Occurrence of *Fusarium* Species in Symptom-free and Overwintered Cornstalks in Northwestern Minnesota

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ABSTRACT

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Incidence of stalk rot in 50 cornfields sampled in early October 1985 and 1986 averaged 1 and 2%, respectively. However, six *Fusarium* species were isolated from cornstalks without stalk rot symptoms. *F. subglutinans* predominated and colonized 30% of stalks in 1985 and 86% in 1986; *F. graminearum* colonized 20 and 37% for the 2 yr; *F. proliferatum*, 5 and 13%; *F. avenaceum*, 3% both years; *F. culmorum*, 1 and 3%; and *F. moniliforme*, 1 and 2%. *F. graminearum* was isolated from 26% of corn debris collected from 14 fields in 2 yr, *F. avenaceum* from 12%, and *F. culmorum* from 4%. Incidence of stalk rot (or infected stalks) in fields containing carryover stalk debris was the same as that in fields where debris was absent. Isolates of *F. graminearum* were in group 2 in that perithecia were produced by 99–100% of the isolates from cornstalks and 1-yr-old stalk debris.

Additional keywords: *Gibberella zeae, Zea mays*

Northwestern Minnesota is an important cereal production area. During 1982–1986, annual averages of 662,000 ha of wheat (*Triticum aestivum* L.) and 337,000 ha of barley (*Hordeum vulgare* L.) were grown in this region and represented 58% of the wheat and 78% of the barley grown in the state. With the development of short-season (75–85 days) corn (*Zea mays* L.) hybrids, more and more corn is being grown in northwestern Minnesota. During 1975–1976, for example, 44,000 ha were planted to corn, whereas 82,200 ha were planted to corn during 1985–1986. Yet corn in this area currently represents only 3% of the area planted to corn in the state (Minnesota Agriculture Statistics Service, St. Paul). Sutton (10) reported that corn residues served as a significant source of scab inoculum in small-grain-growing areas in Canada. Scab of wheat and barley has been observed to be more prevalent in northwestern Minnesota in recent years (M. Johnson, *personal communication*), although any relation with increasing corn culture has not been established.

The most frequently reported causes of stalk rot of corn are *F. graminearum* Schwabe, *F. moniliforme* Sheldon, and *F. subglutinans* (Wollenw. & Reinking) Nelson, Toussoun & Marasas (1,5); *F. proliferatum* (Matsushima) Nirenberg was added to this list in 1987 (4). The fungi causing scab (head blight) of wheat and barley are *F. graminearum, F. culmorum* (W. G. Smith) Sacc., and *F. avenaceum* (Fr.) Sacc. (7,11). Because *F. graminearum* can cause stalk rot of corn and scab of small grains, especially wheat (10,12), as well as seedling blight, it is important wherever cereal crops are grown. Francis and Burgess (2) separated this species into two populations: Group 1 isolates are mainly soilborne, rarely form perithecia, and cause crown rot of cereals, whereas group 2 isolates are mostly airborne, form perithecia of *Gibberella zeae* (Schw.) Petch, and cause stalk and cob rot of corn, scab of small grains, and stub dieback of carnations.

Our objectives were to: 1) ascertain the populations of *Fusarium* species in living and residual stalks in northwestern Minnesota, 2) determine the incidence of stalk rot in the presence and absence of stalk residue in that part of the state, and 3) determine the population group of *F. graminearum*. A preliminary report of this study has been published (15).

MATERIALS AND METHODS

Stalk rot and occurrence of *Fusarium* species were evaluated in 50 randomly chosen fields in 12 counties of northwestern Minnesota in 1985 and 1986 (Fig. 1). Of these fields, 28 were examined from 30 September to 2 October 1985 and 22 on 6 and 7 October 1986. Incidence of stalk rot was determined in each field on 300 plants (100 plants in each of three randomly selected rows) by displacing each stalk about 30 cm from the vertical. Stalks were considered rotten if they were free from corn borers and broke at the lower nodes.

To study the incidence of *Fusarium* species, samples of tissue were collected from 15–25 cornstalks chosen at random and free from external symptoms of stalk rot in each of the three rows used for stalk rot evaluations. Samples were collected with a bark increment hammer from the first internode above the brace roots (6). Cores (15–25 × 4 mm) of stalk tissue were placed in plastic bags and stored at 5 C for 2–3 days until analyzed for *Fusarium* species. The assays were done with 50 randomly selected cores per field. The cores were surface-treated in 0.5% NaOCl for 30 sec, drained on paper towels, and cultured on pentachloronitrobenzene (PCNB) agar supplemented with chlortetracycline HCl (0.05 g/L), a medium selective for *Fusarium* species (8).

