Incidence of *Bipolaris* and *Exserohilum* Species in Corn Leaves in North Carolina

K. J. LEONARD, Research Plant Pathologist, USDA-ARS, Department of Plant Pathology, Box 7616, North Carolina State University, Raleigh 27695-7617; R. P. THAKUR, Plant Pathologist, Pearl Millet Improvement Program, ICRISAT, Patancheru P.O., Andhra Pradesh 502324, India; and S. LEATH, Research Plant Pathologist, USDA-ARS, Department of Plant Pathology, Box 7616, North Carolina State University, Raleigh 27695-7616

ABSTRACT

Leonard, K. J., Thakur, R. P., and Leath, S. 1988. Incidence of *Bipolaris* and *Exserohilum* species in corn leaves in North Carolina. Plant Disease 72: 1034-1038.

Samples of 42–60 corn leaves were collected without regard to disease symptoms at 2-m intervals in each of eight cornfields in western and two in eastern North Carolina in 1985. Sampling dates for the 10 fields ranged from 16 July to 8 August. The leaves were surface-sterilized and incubated in moist chambers for isolation of large-spored species of *Bipolaris* and *Exserohilum*. Incidence of *B. zeicola* infection was high (41–81%) in leaves from nine of the 10 fields. *B. maydis* and *E. turcicum*, which are more aggressive pathogens of hybrid corn than *B. zeicola*, occurred at lower incidences of 0–48% and 0–50%, respectively, in leaves from these fields at the time of sampling. This indicated that initial population densities of these aggressive pathogens were much lower than that of *B. zeicola*. Forty-five isolates of *E. turcicum* collected before 28 August were all race 1. Three isolates of race 2 were obtained later from fields of hybrids with the *Ht1* gene for resistance to race 1. All 75 isolates of *B. maydis* obtained from the 10 extensively sampled fields were race O; they were equally virulent on inbred line B73 with normal cytoplasm or B73 with C, S, or T male sterile cytoplasm. Frequencies of mating types of *B. maydis* and *B. zeicola* varied from field to field, with no correlation between the frequencies in the two species. Races 2 and 3 of *B. zeicola*, however, had similar mating type frequencies in these fields. *B. sorokiniana* was isolated from leaves from six of the 10 fields; the greatest incidence was 37%. *E. rostratum* was found in leaves from five fields at incidences up to 18%, and *E. holmii* occurred in leaves in one field at 11% incidence. These data indicate that a variety of species of *Bipolaris* and *Exserohilum* are able to infect green leaves of mature corn plants in the field. Thus, corn may contribute to the survival of species that are primarily pathogenic on other gramineous hosts.

Additional keywords: *Cochliobolus carbonum*, *C. heterostrophus*, maize, northern leaf blight, southern leaf blight, *Zea mays*

As a continuation of studies of genetic variation in populations of *Bipolaris zeicola* (Stout) Shoemaker (teliomorph *Cochliobolus carbonum* Nelson) (14,19), we sampled leaves of corn (*Zea mays* L.) in 10 fields in North Carolina in 1985. In isolating *B. zeicola* from these leaves, we realized that several species of *Bipolaris* and *Exserohilum* were present at relatively high incidences in leaves from many of the fields. Not all of the species we found are generally regarded as pathogens of corn, and those species that are known as important corn pathogens were not always found with the greatest frequency. Consequently, we expanded the study to include all of the large-spored species of *Bipolaris* and *Exserohilum* that occurred in the leaves collected. A more detailed analysis of genetic variation within the *B. zeicola* populations in these fields will be reported in a separate paper.

In this paper, we report on virulence and mating types of two important leaf blight pathogens of corn: *Bipolaris maydis* (Nisikado) Shoemaker (teliomorph *C. heterostrophus* Drechs.), the cause of southern corn leaf blight, and *Exserohilum turcicum* (Pass.) Leonard & Suggs (teliomorph *Setosphaeria turcica* (Luttrell) Leonard & Suggs.), the cause of northern leaf blight. Two races, race O and race T, of *B. maydis* have been described in the United States, although race T, which is specifically virulent on corn with Texas male sterile cytoplasm (cms-T), has not been important in recent years (13,15). In China, Wei et al (28) recently discovered a third race, race C, which produces a host-specific toxin specifically active against corn lines with C male sterile cytoplasm (cms-C). Cms-C has been used, although sparingly, as an alternative to cms-T in the production of hybrid corn seed in the United States, so we felt that it would be important to screen our isolates for virulence not only on corn with cms-T but also on corn with cms-C.

