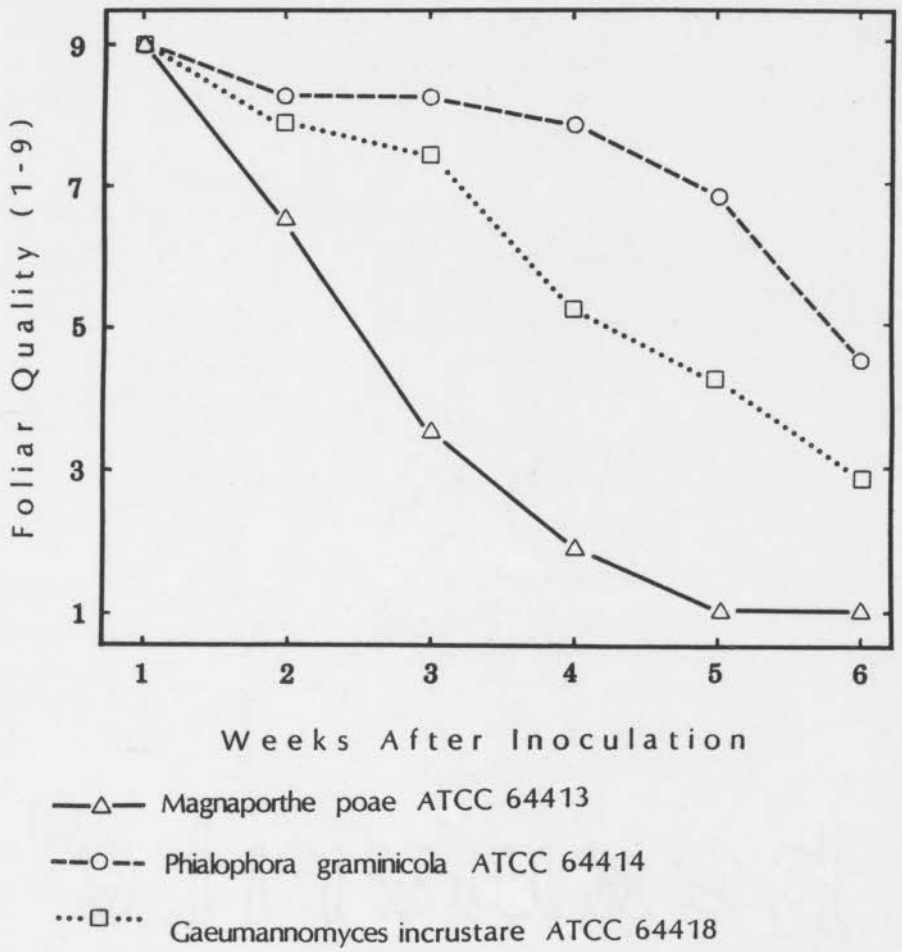


Fig. 26. Comparison of foliar quality between 8-week-old 'Merion' Kentucky bluegrass inoculated with M. poae (ATCC 64413), P. graminicola (ATCC 64414), and G. incrustans (ATCC 64418).

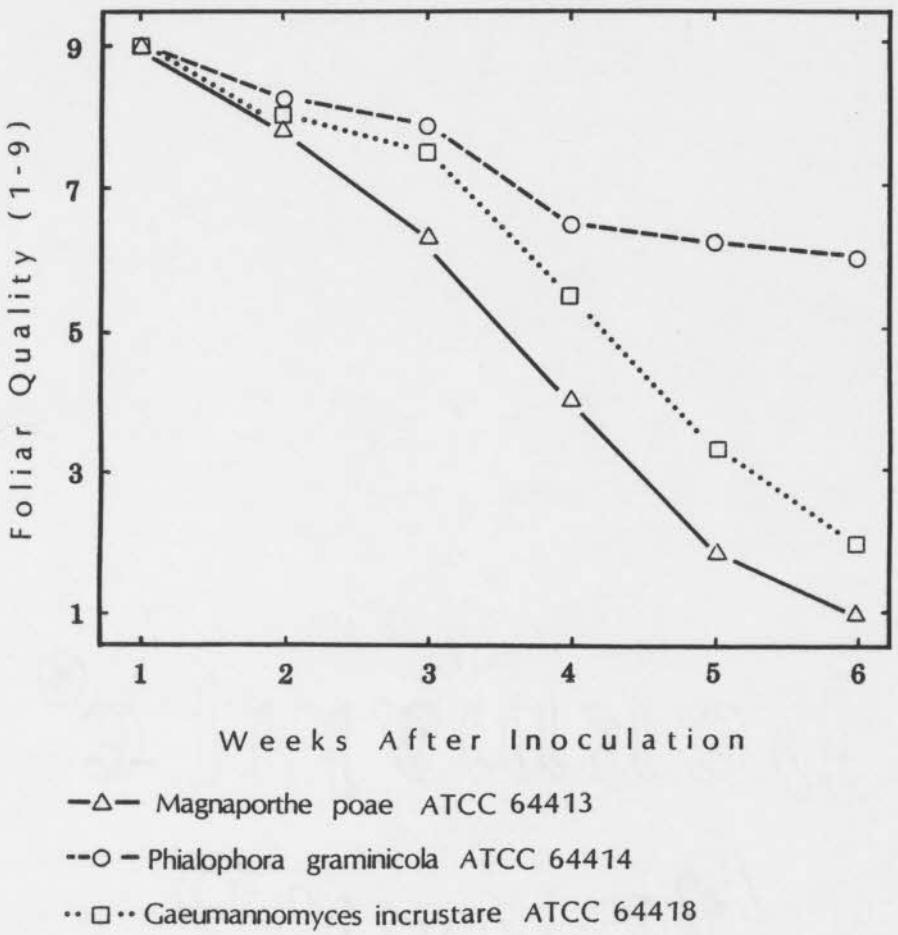


Fig. 27. Comparison of foliar quality between 7-year-old 'Merion' Kentucky bluegrass inoculated with M. poae (ATCC 64413), P. graminicola (ATCC 64414), and G. incrustans (ATCC 64418).

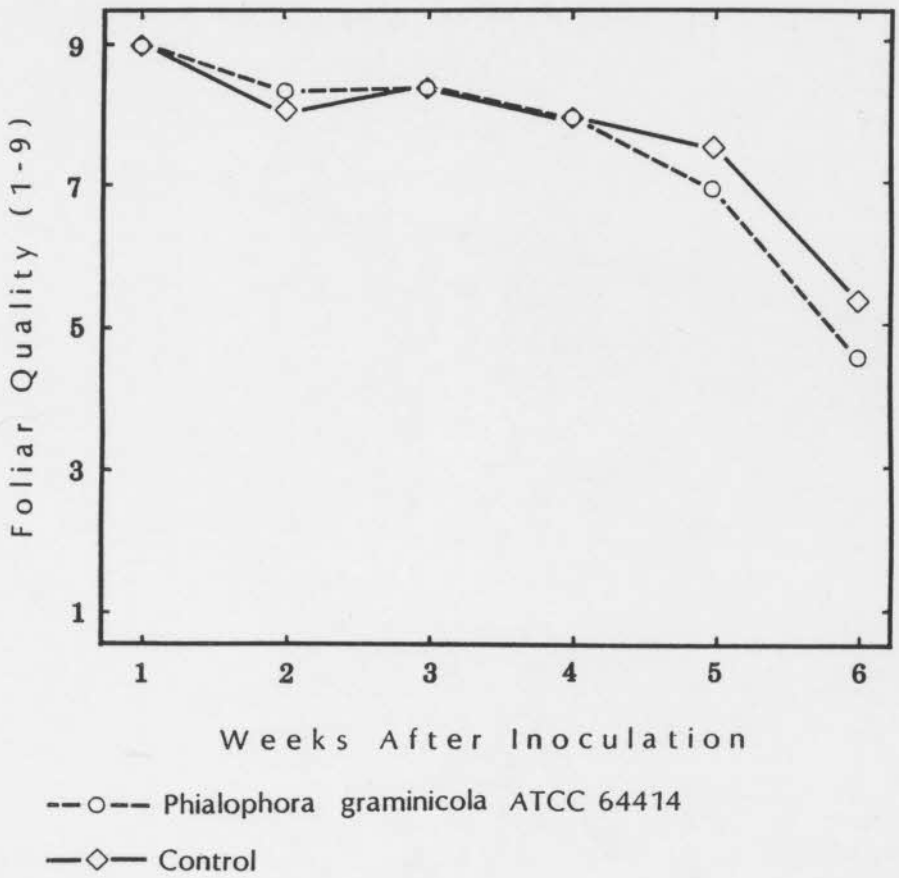


Fig. 28. Comparison of foliar quality between 8-week-old 'Merion' Kentucky bluegrass inoculated with P. graminicola (ATCC 64414) and a control.