Cornstalk debris of the previous crop

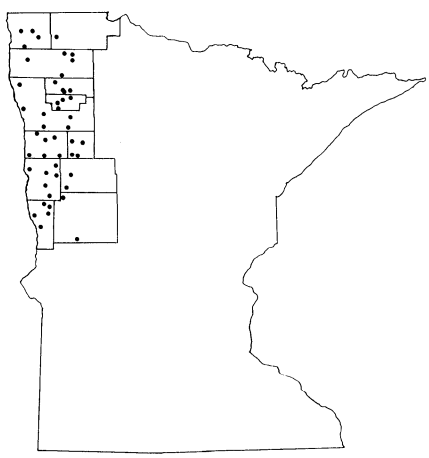

Fig. 1. Sites sampled for stalk rot and *Fusarium* in 1985 and 1986; 300 cornstalks were tested for breakage by the "push" test, and 50 samples were collected for identification to *Fusarium* species per field. One circle sometimes represents two or more adjacent fields.

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Table 1. Incidence of stalk rot and *Fusarium* species generally known as pathogens of cereals in cornfields where cornstalk residues were absent or present in 1985 and 1986

Corn residues		No. of fields	Percent stalk rot[a]		Percent cornstalks with *Fusarium*:[b]											
					subglutinans[c]		*graminearum*		*proliferatum*		*avenaceum*		*culmorum*		*moniliforme*	
			$\bar{x}$	Range	$\bar{x}$	Range	$\bar{x}$	Range	$\bar{x}$	Range	$\bar{x}$	Range	$\bar{x}$	Range	$\bar{x}$	Range
Absent	1985	17	<1	0–0.7	25	0–60	19	4–36	5	0–19	3	0–22	2	0–8	2	0–8
	1986	16	2.2	0–5.3	86	82–90	34	6–86	12	0–36	2	0–10	4	0–22	2	0–8
Present	1985	11	<1	0–0.7	39	0–74	21	0–40	5	0–16	3	0–6	<1	0–10	<1	0–4
	1986	6	1.2	0–4.3	···	···	42	16–66	17	6–32	5	0–18	1	0–6	2	0–10

[a] Three hundred cornstalks were tested per field.
[b] In 1985, based on isolates from 845 cornstalks from fields without residues and 545 cornstalks from fields with residues; in 1986, based on isolates from 800 cornstalks from fields without residues and 300 cornstalks from fields with residues.
[c] Based on 250 cornstalks collected from five fields without residues in 1986.

(1 yr old) was present in 17 of the 50 cornfields examined. Samples were collected from 14 of the 17 fields. Twenty to 100 basal cornstalk pieces were removed from the soil surface or pulled from near the soil surface in each of eight fields in 1985 and six in 1986. Samples were placed in paper bags and stored at room temperature for 5–11 wk until assayed. Because of advanced rotting of pith tissue, each stalk residue fragment was trimmed with pruning shears to the lowermost (basal) stem internode, about 1 cm^3, which still retained its integrity. The basal-stem internode fragments were surface-treated in 1% NaOCl for 30 sec, drained on paper towels, and placed on PCNB agar. The deterioration of residue was caused by a variety of organisms that inhabit soil and plant debris and not necessarily by stalk rot fungi.

Fusarium cultures were grown on PCNB agar under a combination of fluorescent (four General Electric or Sylvania 40W tubes) and black lamps (one or two Sylvania 40W tubes, BLB series) for a 12-hr photoperiod (5,300 lx) for 1–2 wk. Cultures were transferred to homemade and acidified potato-dextrose agar (PDA), and *Fusarium* species were identified 10–14 days later (9). Some cultures also were transferred to carnation leaf agar (CLA) to stimulate sporulation or to facilitate examination of phialides.

To test for the presence of group 1 and 2 populations of *F. graminearum*, cultures were placed on CLA (9). Cultures mixed with other *Fusarium* species were hyphal-tipped (four to six per culture) before being transferred to CLA. After 2–9 wk, cultures were examined for perithecia of *G. zeae*, then 25% of the isolates from each field were checked for mature ascospores.

RESULTS

Stalk rot incidence. Stalk rot incidence was negligible in cornfields with and without cornstalk residues in early October of 1985 and 1986 (Table 1). Although data are not given for individual fields, we observed stalk rot in 12% of the fields without corn residues and in 18% of the fields with corn residues in 1985. In 1986, stalk rot occurred in 75% of the fields without corn residues and in 50% of the fields with corn residues.