The *Ht1* gene for chlorotic lesion resistance to northern leaf blight has been widely, although not exclusively, used in corn hybrids grown in the United States. Because northern leaf blight is generally less important in the southeastern United States than in the Corn Belt, *Ht1* has been less widely used in the Southeast. Berquist and Masias (2) first reported the occurrence of race 2 of *E. turcicum*, which was not controlled by *Ht1* resistance, in corn in Hawaii. Race 2 was found in Indiana in 1979 (26). By 1981, race 2 had been found throughout the Corn Belt as well as in New York, Pennsylvania, and Florida (5,18). The development of a northern leaf blight epidemic in August 1985 in corn in several counties of western North Carolina, where *Ht1* had not been used widely, provided an opportunity to examine relative frequencies of races 1 and 2 in the absence of strong host selection for race 2.

Kline and Nelson (6,22) reported that many species of *Bipolaris* have rather broad host ranges and that species commonly associated with grasses or small grains can infect corn seedlings in the greenhouse. Their tests were done with high inoculum doses and under conditions extremely favorable for infection. The occurrence of several *Bipolaris* and *Exserohilum* species in leaf samples from the fields in 1985 provided a test of the ability of some of these grass and small grain pathogens to infect and survive in corn under field conditions. The results reported herein provide insight into the role of alternative hosts in the survival of leaf-infecting species of *Bipolaris* and *Exserohilum*.

The corn leaves in the sampled fields were collected without regard to disease symptoms. Thus, our objectives in this survey were to obtain an unbiased estimate of the relative prevalence of *Bipolaris* and *Exserohilum* species in leaves of corn plants in the field and, in the case of important corn pathogens, to obtain estimates of the prevalence of pathogenic races and of mating types among the isolates obtained.

MATERIALS AND METHODS

Ten cornfields in North Carolina were sampled extensively for leaf-inhabiting species of *Bipolaris* and *Exserohilum* in

Present address of first author: USDA Cereal Rust Laboratory, University of Minnesota, St. Paul, MN 55108

Journal Article 761 of the International Crops Research Institute for the Semi-Arid Tropics (ICRISAT). Paper No. 11409 of the Journal Series of the North Carolina Agricultural Research Service, Raleigh 27695-7601.

Cooperative investigations of the USDA-ARS and North Carolina Agricultural Research Service.

Accepted for publication 12 July 1988 (submitted for electronic processing).

1985 One leaf was taken per plant at approximately 2-m intervals in a portion of each field The sampled leaves were among the oldest leaves that had not yet senesced on the plants, and they were collected without regard to symptoms Leaves from 60 plants were collected in fields 1–9 and from 42 plants in field 10

Six of the sampled fields were in Wilkes County which lies just east of the Blue Ridge Mountains in western North Carolina (Fig 1) Elevations of these six fields ranged from 283 to 450 m Two other sampled fields also were in western North Carolina field 6 (elevation 326 m) in Yadkin County just east of Wilkes County, and field 9 (elevation 370 m) in McDowell County, southwest of Wilkes County and just east of the Blue Ridge Mountains The other two sampled fields were in Wake County (elevation 102 m) and Edgecombe County (elevation 38 m) in eastern North Carolina Each field was sampled once with sampling dates ranging from 16 July to 8 August 1985

In early August 1985 northern leaf blight lesions were reported in many fields in western North Carolina so we collected symptomatic leaves from 14 additional fields all but one of which were in five western North Carolina counties (Fig 2) Isolates of *E. turcicum* were obtained from one or two northern leaf blight lesions from each of these 14 fields

The leaves from the extensively sampled fields were dried in a plant press and kept at room temperature until they could be incubated for isolation of *Bipolaris* and *Exserohilum* species Because conidia deposited on the surface of corn leaves either in the field or in the laboratory as contaminants can germinate and produce sporulating colonies on the leaf surface within 2 or 3 days (9), it was necessary to surface-sterilize the collected leaves immediately before incubation Pieces up to 8 cm long and several centimeters wide were cut from each leaf and surface-sterilized by immersion for 30 sec in 70% ethanol followed by 30 sec in 0 5% sodium hypochlorite solution The leaf pieces were rinsed in tap water and incubated on wet filter paper in petri dishes at room temperature under fluorescent light with a 12-hr photoperiod