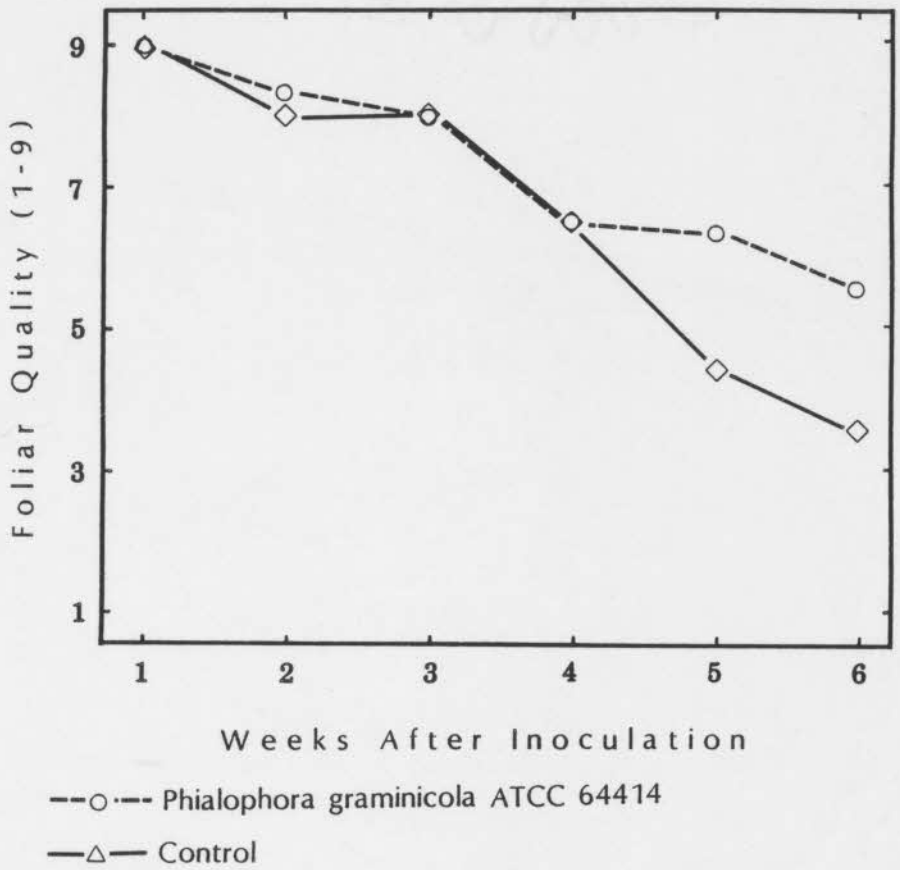


Fig. 29. Comparison of foliar quality between 7-year-old 'Merion' Kentucky bluegrass inoculated with P. graminicola (ATCC 64414) and a control.

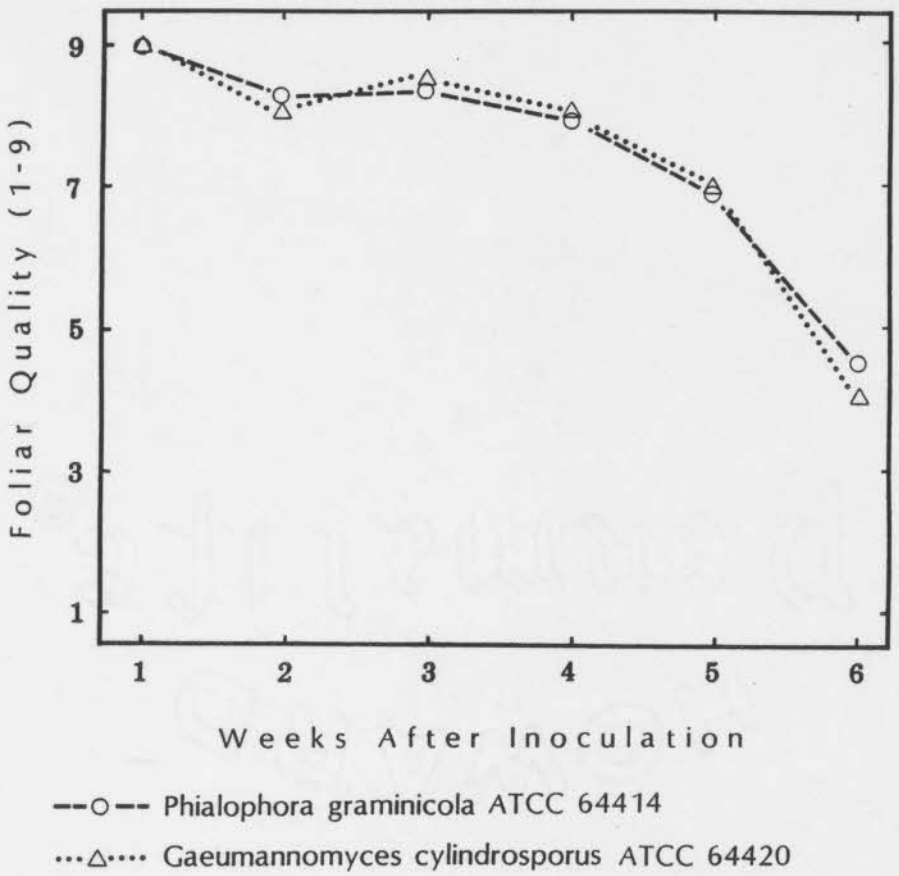


Fig. 30. Comparison of foliar quality between 8-week-old 'Merion' Kentucky bluegrass inoculated with P. graminicola (ATCC 64414) and G. cylindrosporus (ATCC 64420).

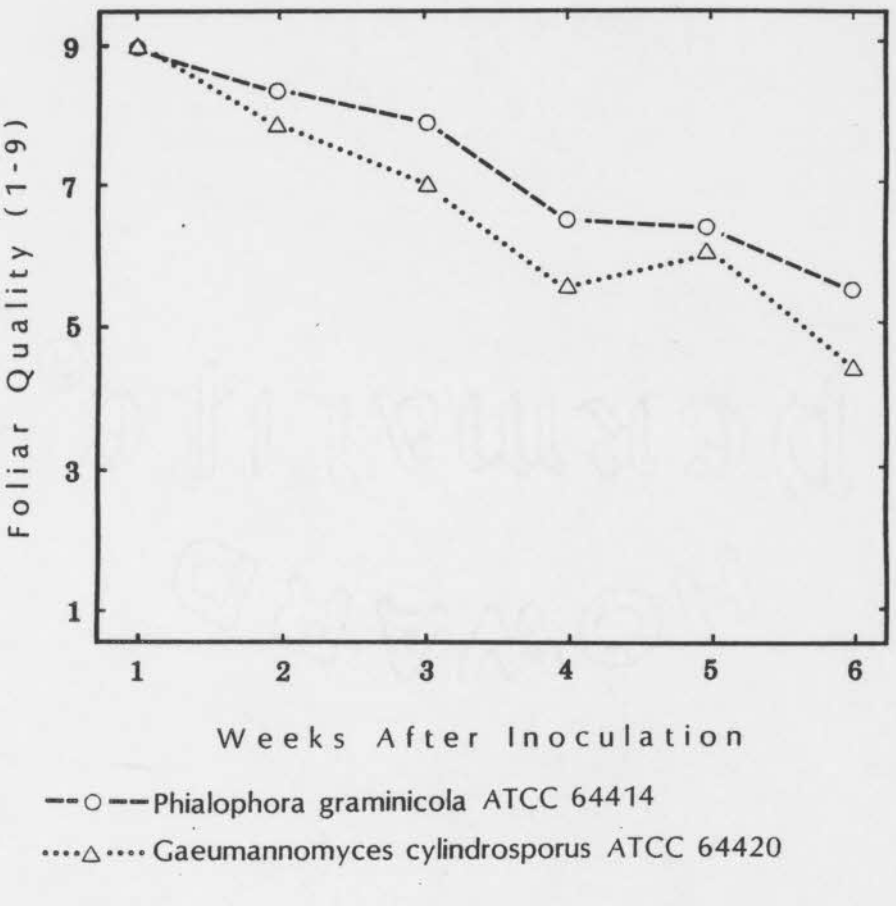


Fig. 31. Comparison of foliar quality between 7-year-old 'Merion' Kentucky bluegrass inoculated with P. graminicola (ATCC 64414) and G. cylindrosporus (ATCC 64420).

Table 11. Influence of ectotrophic Phialophora species on clipping yields (expressed as shoot dry wt.) and root discoloration of 8-week-old 'Merion' Kentucky bluegrass incubated for 6 weeks at 28°C.

Species	Isolate	Shoot Dry wt. Test 1	Shoot Dry wt. Test 2	Root[1] discoloration Test 1	Root[1] discoloration Test 2
		---- g/79 cm^2----			
M. poae	ATCC 64413	.80 c[2]	.80 c	4.8 a	4.7 a
M. poae	ATCC 64412	.86 c	1.02 bc	4.4 b	4.3 a
M. poae	ATCC 60239	.81 c	.84 c	4.5 ab	4.7 a
M. poae	ATCC 64131	.94 c	.96 bc	4.7 ab	4.7 a
M. poae	ATCC 64413 + ATCC 64412	.73 c	1.02 bc	4.7 ab	4.4 a
G. incrustans	ATCC 64418	1.22 b	1.31 b	3.1 c	3.3 b
P. graminicola	ATCC 64414	1.99 a	2.05 a	2.1 d	2.1 d
P. graminicola	ATCC 64415	1.93 a	1.74 a	2.3 d	2.7 c
G. cylindrosporus	ATCC 64420	2.08 a	1.73 a	2.1 d	2.3 d
Control	-------	1.93 a	1.82 a	1.0 e	1.0 e

[1]Based on visual rating scale of 1 to 5: 1 = no colonization, 2 = colonization and light discoloration, 3 = colonization and moderate discoloration, 4 = colonization and moderately severe discoloration, 5 = colonization and severe discoloration.

[2]Means within a column and followed by the same letter are not significantly different (P=0.05) according to Duncan's Multiple Range Test.

Table 12. Influence of ectotrophic Phialophora species on clipping yields (expressed as shoot dry wt.) and root discoloration of 7-year-old 'Merion' Kentucky bluegrass incubated for 6 weeks at 28°C.

Species	Isolate	Shoot Dry wt.		Root[1] Discoloration	
		Test 1	Test 2	Test 1	Test 2
		-----g/79 cm^2 --			
M. poae	ATCC 64413	1.5 cd[2]	0.9 f	4.8 a	4.6 a
M. poae	ATCC 64412	1.6 bcd	1.1 def	4.6 ab	4.2 ab
M. poae	ATCC 60239	1.3 d	1.1 def	4.7 ab	4.5 a
M. poae	ATCC 64131	1.3 cd	1.0 ef	4.6 ab	4.7 a
M. poae	ATCC 64413 + ATCC 64412	1.6 bcd	1.2 de	4.2 b	4.2 ab
G. incrustans	ATCC 64418	1.7 abc	1.2 d	3.2 c	3.9 b
P. graminicola	ATCC 64414	2.0 ab	1.5 c	3.1 c	2.8 c
P. graminicola	ATCC 64415	1.9 ab	1.8 ab	2.9 cd	2.5 c
G. cylindrosporus	ATCC 64420	2.0 ab	1.8 ab	2.5 d	2.8 c
Control	----------	2.1 a	1.8 a	2.8 cd	2.6 c

[1]Based on visual rating scale of 1 to 5: 1 = no colonization, 2 = colonization and light discoloration, 3 = colonization and moderate discoloration, 4 = colonization and moderately severe discoloration, 5 = colonization and severe discoloration.

[2]Means within a column and followed by the same letter are not sigificantly different (P=0.5) according to Duncan's Multiple Range Test.

yields (shoot dry weights) of 8-week-old and 7-year-old kentucky bluegrass inoculated with M. poae were significantly lower than yields from control plants. Root discoloration was extensive on 8-week-old Kentucky bluegrass inoculated with M. poae, whereas control plants showed little evidence of root discoloration. Significant differences in root discoloration between M. poae-inoculated plants and controls were evident on 7-year-old Kentucky bluegrass. However, roots of control plants showed a greater degree of discoloration on field-grown, 7-year-old turf than on the 8-week-old turf. This could be due to the presence of other root-infecting fungi on the mature turf as well as naturally senescing roots.

Gaeumannomyces incrustans (ATCC 64418) was only mildly pathogenic on both 8-week-old and 7-year-old Kentucky bluegrass. Significant differences in quality occurred between Kentucky bluegrass inoculated with G. incrustans and controls following 4-6 weeks at 28°C on 8-week-old and 7-year-old turf. Quality ratings of 8-week-old and 7-year-old plants inoculated with G. incrustans were usually higher than those inoculated with M. poae, although significant differences were not always apparent. Total clipping yields from G. incrustans inoculated plants were intermediate between control plants and M. poae inoculated plants. Root discoloration was moderate on both 8-week-old and 7-year-old Kentucky bluegrass turf.

Quality ratings of 8-week-old Kentucky bluegrass turf

inoculated with P. graminicola and G. cylindrosporus were significantly lower than controls on only one occasion (test 1, week 6). Clipping weights of 8-week-old and 7-year-old Kentucky bluegrass turf inoculated with P. graminicola and G. cylindrosporus were significantly lower than controls on one occasion (ATCC 64414 in test 2, 7-year-old turf) and were higher than M. poae in all tests. Root discoloration was light on roots colonized by these fungi. This may have been due to either poor colonization under the conditions of this test (P. graminicola and G. cylindrosporus have slow growth rates at temperatures of 28°C) or it reflects the inability of these fungi to cause much discoloration when established on turfgrass roots.

2. Host Range of M. poae.

Quality ratings of seven turfgrass species/cultivars inoculated with M. poae (ATCC 64413) are presented in Fig. 32-38 and Tables 5 and 6 in Appendix III. Clipping yields and estimates of root colonization/discoloration are shown in Table 13. Shoot lengths, shoot dry weights, root dry weights, and estimates of root colonization/discoloration of four cereal species inoculated with M. poae (ATCC 64413) and controls are presented in Tables 14 and 15.

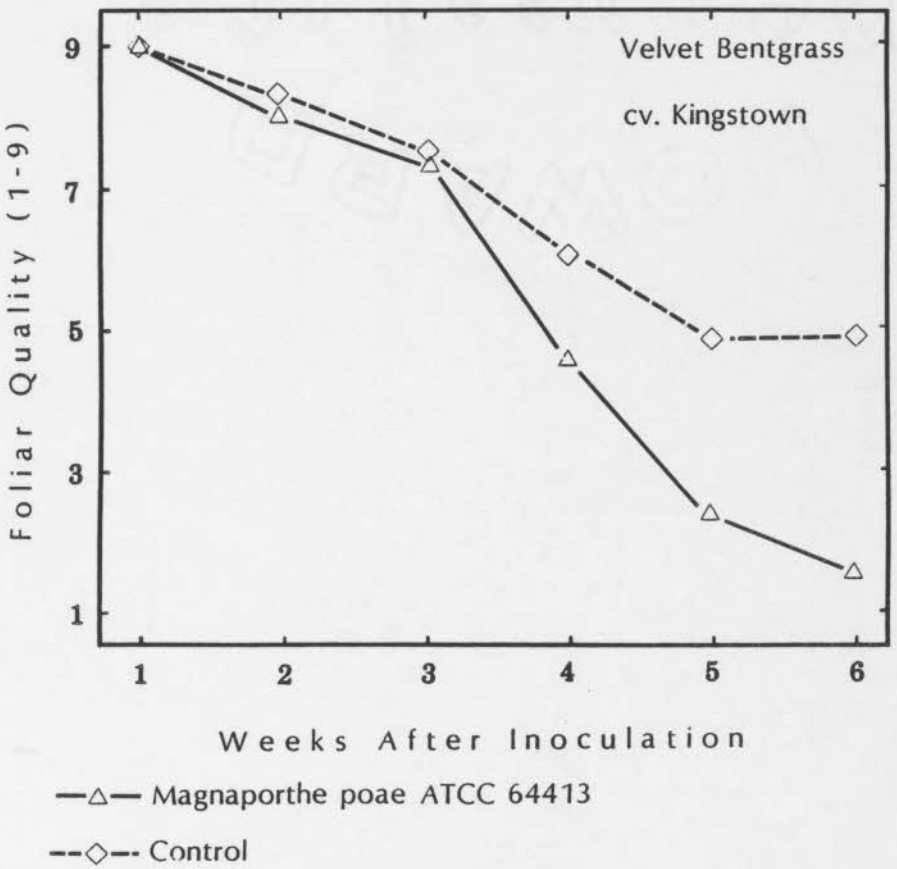


Fig. 32. Comparison of foliar quality between 'Kingstown' velvet bentgrass inoculated with M. poae (ATCC 64413) and a control.

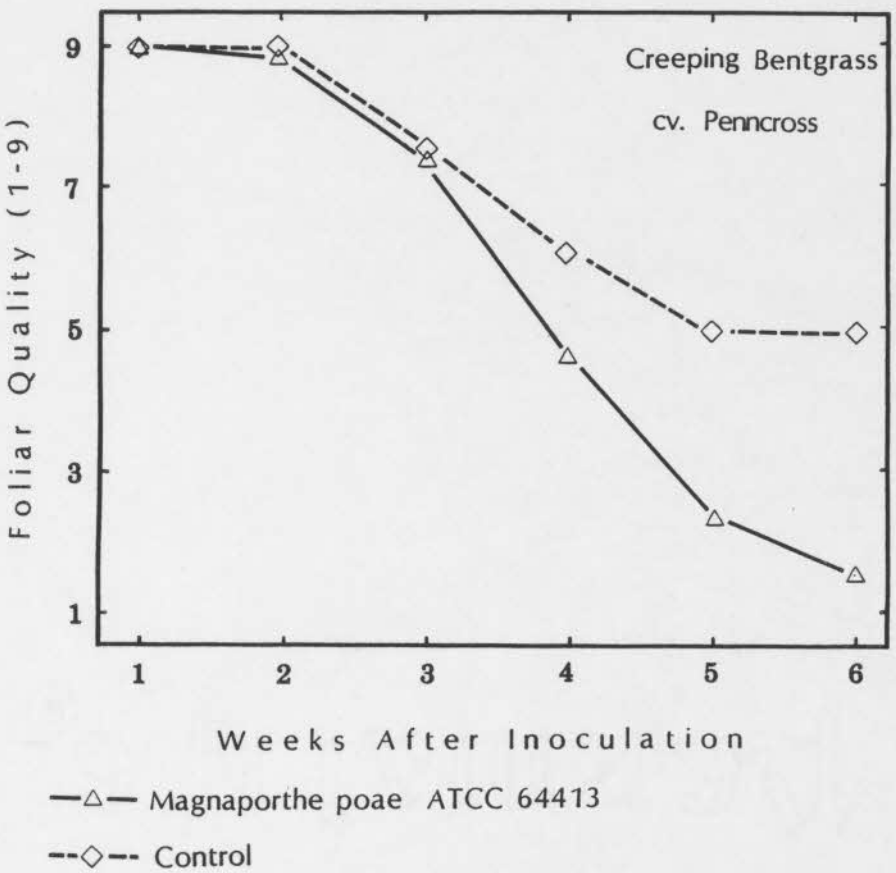


Fig. 33. Comparison of foliar quality between 'Penncross' creeping bentgrass inoculated with M. poae (ATCC 64413) and a control.