After 2–4 days, the incubated leaf pieces were examined under a stereo microscope at 25× for conidiophores and conidia of large-spored species of *Bipolaris* and *Exserohilum* A representative of each type present on each leaf was collected by picking conidia directly from the conidiophores with a sterile needle The conidia were transferred to potato-dextrose agar (PDA) with 10 gm of dextrose per liter The resulting cultures were transferred once to ensure purity, after which the isolates were stored as conidial suspensions in 30% glycerol frozen at −70 C

Inocula for tests of virulence of *Bipolaris* and *Exserohilum* isolates were prepared by washing conidia from 7- to 10-day-old cultures on PDA for *Bipolaris* species or on lactose-casein hydrolysate agar (25) for *E. turcicum* Conidial suspensions were filtered through four layers of cheesecloth The conidial suspensions were sprayed onto corn seedlings in the four- to six-leaf stage with a DeVilbiss atomizer attached to an air pump and the inoculated plants were incubated overnight in a moist chamber Atomizers were rinsed with 70% ethanol between inoculations For isolates of *Bipolaris* species, seedlings of Pioneer Brand 3369A were inoculated in the initial virulence tests Isolates of *B. maydis* also were inoculated onto seedlings of B73 with normal cytoplasm cms-C cms-S or cms-T Isolates of *E. turcicum* were inoculated onto seedlings of Pioneer Brand 3065 which lacks the *Ht1* gene for resistance to northern leaf blight and Pioneer Brand 3535 which has the *Ht1* gene Disease reactions were evaluated after 1 wk for *Bipolaris* species and after 2 wk for *E. turcicum*

Mating types of isolates of *B. zeicola* and *B. maydis* were determined by pairing each isolate with known tester isolates of MAT1-1 and MAT1-2 (formerly designated mating types A and a, respectively) of each species Mycelial plugs for paired isolates from 5- to 7-day-old cultures on PDA were placed on opposite sides of autoclaved 1-cm-diameter disks of senescent corn leaves on modified Sachs agar (4) in petri dishes Pseudothecia formed within 7 days in fertile combinations and the pseudothecia were crushed and examined for asci and ascospores after 21 days

Mating types of isolates of *E. turcicum* and *E. holmii* (Luttrell) Leonard & Suggs (teliomorph *Setosphaeria holmii* (Luttrell) Leonard & Suggs) were determined by pairing mycelial plugs from PDA cultures of unknown isolates and known mating type testers approximately 1 cm apart on sterilized barley grains on modified Sachs agar in petri dishes Pseudothecia of *S. turcica* if they formed were crushed and examined for asci and ascospores after 20–25 days Pseudothecia of *S. holmii* were examined after 18 days

RESULTS

Incidence. *B. zeicola* occurred at high frequencies in leaves of corn plants in nine of the 10 extensively sampled fields (Table 1) even though the leaves were collected independently of the presence of leaf spot symptoms *B. maydis* and *E. turcicum* were more erratic in their distribution and occurred at lower frequencies than *B. zeicola* at the time of sampling from mid-July to early August Surprisingly *B. sorokiniana* (Sacc in Sorok) Shoemaker, a pathogen of barley and wheat, was present in corn leaves from six of the 10 fields In two of the fields, the incidence of *B. sorokiniana* was greater than that of either *B. maydis*

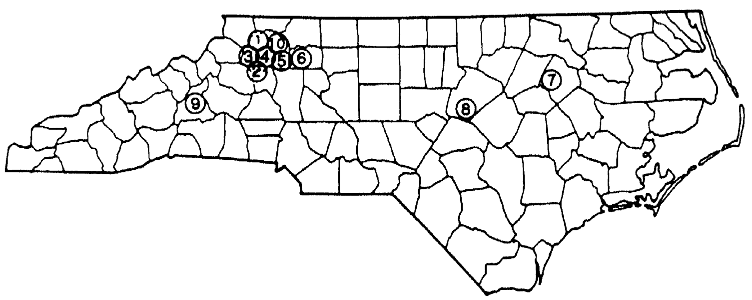

Fig 1 Locations of 10 cornfields in North Carolina that were extensively sampled for incidence of infections of large-spored species of *Bipolaris* and *Exserohilum* in 1985