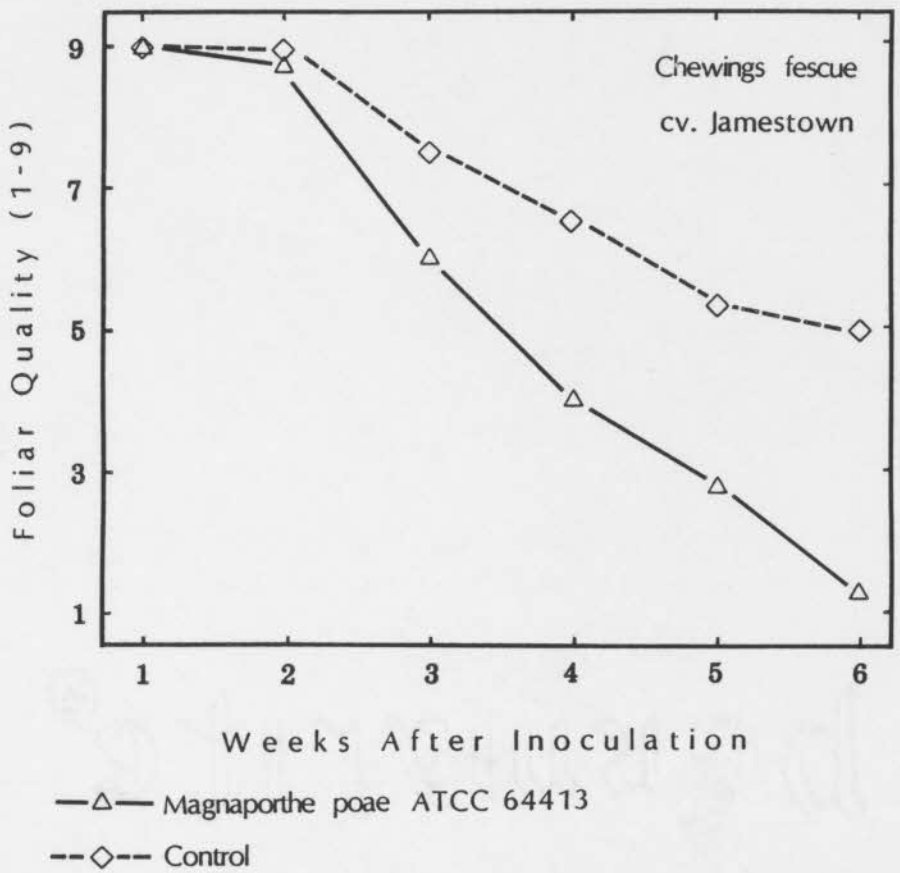


Fig. 34. Comparison of foliar quality between 'Jamestown' chewings fescue inoculated with M. poae (ATCC 64413) and a control.

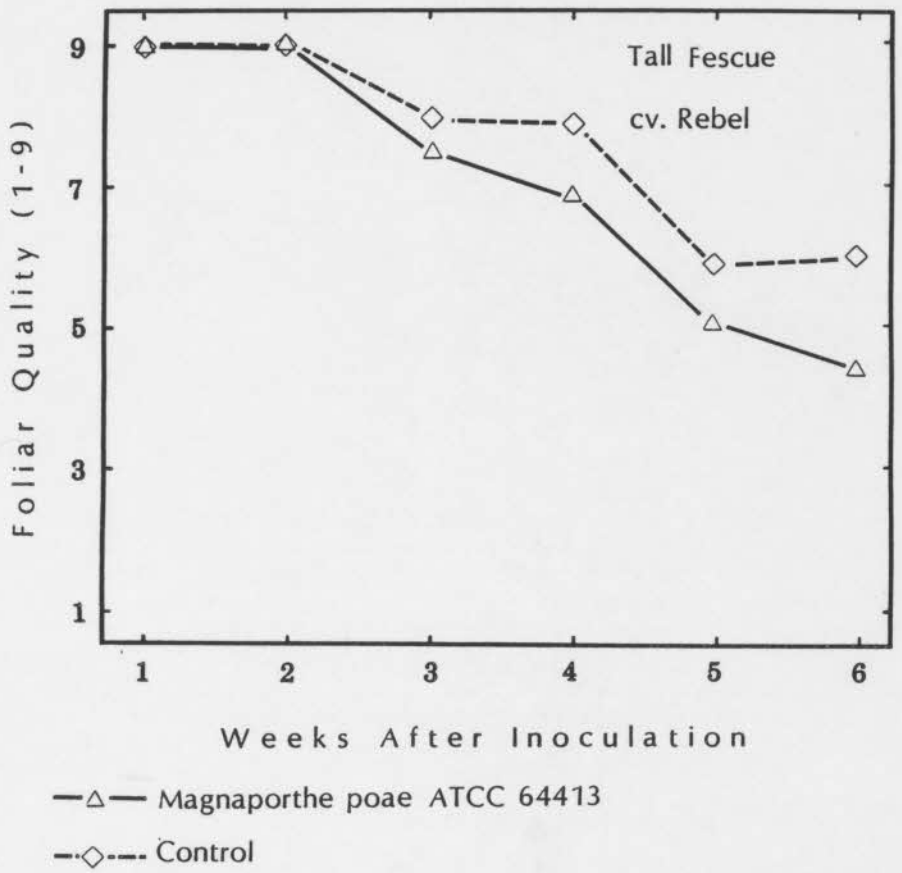


Fig. 35. Comparison of foliar quality between 'Rebel' tall fescue inoculated with M. poae (ATCC 64413) and a control.

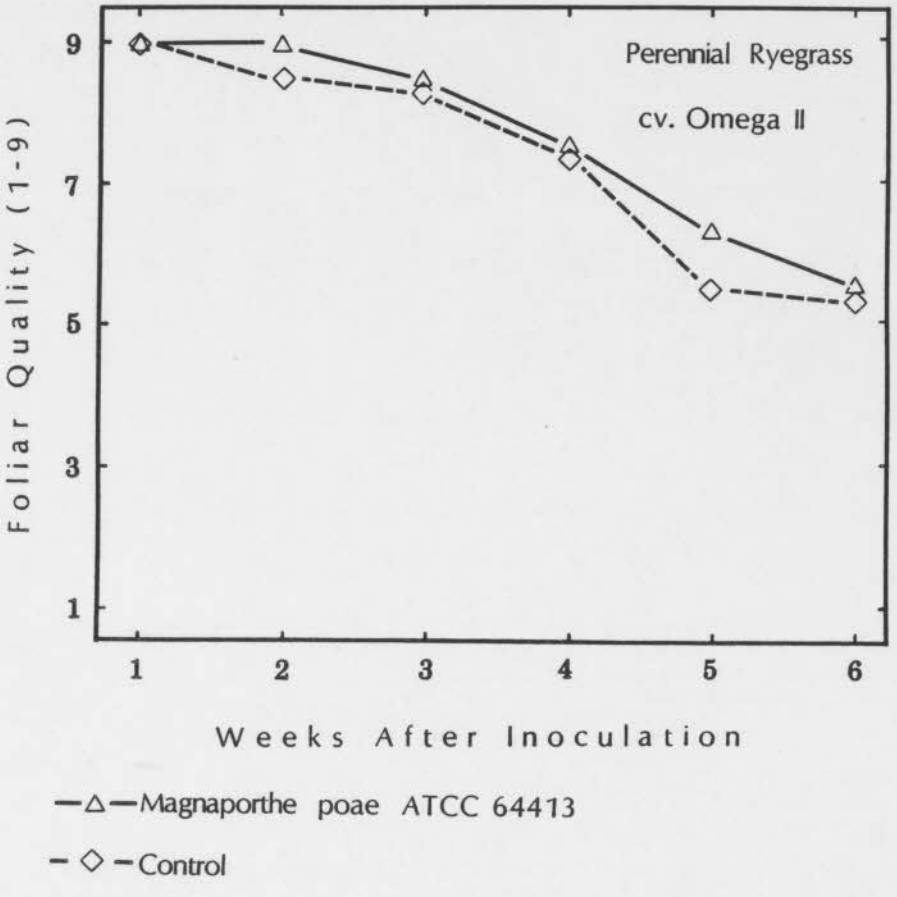


Fig. 36. Comparison of foliar quality between 'Omega II' perennial ryegrass inoculated with M. poae (ATCC 64413) and a control.

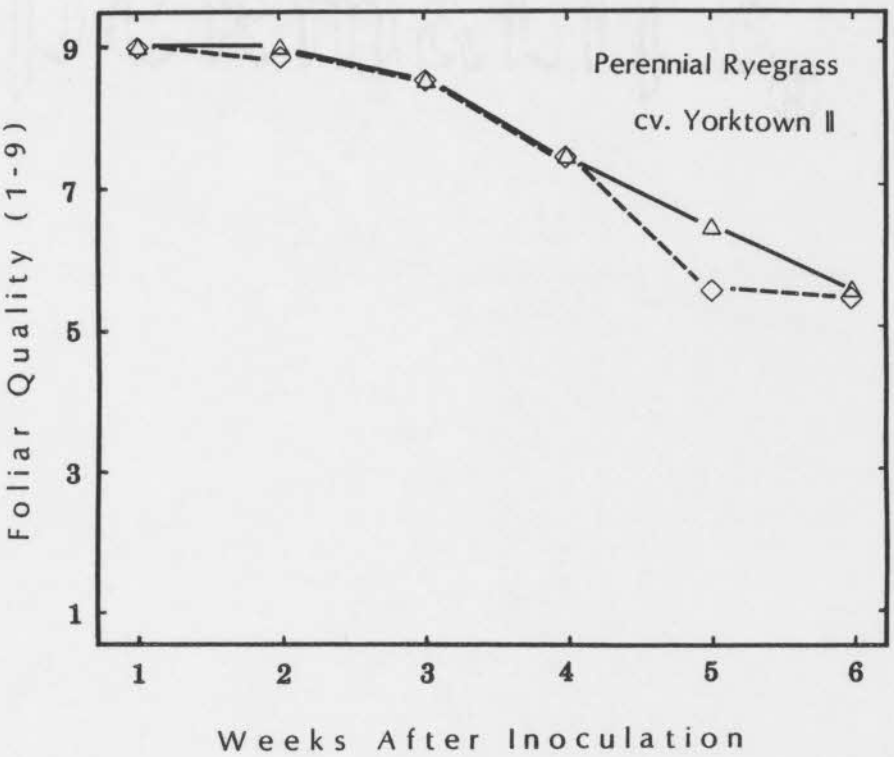


Fig. 37. Comparison of foliar quality between Yorktown II perennial ryegrass inoculated with M. poae (ATCC 64413) and a control.