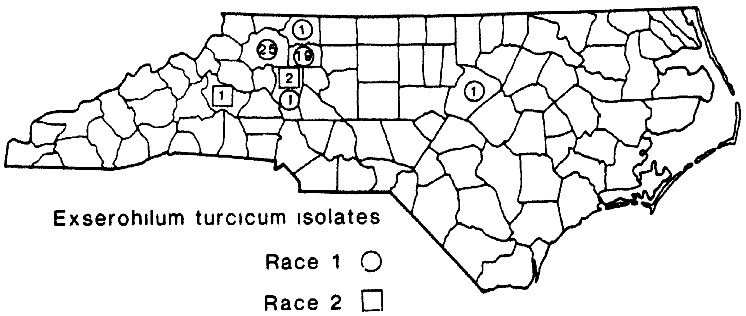

Fig. 2. Distribution of race 1 and race 2 of *Exserohilum turcicum* isolates collected from cornfields in North Carolina in 1985 Numbers within the circles and squares indicate the number of isolates of race 1 and race 2, respectively, obtained in the indicated county

or *E. turcicum*.

An unidentified species of *Bipolaris* was found in corn leaves from two fields, one in western and one in eastern North Carolina. The unidentified species was morphologically similar to *B. zeicola* but was completely infertile in attempted matings with mating type testers of *B. zeicola*. Furthermore, isolates of the unidentified species induced either no visible symptoms or only a few small flecks on inoculated leaves of corn seedlings. The unidentified species also was distinguished from *B. zeicola* in that more than 75% of the isolates of this species failed to grow or grew very poorly in PDA amended with 100 μg/ml of carboxin, whereas less than 10% of *B. zeicola* isolates were severely inhibited by carboxin at 100 μg/ml. All isolates of *B. sorokiniana*, which is morphologically distinct from the unidentified species, also failed to grow on PDA with carboxin at 100 μg/ml.

E. rostratum (Drechs.) Leonard & Suggs, a species known to have a broad host range, was found in corn leaves from five of the 10 fields. Incidence of *E. rostratum* was low except in field 8 in Wake County in eastern North Carolina. One of the isolates from field 8 produced extremely long conidia, averaging 274 × 15.6 μm, on the incubated leaf. This isolate fits within the range described for *E. longirostratum* (Subram.) Sivanesan (24), but Leonard and Suggs (17) regarded *E. longirostratum* as probably a variant of *E. rostratum*. Isolates of *E. rostratum* induced small, round necrotic lesions on leaves of corn seedlings similar to, though generally smaller than, those induced by race 2 of *B. zeicola*.

E. holmii, a leaf blight pathogen of the wild grass *Dactyloctenium aegyptium* (L.) Richter (20), occurred in corn leaves only from the field in McDowell County in western North Carolina. Isolates of *E. holmii* induced either no visible symptoms or only small flecks on inoculated leaves of corn seedlings.

Virulence. The frequencies of race 2 and race 3 of *B. zeicola* in the extensively sampled fields have been described briefly (8) and will be reported elsewhere in greater detail. All of the 75 *B. maydis* isolates from these fields were race O. None showed any greater virulence to any of the cms lines of B73 than to B73 with normal cytoplasm.

None of the leaves collected from 16 to 28 July had northern leaf blight lesions (Table 1). In fact, only one northern leaf blight lesion (found in field 6 in Yadkin County) was observed in any of the fields sampled during that time. Thus, the isolates of *E. turcicum* from leaves collected on 16 July from fields in Wilkes and Yadkin counties represented latent infections in the early stages of an epidemic that became apparent a few weeks later (16).

On 8 August, in response to calls from extension agents in these counties, we returned to Wilkes and Yadkin counties to obtain additional leaf samples. In field 10, we collected 42 leaves without regard to symptoms and found 50% incidence of *E. turcicum*. In addition, we obtained seven more isolates from another field in Wilkes County and five fields in Yadkin County. Other leaf samples with northern leaf blight lesions were sent by county agents to the North Carolina State University Plant Disease and Insect Clinic.

Of the 50 isolates of *E. turcicum* that we obtained, 47 were race 1 and only three were race 2. The three isolates of race 2 were collected on hybrids with the *Ht1* gene for resistance on 28 and 29 August (Table 2). All isolates of *E. turcicum* except the last isolate collected in September were from western North Carolina (Fig. 2).