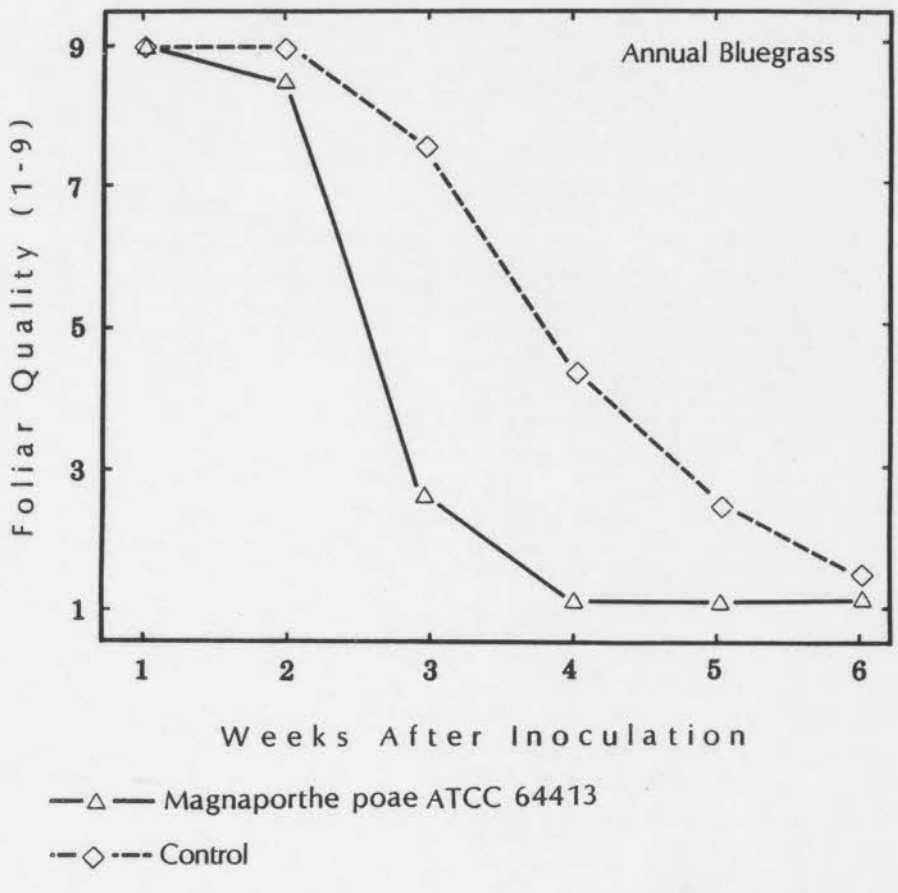


Fig. 38. Comparison of foliar quality between annual bluegrass inoculated with M. poae (ATCC 64413) and a control.

Table 13. Influence of Magnaporthe poae (ATCC 64413) on clipping yields (expressed as shoot dry wt.) and root discoloration of 6 turfgrasses incubated for 6 weeks at 28°C.

Species	Treatment Comparison	Shoot Dry Wt. Test 1	Shoot Dry Wt. Test 2	Root[1] Discoloration Test 1	Root[1] Discoloration Test 2
		---- g/79 cm^2 --			
Creeping bentgrass (cv. Penncross)	Control	1.5	1.7	1.0	1.0
	ATCC 64413	1.3 ns[2]	1.3 *	2.8 ***	3.0 ***
Velvet bentgrass (cv. Kingstown)	Control	1.4	1.2	1.0	1.0
	ATCC 64413	1.1 *	0.9 *	3.1 ***	3.0 ***
Chewings fescue (cv. Jamestown)	Control	1.2	1.0	1.0	1.0
	ATCC 64413	0.8 **	0.7 **	3.6 ***	3.6 ***
Tall fescue (cv. Rebel)	Control	2.0	1.4	1.0	1.0
	ATCC 64413	1.5 *	1.2 ns	2.7 ***	2.5 ***
Perennial Ryegrass (cv. Yorktown II)	Control	2.1	1.4	1.0	1.0
	ATCC 64413	1.8 ns	1.5 ns	2.1 ***	2.0 ***
Perennial Ryegrass (cv. Omega II)	Control	2.2	1.4	1.0	1.0
	ATCC 64413	2.0 ns	1.5 ns	2.5 ***	2.1 ***
Annual Bluegrass	Control	1.8	1.9	1.0	1.0
	ATCC 64413	0.8 ***	1.1 ***	4.9 ***	4.9 ***

[1]Based on visual rating scale of 1 to 5: 1 = no colonization, 2 = colonization and light discoloration, 3 = colonization and moderate discoloration, 4 = colonization and moderately severe discoloration, 5 = colonization and severe discoloration.

[2]Leaf dry weights analyzed using t-test. Significance levels are $P<0.001$ (***), $P<0.01$ (**), $P<0.05$ (*), or $P>0.05$ (ns).

Table 14. Influence of Magnaporthe poae (ATCC 64413) on shoot length, shoot weight, root weight, and root discoloration of four cereal species incubated for 6 weeks at 28°C (Test 1).

Species	Treatment Comparison	Shoot Length	Shoot Dry Wt.	Root Dry Wt.	Root Discoloration[1]
		- cm -	------- g --------		
Barley (Arivat)	Control	56.1	3.2	1.1	1.0
	ATCC 64413	12.1 ***	0.1 ***	0.1 ***	5.0 ***
Wheat (Redcoat)	Control	47.0	1.9	0.6	1.0
	ATCC 64413	30.1 ***	0.4 ***	0.2 ***	3.5 ***
Oats	Control	55.1	1.9	0.6	1.0
	ATCC 64413	46.4 **	1.1 ***	0.4 ***	2.3 ***
Rye	Control	39.3	1.9	1.0	1.0
	ATCC 64413	37.4 ns	1.0 *	0.4 **	2.3 ***

[1]Based on visual ratings of 1 to 5: 1 = no colonization, 2 = colonization and light discoloration, 3 = colonization and moderate discoloration, 4 = colonization and moderately severe discoloration, 5 = colonization and severe discoloration.

[2]Growth parameters analysed using t-test. Significance levels are $P<0.001$ (***), $P<0.01$ (**), $P<0.05$ (*), $P>0.05$ (ns).

Table 15. Influence of Magnaporthe poae (ATCC 64413) on shoot length, shoot weight, root weight, and root discoloration of four cereal species incubated for 6 weeks at 28 °C (Test 2).

Species	Treatment Comparison	Shoot Length	Shoot Dry Wt.	Root Dry Wt.	Root[1] Discoloration
		- cm -	------- g -------		
Barley (Arivat)	Control	52.5	3.3	1.7	1.0
	ATCC 64413	15.2 ***	0.1 ***	0.1 ***	5.0 ***
Wheat (Redcoat)	Control	57.7	2.1	0.8	1.0
	ATCC 64413	24.7 ***	0.1 ***	0.1 ***	4.8 ***
Oats	Control	59.1	3.0	1.0	1.0
	ATCC 64413	41.0 **	1.2 ***	0.7 ns	2.5 ***
Rye	Control	42.7	2.5	0.8	1.0
	ATCC 64413	37.3 ns	1.8 ns	1.0 ns	2.3 ***

[1]Based on visual ratings of 1 to 5: 1 = no colonization, 2 = colonization and light discoloration, 3 = colonization and moderate discoloration, 4 = colonization and moderately severe discoloration, 5 = colonization and severe discoloration.

[2]Growth parameters analysed using t-test. Significance levels are $P<0.001$ (***), $P<0.01$ (**), $P<0.05$ (*), $P>0.05$ (ns).

Turfgrasses

Magnaporthe poae was more pathogenic on some turfgrass species/cultivars than others. By the third week of Test 1 and the forth week of test 2, 'Jamestown' chewings fescue and annual bluegrass inoculated with M. poae were significantly lower in quality than control plants of the same species. Significantly lower quality ratings were apparent between inoculated and control 'Penncross' creeping bentgrass by the forth week of test 1 and the fifth week of test 2. 'Rebel' tall fescue inoculated with M. poae showed a significant decline in quality during the fifth and sixth week of test 1 while no differences occurred between inoculated and control plants of 'Yorktown II' and 'Omega II' perennial ryegrass in either test.

With two exceptions, turfgrasses showing a significant decline in quality also showed significantly lower clipping yields. Exceptions occurred for 'Penncross' creeping bentgrass in test 1, and 'Rebel' tall fescue in test 2.

Root colonization/discoloration was greatest for species/cultivars most adversely influenced by M. poae. Annual bluegrass and 'Jamestown' chewings fescue roots were severely discolored while roots of both 'Penncross' creeping and 'Kingstown' velvet bentgrasses showed only moderate discoloration. 'Rebel' tall fescue and the perennial ryegrasses were least affected, showing only slight root discoloration.

The high degree of pathogenicity of M. poae to annual bluegrass confirms field observations of summer patch on annual bluegrass fairways and putting greens. Dernoeden (personal communication, 1987) has observed summer patch symptoms on 'Pennlawn' fine fescue in areas adjacent to summer patch affected Kentucky bluegrass. Although bentgrasses were susceptible to M. poae in pathogenicity studies, summer patch has not been reported on bentgrasses in the field. Bentgrasses and perennial ryegrasses have been observed colonizing patches of turf killed by M. poae. The small discrepencies shown in the pathogenicity studies and field observations may be due to the high inoculum potential and immature stage of turf used in this study.

Cereals

M. poae was pathogenic to all cereal species but to varying degrees. Based on the parameters used in this test, barley (cv. Arivat) was most susceptible followed by wheat (cv. redcoat) and oats. Rye was least affected as indicated by non-significant differences between shoot length in test 1 and shoot length, shoot dry weight, and root dry weights in test 2. Root discoloration estimates reflected the degree of susceptibility among cereal species. Barley roots were most severely discolored followed by wheat, oats, then rye. Although these results indicated a high degree of susceptibility of some cereal species/

cultivars, to date M. poae has not been isolated from cereal roots in the field. These pathogenicity studies were conducted on seedling plants under extended periods of high temperatures. Whether M. poae is able to cause root dysfunction on mature plants under less severe environmental conditions is uncertain.