Mating type. Mating type frequencies for isolates of *B. maydis* and *B. zeicola* from fields with more than 10% incidence of *B. maydis* are shown in Table 3. Both mating types of *B. maydis* were found in all of the fields from which more than five isolates of *B. maydis* were obtained. Frequencies of MAT1-1 in *B. maydis* varied among the five fields with greater than 10% incidence of *B. maydis*. In these fields, the frequency of MAT1-1 was not correlated with the frequency of MAT1-1 in *B. zeicola* ($r = -0.142$). In three of the five fields, both race 2 and race 3 of *B. zeicola* occurred at greater than 10% incidence. In those three fields, the frequencies of MAT1-1 in race 2 were significantly correlated with the frequencies of MAT1-1 in race 3 ($r = 0.954$). Mating types could be determined for only 12 of the 50 *E. turcicum* isolates. Three race 1 isolates were mating type A and seven were mating type a. Two of the three race 2 isolates were fertile and were mating type a. All six of the *E. holmii* isolates collected from field 9 were fertile, three were mating type A and three were mating type a. Isolates of *E. rostratum* were not tested for mating type because previous experience had shown that fertility is rare in this species (12).

DISCUSSION

The high incidence of *B. zeicola* in cornfields may seem inconsistent with its relative lack of importance as a pathogen of corn. Although *B. zeicola* is generally associated with corn production, it causes little or no yield loss (3). Race 1 of *B. zeicola*, which can cause severe losses on inbred lines or hybrids of corn sensitive to its toxin (27), has rarely been found since the toxin-sensitive genotypes were removed from corn breeding programs (14). Fisher et al (3) found no yield loss in 19 hybrids inoculated with race 2 of *B. zeicola*, but *B. maydis* race O and *E. turcicum* caused yield losses of 0.40 and 0.23%, respectively, for each 1% disease severity at 3–4 wk after mid-silk stage (3).

The high incidence of *B. zeicola* in mid-

Table 1. Incidence of *Bipolaris* and *Exserohilum* species[a] in leaves of *Zea mays* collected from 10 cornfields in North Carolina in 1985

County	Field[b] no.	Date	Incidence (%)[c]						
			Bz	Bm	Bs	B-	Et	Er	Eh
Wilkes	1	7/16	63	22	3	0	0	0	0
Wilkes	2	7/16	70	13	0	0	0	5	0
Wilkes	3	7/16	67	2	4	0	4	2	0
Wilkes	4	7/16	81	3	0	0	0	0	0
Wilkes	5	7/16	41	0	20	0	2	0	0
Yadkin	6	7/16	77	5	37	0	18	3	0
Edgecombe	7	7/24	66	22	0	0	0	0	0
McDowell	9	7/25	11	12	9	25	0	11	11
Wake	8	7/28	62	48	2	3	0	32	0
Wilkes	10	8/8	95	5	0	0	50	0	0

[a] Bz = *Bipolaris zeicola*, Bm = *B. maydis*, Bs = *B. sorokiniana*, B- = unidentified *Bipolaris* sp., Et = *Exserohilum turcicum*, Er = *E. rostratum*, Eh = *E. holmii*.

[b] See Figure 1 for locations.

[c] Percentage of leaves from which the indicated species was isolated. Sample sizes were 60 leaves each in fields 1–9 and 42 leaves in field 10. Leaves were collected at 2-m intervals in fields without regard to symptoms.

Table 2. Pathogenic races of *Exserohilum turcicum* isolated from leaves of *Zea mays* in North Carolina in 1985

County	Date	Number of isolates	
		Race 1	Race 2
Wilkes[a]	7/16	3	0
Yadkin[b]	7/16	11	0
Wilkes[c]	8/8	22	0
Yadkin[d]	8/8	6	0
Surry	8/12	1	0
Yadkin[d]	8/19	2	0
Iredell	8/28	1	2
Burke	8/29	0	1
Wake	9/4	1	0
Total		47	3

[a] Two isolates from field 3, one from field 5 (see Table 1).

One isolate from unnumbered field, 21 from field 10.

[d] Isolates from four unnumbered fields in Yadkin County.

July can be accounted for only by high levels of initial inoculum. Evidently, *B. zeicola* is better adapted than either *B. maydis* or *E. turcicum* for survival during its saprophytic phase in corn crop debris under the environmental conditions in North Carolina. As the growing season progresses, *B. maydis* and *E. turcicum* populations overtake those of *B. zeicola* because of their greater capacity for reproduction on living corn leaves. The lesions induced by *B. zeicola* are small and sporulation generally does not occur in them until the leaves senesce (19). Conidia of *B. zeicola*, however, are capable of infecting corn leaves in all stages of the plant's development. Thus, *B. zeicola* is effective in establishing itself in living corn leaves before they can be invaded by saprophytes, but it causes little damage to the plant. In this way, *B. zeicola* occupies an ecological niche between that of an aggressive parasite and that of an opportunistic saprophyte.