3. Inducement of patch symptoms on mature Kentucky bluegrass sod

Patch symptoms were first observed in plots inoculated with M. poae on 24 July 1986 (Fig. 39 and 40). Patches occurred on two of three replicate plots inoculated with isolates ATCC 56773 and ATCC 64413. One other isolate of M. poae (MI-1) did not produce patch symptoms. Inoculation of one isolate of G. cylindrosporus (SAK), one isolate of P. graminicola (REY), and three isolates of G. incrustans (ATCC 64418, CREST, ATBM) failed to induce any patch symptoms.

The first evidence of symptom development was the appearance of dark green patches of turf that wilted despite adequate soil moisture. The turf within these patches was stunted, turning a dark reddish-brown color over the course of 2 days. As symptoms progressed, the turf turned a tan color at the margins of the patches. Individual patches were roughly circular and ranged in size from 27 x 18 cm to 45 x 43 cm. (The size of these patches increased only 2 to 5 cm from the time symptoms first became

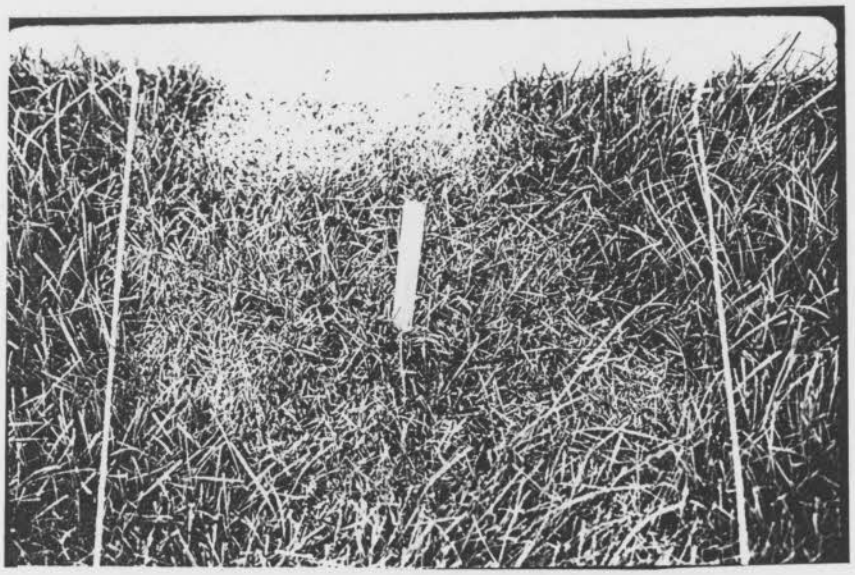


Fig. 39. Patch symptoms on Kentucky bluegrass turf inoculated with M. poae (ATCC 64413).

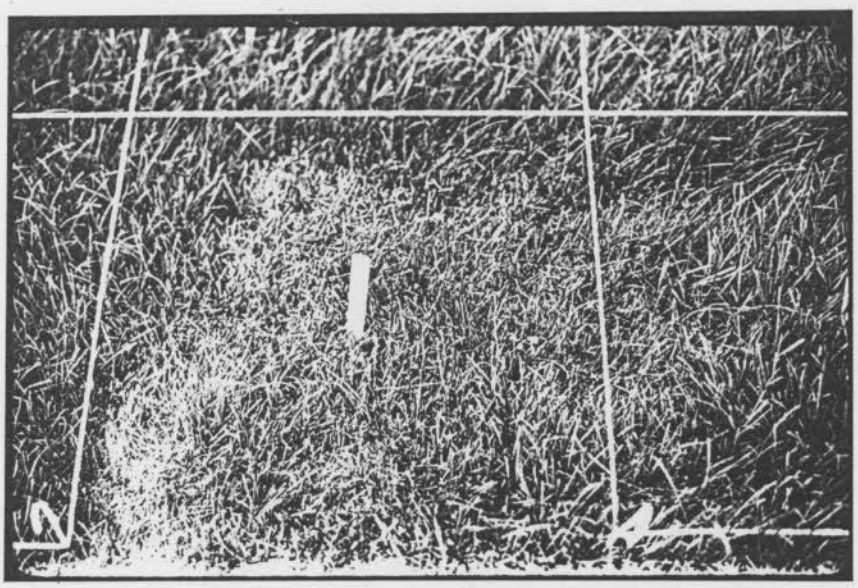


Fig. 40. Patch symptoms on Kentucky bluegrass turf inoculated with M. poae (ATCC 56773).

visible.) Many live and apparently healthy tillers remained in the center of the patch. No distinct leaf leasions were apparent although some dieback was evident on many tillers.

Roots and crowns of affected plants were rotted, most showing a distinct vascular discoloration. On some stem bases, large aggregates of swollen, pigmented hyphae were apparent (Fig. 10).

Isolation from diseased roots and crowns yielded several common root-inhabiting fungi as well as M. poae, G. incrustans, and P. graminicola. Of these fungi, only M. poae showed a consistent and dramatic increase over the fungi isolated from control plants. All M. poae isolates produced mature perithecia when mated with opposing mating types of the same species.

Since a synchronous wilt of plants in large patches was apparent at the onset of symptom development, it seems likely that M. poae grew up to 22 cm from the point of inoculation over a 1 1/2 year period and inhabited the roots without killing plants. When the turf became heat stressed following several days of $>32^{o}C$ temperatures, the fungus apparently gained an advantage over the suscept and began to invade the vascular system of the roots, causing dysfunction and eventually death.

It is not clear why symptoms did not appear in the third replication for isolates ATCC 64413 and ATCC 56773 or why MI-1 did not induce patch symptoms in any plot. It is possible that soil microflora or microfauna inhibited the

dissemination and colonization of this fungus or that physical conditions were limiting at microsites in the sod or in the underlying soil.

E. Histology

The ectotrophic, Phialophora fungi used in this study colonize and infect grass roots in a manner similar to other Phialophora and Gaeumannomyces fungi (Deacon, 1981 and Skou, 1981). Magnaporthe poae grows ectotrophically over the roots as dark brown mycelium and in some cases onto the leaf sheaths of grasses. Following extended periods of high temperatures, the fungus enters the plant by means of infection hyphae produced from single hyphopodia (Fig. 9) or hyphopodial aggregates (Fig. 10). Magnaporthe poae colonizes cortical cells by slender hyaline hyphae that may stimulate the production of lignitubers. In the advanced stages of infection, M. poae enters the vascular elements causing a brown discoloration and plugging of these tissues.

Gaeumannomyces incrustans also grows ectotrophically over the roots and leaf sheaths of grasses (Fig. 20). The fungus enters the plant by infection hyphae from hyphopodia (Fig. 18). Darkly pigmented swollen hyphae fill the cells in, and immediately below, the root epidermis. Infection hyphae ramify the remainder of the cortical cells in the area of infection and often become ensheathed by a

deposition of host material forming lignitubers. Gaeumannomyces incrustans may enter subsequently the vascular elements of grass roots and causes moderate vascular discoloration.

Phialophora graminicola and G. cylindrosporus produce ectotrophic mycelium on root surfaces and enter the host by means of infection hyphae from hyphopodia. Infection hyphae, however, rarely seem to progress to cortical cells near the stele and have not been observed colonizing vascular tissues.

Discussion

Magnaporthe poae is a non or weakly-pathogenic fungus on turfgrass roots when conditions are favorable for root growth. However, when conditions are less favorable for root growth and/or when the host's defense mechanisms are impaired due to environmental stress factors, M. poae is able to exploit the roots and crowns of turfgrasses and cause a severe vascular dysfunction. Smiley et al. (1985) reported that pathogenicity of isolates now known to be M. poae were strongly influenced by temperature, the highest degree of pathogenicity occurring at temperatures of 29oC. Other factors implicated in increasing the severity of summer patch include high levels of soil N, low mowing heights, arsenical herbicides, and irrigated turf suddenly subjected to drought stress (Smiley et al., 1985b).

Magnaporthe poae colonizes the roots and crowns of turfgrasses as ectotrophic mycelium at moderate soil temperatures without causing disease symptoms (provided conditions are favorable for ectotrophic growth). Inoculation of mature, field-grown sod has shown this fungus can grow up to 18 cm over 1 1/2 years without causing visible disease symptoms. Garrett (1970) noted that growth of ectotrophic mycelium is accellerated following a decline in plant vigor. Evidence of this was provided by Leach (1939) when he noticed a large increase in the rate of spread of Armillaria mellae (Fr.) Kummer over the roots of felled forest trees in East Arica. Another example involves the increased spread of Gaeumannomyces graminis following the close grazing of young winter wheat in Australia (Garrett, 1970). Close mowing of susceptible turfgrasses may similarly enhance the growth of M. poae over the root system.

The ectotrophic growth habit provides a competitive advantage for M. poae (as well as other ectotrophic, root-inecting fungi) in that they are present on the root surface (infection court) when host resistance breaks down. These fungi are thus able to incite infection before antagonistic facultative symbionts (such as Fusarium spp., Rhizoctonia spp., Pythium spp., etc.) are able to infect the roots (Garrett, 1970). Another advantage afforded by the ectotrophic habit is that it provides a mechanism whereby the fungus can incite multiple infections in comparatively

rapid succession along the length of the root, thus, diluting and eventually overwhelming the remaining host defenses (Garrett, 1970). Garrett, 1970 speculated that the ectotrophic infection-habit is conditional upon the active defense response of roots. If no active defense response is provided, internal infection can proceed without obstruction.

It should be noted that a complete breakdown in host resistance is not necessarily an advantage to specialized pathogens such as M. poae because it permits invasion by weak pathogens and then by saprotrophs. Thus, at its maximum, host resistance affects ectotrophic root-infecting fungi by restricting infection and spread; at an intermediate level, it affords protection against invasion of their living substrate by weak pathogens and saprotrophs which come in during the final stages of dwindling resistance (Garrett, 1970).