To be successful in the niche to which it has adapted, *B. zeicola* must sporulate profusely after the infected leaves senesce and must compete strongly with other microorganisms to maintain itself in the dead host tissue until the next growing season. The success of *B. zeicola* is enhanced by its ability to invade and survive in stalks as well as leaves of corn (1). In the comparison of *B. zeicola* with *B. maydis*, it appears that by adapting to increasingly aggressive parasitism, the pathogen sacrifices part of its ability to survive saprophytically, and vice versa.

Wheat and corn often are grown in adjacent small fields in North Carolina or in the same fields in successive seasons in a corn, winter wheat, soybean rotation, so it is not so surprising that conidia of *B. sorokiniana* can be found in cornfields. Kline and Nelson (6,21) showed that *B. sorokiniana* has a broad host range, but their isolates were avirulent to corn seedlings inoculated in the greenhouse. Our results show that *B. sorokiniana* can infect adult corn plants in the field. In field 6 in Yadkin County, the incidence of *B. sorokiniana* in corn leaves was 37%. This indicates that most of the corn plants must have carried *B. sorokiniana* infections in at least one leaf. Such an abundance of the pathogen in corn leaves would provide significant initial inoculum for infection of a wheat crop in the field in a corn, winter wheat, soybean rotation.

Northern leaf blight, as its name suggests, is not a common problem of corn in the Southeast. The disease occurs sporadically in North Carolina, with epidemics at infrequent intervals of up to 10 yr. Because temperatures are cooler in the mountains and western Piedmont than in eastern North Carolina, *E. turcicum* is found more frequently in corn in the western counties, as it was in the 1985 survey. In contrast, southern leaf blight caused by *B. maydis* is common in North Carolina and tends to be more severe in eastern than in western North Carolina (11). In 1985, *B. maydis* incidence appeared to be slightly greater in the two extensively sampled fields in eastern North Carolina than in the western fields, but the differences were not significant.

E. rostratum has a very broad host range and has often been reported in leaves of corn and other gramineous species (12,29). Its pathogenicity to corn resembles that of *B. zeicola*, but the incidence of *E. rostratum* in corn leaves in North Carolina in 1985 was much lower than that of *B. zeicola*. It seems likely that *E. rostratum* is less efficient than *B. zeicola* in infecting green leaves of adult corn plants. Like *B. zeicola*, *E. rostratum* can invade the stalks of mature corn plants (7). This undoubtedly enhances its ability to survive saprophytically and produce abundant inoculum for the following growing season.

The presence of *E. holmii* in leaves of corn plants in field 9 in McDowell County suggests that *Dactyloctenium*, the known host of *E. holmii*, must have been abundant around the edges of that cornfield. We suspect that the unidentified *Bipolaris* species found in field 9 is probably also a pathogen of a grass that was common at the edges of the field.

The presence of *E. holmii*, *B. sorokiniana*, and the unidentified *Bipolaris* species in leaves of adult corn plants at incidences as high as 37% raises the possibility that the corn pathogens *B. maydis* and *E. turcicum* may also invade leaves of grasses and small grains in the vicinity of cornfields. Dead leaves of these plants might serve as sources of inoculum for corn during the following growing season.

In virulence tests, we found no evidence of a race virulent on corn with cms-C as reported in China (28). Whereas this information is welcome in terms of the prospects for hybrids with cms-C in the immediate future, it does not ensure continued safe use of cms-C in the production of future hybrids in the United States. Evidence from the epidemic caused by race T of *B. maydis* in 1970 shows that such a race of *B. maydis* could arise and spread throughout the corn-growing areas of North America within a few years if susceptible hybrids were to be extensively grown (15). On the other hand, the evidence of race T also indicates that such a race will probably not arise if susceptible hybrids are not grown in the United States. We found no race T isolates among the 75 isolates of *B. maydis* that we collected in 1985. This is consistent with earlier reports that race T quickly disappeared from the *B. maydis* population of the Southeast after the use of cms-T in the production of hybrid corn seed was abandoned (13,15).

As we reported earlier (16), our evidence indicates that the 1985 epidemic of northern corn leaf blight in western North Carolina was caused by race 1 of *E. turcicum*. Although race 2 did eventually increase on corn hybrids with the gene *Ht1* for resistance to race 1, there was no significant damage from race 2. Either the amount of initial inoculum of race 2 was extremely low early in 1985 or the genotypes of race 2 present are less aggressive than those of race 1.