Smiley and Craven-Fowler (1984) claimed that summer patch occurs under the same cultural and environmental conditions as Fusarium blight syndrome and that their disease symptoms are indistinguishable. The authors noted that repeated attempts to reproduce the typical patch symptoms on mature turf with Fusarium spp. have failed, thus raising doubts concerning the role of these fungi as primary causal agents of Fusarium blight syndrome. It is possible that early ivestigators of Fusarium blight failed to isolate M. poae from turfgrass roots due to isolation

techniques which favored faster growing fungi. Fusarium spp. are usually the dominant fungi present on turfgrasses during the summer months and are readily isolated from non-diseased turfgrasses. It is therefore unclear which primary pathogen(s) were associated with turfgrasses exhibiting patch disease symptoms reported over the past 20 years.

CONCLUSION

The three ectotropic Phialophora species found during this investigation included the anamorphs of Magnaporthe poae sp. nov., Gaeumannomyces incrustans sp. nov., and Gaeumannomyces cylindrosporus (anamorph P. graminicola). Magnaporthe poae was isolated most frequently from patches in an active state of disease development. Gaeumannomyces incrustans and G. cylindrosporus were most often isolated from patches no longer exhibiting disease activity or from turf not affected with summer patch.

Of the three species, M. poae was the most pathogenic on both 8-week-old and 7-year-old Kentucky bluegrass turf at 28oC. Gaeumannomyces incrustans was mildly pathogenic while G. cylindrosporus (P. graminicola), the published causal agent of summer patch, was not pathogenic. When inoculated into mature (22-month-old) Kentucky bluegrass turf, M. poae produced patches that were identical to field symptoms of summer patch. Magnaporthe poae was reisolated from the roots and crowns of tillers from diseased patches. The results of this study indicate a causal relationship between M. poae and summer patch disease.

During this investigation it was established that the isolates designated as P. graminicola by Smiley and associates (ATCC 64413, ATCC 56773, NY-258, NE-179, and ATCC 60239) were actually M. poae. Therefore, the explanation by Smiley et al. (1985) concerning the differences in

morphology and virulence between North American isolates of P. graminicola and those from Australia and Europe is not valid.

Magnaporthe poae is pathogenic on Kentucky bluegrass and annual bluegrass turf during extended periods of hot, humid weather, particularly on turf with poor root development (Smiley and Craven Fowler, 1984 and Smiley et al., 1985b). Under these conditions, this fungus is able to exploit the root and crown tissues, causing severe vascular dysfunction and eventually death. The ability of M. poae to gain an advantage over the host is undoubtedly facilitated by its presence on the host's root system, as ectotrophic mycelium, prior to penetration and infection.

APPENDIX I

Media used for isolation and maintenance of ectotrophic *Phialophora* species and analysis of Terra Green® calcined clay.

1. Gaeumannomyces selective medium:

Autoclave 39 g Difco PDA in 1 l distilled water. Add the following compounds to 10 ml sterile distilled water;

100 mg streptomycin sulfate
500 mg L-B 3,4-dihydroxyphenylalaine (L-DOPA)
0.04 ml metalaxyl (Subdue®)
13.3 mg dicloran (Botran®)
50 mg 1-(3,5-dichlorophenyl)-3-methoxymethyl pyrroliden-2,4-dion (HOE 00703)

Pour this mixture into autoclaved PDA medium when cooled (50°C) and mix by swirling flask.

2. Potato dextrose agar (PDA):

Autoclave 15 g Difco PDA in 1 l distilled water.

3. PDA with antibiotics:

Autoclave 40 g Difco PDA in 1 l distilled water and add the following antibiotics to 10 ml sterile distilled water;

60 mg oxytetracycline

40 mg penicillin-G

50 mg streptomycin sulfate

Pour mixture into autoclaved PDA when cooled (50°C) and mix by swirling flask.

4. Water agar (WA):

Autoclave 15 g Difco Bacto-Agar in 1 l distilled water.

5. Analysis of Terra Green calcined clay:

a. washed	N	P	K	Ca	Mg	pH
	ppm					
	7.0	20	842	846.6	369.4	5.4

b. unwashed	N	P	K	Ca	Mg	pH
	ppm					
	3.0	20	929	779.4	367.1	5.4

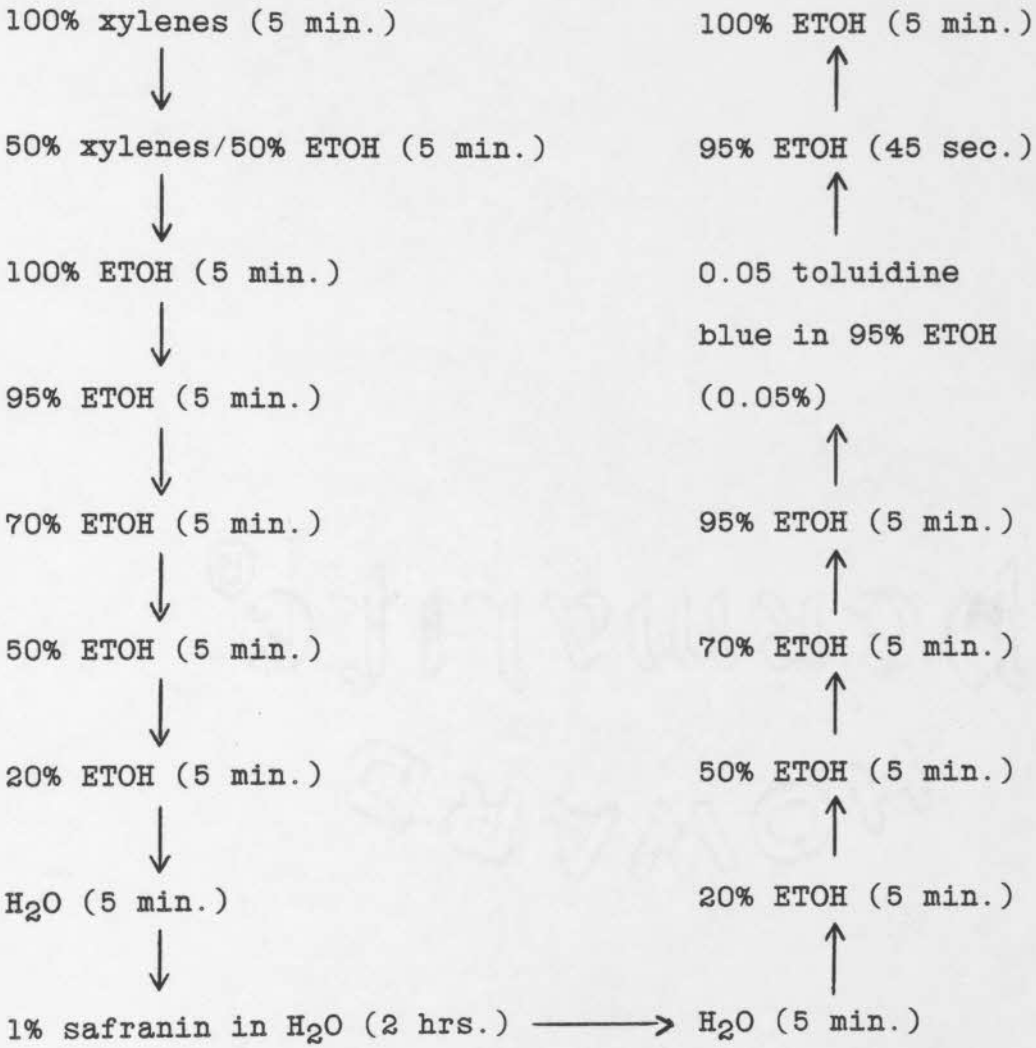
APPENDIX II

Histology

1. Graded tertiary-butyl-alcohol (TBA) series:

	Time of immersion				
	2 hrs.	2 hrs.	2 hrs.	24 hrs.	24 hrs.
H2O	30%	15%	5%	-	-
ETOH	50%	50%	40%	25%	-
TBA	20%	35%	55%	75%	100%

2. Hydration sequence for staining perithecia in 1% safranin and dehydration sequence for counterstain with toluidine blue.



APPENDIX III

Influence of ectotrophic *Phialophora* species on the foliar quality of turfgrasses

Table 1. Influence of ectotrophic Phialophora species on foliar quality of 8-week-old 'Merion' Kentucky bluegrass (Test 1).

Fungus Species	Isolate	Foliar Quality [1] Week 1	2	3	4	5	6
M. poae	ATCC 64413	9.0 a [2]	6.5 b	3.5 b	1.8 c	1.0 d	1.0 d
M. poae	ATCC 64412	9.0 a	6.8 b	3.3 b	2.3 c	1.5 d	1.0 d
M. poae	ATCC 60259	9.0 a	6.3 b	4.0 b	1.3 c	1.0 d	1.0 d
M. poae	ATCC 64131	9.0 a	6.5 b	3.5 b	1.8 c	1.0 d	1.0 d
M. poae	ATCC 64413 + ATCC 64412	9.0 a	6.3 b	4.0 b	1.3 c	1.0 d	1.0 d
G. incrustans	ATCC 64418	9.0 a	7.8 a	7.5 a	5.3 b	4.3 c	2.8 c
P. graminicola	ATCC 64414	9.0 a	8.3 a	8.3 a	7.8 a	6.8 b	4.5 b
P. graminicola	ATCC 64415	9.0 a	8.0 a	8.5 a	8.0 ab	7.0 ab	4.5 b
G. cylindrosporus	ATCC 64420	9.0 a	8.0 a	8.5 a	8.0 a	7.0 ab	4.8 b
Control	----------	9.0 a	8.0 a	8.3 a	7.8 ab	7.5 a	5.3 a

[1] Based on a scale of 1-9, 1 = dead plants (straw-yellow color), 9 = dense, dark-green and actively growing turf.