Pedersen and Brandenburg (23) reported that 27 of 65 isolates of race 2 of *E. turcicum* collected over a wide geographical area from Delaware to Iowa were mating type a and that the other 38 isolates failed to form pseudothecia in any mating tests. We also had difficulty inducing pseudothecial production in mating type tests. Only two of our three race 2 isolates were sufficiently fertile to identify mating types; both were mating type a. Although the evidence is not conclusive, the situation seems similar to that in race T of *B. maydis* in which a single mating type predominated in the new race of the pathogen, particularly in the Corn Belt and northeastern United States (10). More fertile testers of *E. turcicum* must be identified so that the mating types of a higher frequency of field isolates can be determined. If, indeed, all isolates of race 2 of *E. turcicum* are mating type a, that would indicate that hybridization between race 1 and race 2 has rarely, if ever, occurred in the field. On the basis of that possibility and the possibility that

Table 3. Mating type[a] frequencies among isolates of *Bipolaris zeicola* and *B. maydis* isolates collected from leaves of *Zea mays* in five cornfields[b] in North Carolina in 1985

			Frequency of MAT1-1 (%)			
			B. zeicola			
County	Field no.	Date	Race 2	Race 3	Total	*B. maydis*
Wilkes	1	7/16	24	38	28	23
Wilkes	2	7/16	76	67	71	29
Edgecombe	7	7/24	22	46	31	62
McDowell	9	7/25			83	43
Wake	8	7/28			59	28
Mean			40.7	50.3	49.4	34.8

[a] MAT1-1 and MAT1-2 were formerly designated as A and a, respectively.
[b] Only fields with greater than 10% incidence of *B. zeicola* and *B. maydis* in the leaves are included; fields 8 and 9 had less than 5% incidence of race 3 of *B. zeicola*.

race 2 is less aggressive than race 1, it is important that every precaution should be taken to prevent laboratory recombinants between the two races from escaping to the field.

Frequencies of the two mating types of *B. zeicola* and *B. maydis* fluctuate from year to year and from field to field, but with the exception of the first years of occurrence of race T of *B. maydis* in the United States, both mating types can be found at reasonably high frequencies in any population of either species. The persistence of the mating type polymorphism is typical of systems of frequency-dependent selection. In stable polymorphisms protected by frequency-dependent selection, whichever allele is less frequent in the population tends to have a selective advantage. Frequency-dependent selection would occur if the sexual stage was essential to survival of these fungi, but there is no evidence that pseudothecia and ascospores are even common in nature, much less essential.

Thus, it appears that some other type of frequency-dependent or balancing selection must account for the persistence of the mating type polymorphism in *B. zeicola* and *B. maydis*. Although the evidence from just three fields is too limited to be convincing, there is some indication that within fields, selection may affect mating types in races 2 and 3 of *B. zeicola* similarly. At least, the frequencies of mating types of races 2 and 3 appeared to be correlated, whereas the frequencies of mating types over both races of *B. zeicola* were not correlated with mating type frequencies in *B. maydis* in the same fields. This suggests that putative factors responsible for frequency-dependent selection of mating types differ for different species of *Bipolaris*. Lodge and Leonard (19) found a general similarity of mating type frequencies in races 2 and 3 of *B. zeicola* over rather broad geographical regions of North Carolina. Further comparisons of mating type frequencies of races 2 and 3 of *B. zeicola* in samples from other fields sampled in 1985 will be reported in a separate paper.

LITERATURE CITED

1. Anderson, B., and White, D. G. 1987. Fungi associated with cornstalks in Illinois in 1982 and 1983. Plant Dis. 71:135-137.
2. Berquist, R. R., and Masias, O. R. 1974. Physiologic specialization in *Trichometasphaeria turcica* f. sp. *zeae* and *T. turcica* f. sp. *sorghi* in Hawaii. Phytopathology 64:645-649.
3. Fisher, D. E., Hooker, A. L., Lim, S. M., and Smith, D. R. 1976. Leaf infection and yield loss caused by four Helminthosporium leaf diseases of corn. Phytopathology 66:942-944.
4. Hebert, T. T. 1971. The perfect stage of *Pyricularia grisea*. Phytopathology 61:83-87.
5. Jordan, E. G., Perkins, J. M., Schall, R. A., and Pedersen, W. L. 1983. Occurrence of race 2 of *Exserohilum turcicum* on corn in the central and eastern United States. Plant Dis. 67:1163-1165.
6. Kline, D. M., and Nelson, R. R. 1963. Pathogenicity of isolates of *Cochliobolus sativus* from cultivated and wild gramineous hosts from the Western Hemisphere to species of the Gramineae. Plant Dis. Rep. 47:890-894.
7. Kucharek, T. A. 1973. Stalk rot of corn caused by *Helminthosporium rostratum*. Phytopathology 63:1336-1338.
8. Leath, S., and Leonard, K. J. 1986. Distribution of race 3 of *Cochliobolus carbonum* on corn in North Carolina. Plant Dis. 70:800.
9. Leonard, K. J. 1972. *Helminthosporium* spp. as laboratory contaminants on incubated leaf specimens. Plant Dis. Rep. 56:834-836.
10. Leonard, K. J. 1973. Association of mating type and virulence in *Helminthosporium maydis*, and observations on the origin of the race T population in the United States. Phytopathology 63:112-115.
11. Leonard, K. J. 1974. Foliar pathogens of corn in North Carolina. Plant Dis. Rep. 58:532-534.
12. Leonard, K. J. 1976. Synonomy of *Exserohilum halodes* with *E. rostratum*, and induction of the ascigerous state, *Setosphaeria rostrata*. Mycologia 68:402-411.
13. Leonard, K. J. 1977. Races of *Bipolaris maydis* in the southeastern U.S. from 1974-1976. Plant Dis. Rep. 61:914-915.
14. Leonard, K. J. 1978. Polymorphisms for lesion type, fungicide tolerance, and mating capacity in *Cochliobolus carbonum* isolates pathogenic to corn. Can. J. Bot. 56:1809-1815.
15. Leonard, K. J. 1987. The host population as a selective factor. Pages 161-179 in: Populations of Plant Pathogens: Their Dynamics and Genetics. M. S. Wolfe and C. E. Caten, eds. Blackwell Scientific Publications, Oxford, England.
16. Leonard, K. J., and Leath, S. 1986. Evidence that race 1 of *Setosphaeria turcica* caused the 1985 northern corn leaf blight epidemic in North Carolina. Plant Dis. 70:981.
17. Leonard, K. J., and Suggs, E. G. 1974. *Setosphaeria prolata*, the ascigerous state of *Exserohilum prolatum*. Mycologia 66:281-297.
18. Lipps, P. E. 1982. *Exserohilum turcicum* virulent on corn with the *Ht* resistance gene in Ohio. Plant Dis. 66:397-398.
19. Lodge, D. J., and Leonard, K. J. 1984. A cline and other patterns of genetic variation in *Cochliobolus carbonum* isolates pathogenic to corn in North Carolina. Can. J. Bot. 62:995-1005.
20. Luttrell, E. S. 1963. A *Trichometasphaeria* perfect stage for a *Helminthosporium* causing a leaf blight of *Dactyloctenium*. Phytopathology 53:281-285.
21. Nelson, R. R., and Kline, D. M. 1961. The pathogenicity of certain species of *Helminthosporium* to species of the Gramineae. Plant Dis. Rep. 45:644-648.
22. Nelson, R. R., and Kline, D. M. 1966. The pathogenicity of 91 morphologically similar isolates of *Helminthosporium* to 30 gramineous species. Plant Dis. Rep. 50:382-384.
23. Pedersen, W. L., and Brandenburg, L. J. 1986. Mating types, virulence, and cultural characteristics of *Exserohilum turcicum* race 2. Plant Dis. 70:290-292.
24. Subramanian, C. V. 1956. Hyphomycetes II. J. Indian Bot. Soc. 35:446-449.
25. Tuite, J. 1969. Plant Pathological Methods. Burgess Publishing Co., Minneapolis, MN. 239 pp.
26. Turner, M. T., and Johnson, E. R. 1980. Race of *Helminthosporium turcicum* not controlled by *Ht* genetic resistance in corn in the American corn belt. Plant Dis. 64:216-217.
27. Ullstrup, A. J. 1944. Further studies on a species of *Helminthosporium* parasitizing corn. Phytopathology 34:214-222.
28. Wei, J.-K., Liu, K.-M., Chen, J.-P., and Lee-Stadelmann, O. Y. 1988. Pathological and physiological identification of race C of *Bipolaris maydis* in China. Phytopathology 78:550-554.
29. Whitehead, M. D., and Calvert, O. H. 1959. *Helminthosporium rostratum* inciting ear rot of corn and leaf-spot of thirteen grass hosts. Phytopathology 49:817-820.