[2] Means within a column and followed by the same letter are not significantly different (P = 0.05) according to Duncan's Multiple Range Test.

Table 2. Influence of ectotrophic Phialophora species on foliar quality of 8-week-old 'Merion' Kentucky bluegrass (Test 2).

Fungus Species	Isolate	Foliar Quality[1] Week 1	2	3	4	5	6
M. poae	ATCC 64413	8.8 a[2]	7.5 b	4.0 d	1.0 c	1.0 c	1.0 d
M. poae	ATCC 64412	8.8 a	8.0 ab	5.0 cd	1.0 c	1.0 c	1.0 d
M. poae	ATCC 60259	9.0 a	6.8 c	4.3 d	1.0 c	1.0 c	1.0 d
M. poae	ATCC 64131	9.0 a	6.7 c	4.0 d	1.0 c	1.0 c	1.0 d
M. poae	ATCC 64413 + ATCC 64412	9.0 a	8.0 ab	4.5 d	1.0 c	1.0 c	1.0 d
G. incrustans	ATCC 64418	9.0 a	8.0 ab	6.0 bc	2.8 b	2.8 b	2.0 c
P. graminicola	ATCC 64414	8.8 a	7.8 ab	7.0 ab	5.3 a	5.3 a	3.8 a
P. graminicola	ATCC 64415	9.0 a	7.8 ab	7.5 a	4.8 a	4.8 a	3.3 ab
G. cylidrosporus	ATCC 64420	9.0 a	8.3 a	7.3 a	4.5 a	5.0 a	3.8 b
Control	----------	8.8 a	7.8 ab	7.0 ab	5.0 a	4.5 a	3.0 a

[1]Based on a scale of 1-9, 1 = dead plants (Straw-yellow color), 9 = dense, dark-green, actively growing turf.

[2]Means within a column and followed by the same letter are not significantly different (P = 0.05) according to Duncan's Multiple Range Test.

Table 3. Influence of ectotrophic Phialophora species on foliar quality of 7-year-old 'Merion' Kentucky bluegrass (Test 1).

Fungus Species	Isolate	Foliar Quality[1] Week 1	2	3	4	5	6
M. poae	ATCC 64413	9.0 a[2]	7.8 a	6.3 cd	4.0 c	1.8 d	1.0 c
M. poae	ATCC 64412	9.0 a	8.0 a	6.5 cd	5.0 c	3.3 bc	1.8 c
M. poae	ATCC 60259	9.0 a	7.8 a	5.0 f	3.8 d	1.5 d	1.0 c
M. poae	ATCC 64131	9.0 a	7.5 a	5.5 ef	4.0 cd	2.0 cd	1.0 c
M. poae	ATCC 64413 + ATCC 64412	9.0 a	7.5 a	6.0 de	5.0 bc	2.5 cd	1.3 c
G. incrustans	ATCC 64418	9.0 a	8.0 a	7.5 ab	5.5 ab	3.3 bc	2.0 c
P. graminicola	ATCC 64414	9.0 a	8.3 a	7.8 a	6.5 a	6.3 a	5.5 a
P. graminicola	ATCC 64415	9.0 a	7.5 a	6.5 cd	5.5 ab	4.5 b	3.8 b
G. cylindrosporus	ATCC 64420	9.0 a	7.8 a	7.0 bc	5.5 ab	6.0 a	4.3 b
Control	----------	9.0 a	8.0 a	8.0 a	6.5 a	4.3 b	3.5 b

[1]Based on a scale of 1-9, 1 = dead plants (straw-yellow color), 9 = dense, dark-green, actively growing turf.

[2]Means within a column and followed by the same letter are not significantly different (P = 0.05) according to Duncan's Multiple Range Test.

Table 4. Influence of ectotrophic Phialophora species on foliar quality of 7-year-old 'Merion' Kentucky bluegrass (Test 2).

Fungus Species	Isolate	Foliar Quality[1] Week 1	2	3	4	5	6
M. poae	ATCC 64413	8.0 a[2]	6.8 b	5.5 d	2.8 cd	1.3 d	1.0 c
M. poae	ATCC 64412	8.3 a	7.3 ab	6.5 bc	3.0 c	1.8 d	1.3 c
M. poae	ATCC 60259	8.8 a	7.5 ab	6.5 bc	3.5 bc	1.5 d	1.0 c
M. poae	ATCC 64131	8.5 a	7.3 ab	6.3 cd	1.8 d	1.3 d	1.0 c
M. poae	ATCC 64413 + ATCC 64412	8.3 a	7.8 a	6.8 abc	2.5 cd	2.3 d	1.5 c
G. incrustans	ATCC 64418	8.5 a	7.3 ab	6.5 bc	4.5 b	3.5 c	2.5 b
P. graminicola	ATCC 64414	8.0 a	7.0 ab	7.0 abc	6.3 a	4.5 ab	2.8 b
P. graminicola	ATCC 64415	8.8 a	7.5 ab	7.5 a	6.5 a	5.0 a	4.3 a
G. cylindrosporus	ATCC 64420	8.5 a	7.5 ab	6.8 abc	5.8 a	4.0 bc	2.8 b
Control	----------	8.3 a	7.3 ab	7.3 ab	6.3 a	4.3 abc	3.3 b

[1]Based on a scale of 1-9, 1 = dead plants (straw-yellow color), 9 = dense, dark-green, actively growing turf.

[2]Means within a column and followed by the same letter are not significantly different (P= 0.05) according to Duncan's Multiple Range Test.

Table 5. Influence of *Magnaporthe poae* (ATCC 64413) on foliar quality of 7 turfgrasses incubated for 6 weeks at 28° C (Test 1).

Species	Treatment Comparison	Foliar Quality[1] Week 1	2	3	4	5	6
Creeping bentgrass (cv. Penncross)	Control	9.0	9.0	7.5	6.0	4.8	4.8
	ATCC 64413	9.0 ns[2]	8.8 ns	7.3 ns	4.5 *	2.3 **	1.5 **
Velvet bentgrass (cv. Kingstown)	Control	9.0	8.3	7.5	6.8	5.5	5.0
	ATCC 64413	9.0 ns	8.0 ns	6.3 ns	5.0 **	3.5 **	2.5 ***
Chewings fescue (cv. Jamestown)	Control	9.0	9.0	7.5	6.5	5.3	5.0
	ATCC 64413	9.0 ns	8.8 ns	6.0 *	4.0 ***	2.8 ***	1.3 ***
Tall fescue (cv. Rebel)	Control	9.0	9.0	8.0	7.8	6.8	6.0
	ATCC 64413	9.0 ns	9.0 ns	7.5 ns	6.8 ns	5.0 *	4.3 ***
Perennial ryegrass (cv. Yorktown II)	Control	9.0	8.8	8.5	7.3	6.0	5.0
	ATCC 64413	9.0 ns	9.0 ns	8.5 ns	7.3 ns	5.8 ns	4.0 ns
Perennial ryegrass (cv. Omega II)	Control	9.0	8.5	8.3	7.3	5.5	5.3
	ATCC 64413	9.0 ns	9.0 ns	8.5 ns	7.5 ns	6.3 ns	5.5 ns
Annual bluegrass	Control	9.0	9.0	7.5	4.3	2.3	1.3
	ATCC 64413	9.0 ns	8.5 ns	2.5 ***	1.0 ns	1.0 *	1.0 ns

[1] Based on a scale of 1-9. 1 = dead plants (straw-yellow color), 9 = dence, dark-green, actively growing turf.

[2] Significance levels are $P<0.001$ (***), $P<0.01$ (**), $P<0.05$ (*), or $P>0.05$ (ns).

Table 6. Influence of Magnaporthe poae (ATCC 64413) on foliar quality of 7 turfgrasses incubated for 6 weeks at 28 °C (Test 2).

Species	Treatment Comparison	Foliar Quality[1] Week 1	2	3	4	5	6
Creeping bentgrass (cv. Penncross)	Control	9.0	9.0	8.0	7.0	6.3	4.5
	ATCC 64413	9.0 ns[2]	8.5	7.3	6.3	3.8 **	2.0 **
Velvet bentgrass (cv. Kingstown)	Control	9.0	8.0	7.5	6.3	5.0	4.5
	ATCC 64413	9.0 ns	8.0 ns	7.3 ns	6.3 ns	4.5 ns	1.5 ***
Chewings fescue (cv. Jamestown)	Control	9.0	8.8	7.5	7.5	6.5	4.8
	ATCC 64413	9.0 ns	8.5 ns	6.8 ns	5.3 ***	3.5 ***	2.0 ***
Tall fescue (cv. Rebel)	Control	9.0	9.0	7.8	7.0	5.8	5.0
	ATCC 64413	9.0 ns	9.0 ns	7.8 ns	7.0 ns	5.3 ns	4.3 *
Perennial ryegrass (cv. Yorktown II)	Control	9.0	9.0	7.5	6.8	5.0	5.3
	ATCC 64413	9.0 ns	9.0 ns	8.0 ns	7.0 ns	5.5 ns	5.0 ns
Perennial ryegrass (cv. Omega II)	Control	9.0	9.0	7.8	7.0	5.3	5.0
	ATCC 64413	9.0 ns	9.0 ns	8.0 ns	7.0 ns	5.3 ns	4.8 ns
Annual bluegrass	Control	9.0	8.0	7.3	6.3	4.5	1.5
	ATCC 64413	9.0 ns	7.5 ns	4.5 ***	2.8 **	1.0 ***	1.0 ns

[1]Based on a scale of 1-9, 1 = dead plants (straw yellow color), 9 = dence, dark-green, actively growing turf.

[2]Significance levels are $P<0.001$ (***), $P<0.01$ (**), $P<0.05$ (*), or $P>0.05$ (ns).

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