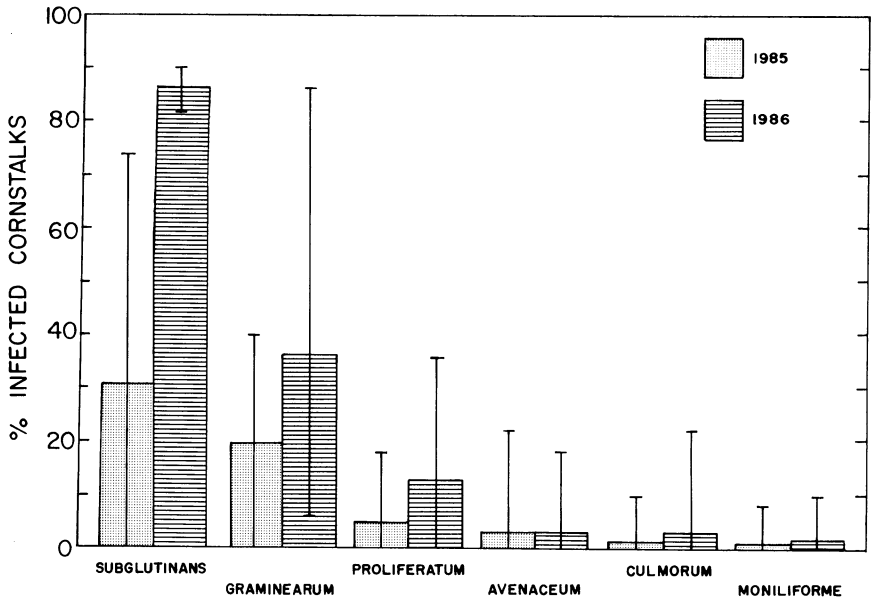

Fig. 2. Percent cornstalks from which *Fusarium* species were recovered in early October 1985 and 1986; 1,390 cornstalks were assayed in 1985 and 1,100 in 1986. Line within each bar represents minimum and maximum occurrences among fields per year.

***Fusarium* in symptom-free stalks.** Of the 1,390 cornstalks sampled in 1985, 72% yielded *Fusarium* (often with more than one species per stalk), and each of the 1,635 isolates was identified to species. Of the 1,100 cornstalks sampled in 1986, 72% yielded *Fusarium*, and 1,746 isolates were identified to species. In five of 22 fields sampled in 1986, *F. subglutinans* and *F. oxysporum* Schlecht. emend. Snyd. & Hans. were identified; these two species were not distinguished (based on presence or absence of polyphialides or chlamydospores) in the other 17 samples.

Six *Fusarium* species known to be pathogenic to corn and other cereals were isolated from symptom-free cornstalks collected in 1985 and 1986 (Fig. 2). *F. subglutinans* was isolated from the greatest percentage of stalks in 1985 and 1986, averaging 30 and 86%, respectively, followed by *F. graminearum* (20 and 37%), *F. proliferatum* (5 and 13%), *F. avenaceum* (3% both years), *F. culmorum* (1 and 3%), and *F. moniliforme* (1 and 2%). *F. graminearum* occurred in a slightly greater percentage of cornstalks when cornstalk debris was present, but fields with and without residues overlapped in ranges of the percentage of cornstalks infected by this fungus and the other five species (Table 1).

Other *Fusarium* species isolated from cornstalks in 1985 and 1986 included: *F. equiseti* (Corda) Sacc. sensu Gordon (18 and 50%), *F. acuminatum* Ell. & Ev. sensu Gordon (16 and 8%), *F. sporotrichioides* Sherb. (12 and 10%), *F. oxysporum* (8 and 15%), and *F. solani* (Mart.) Appel & Wollenw. emend. Snyd. & Hans. (1 and 9%). Among species occurring in <1% of the cornstalks were: *F. camptoceras* Wollenw. & Reinking, *F. crookwellense* Burgess, Nelson &

Toussoun, *F. graminum* Corda, *F. poae* (Peck) Wollenw., *F. sambucinum* Fuckel, *F. semitectum* Berk. & Rav., and *F. tricintum* (Corda) Sacc.

***Fusarium* in corn debris.** Identification of *Fusarium* from 1-yr-old corn debris was limited to species known as pathogens of cereals. *F. graminearum* predominated (Fig. 3) and was isolated from 29% of the debris collected in 1985 and from 24% collected in 1986, followed by *F. avenaceum* (5 and 15%) and *F. culmorum* (6 and 3%). Although data are not shown for individual fields, *F. graminearum* was present in debris collected from 100% of the fields sampled. *F. culmorum* was present in debris collected from 50% of the fields in 1985 and from 83% in 1986. *F. avenaceum* was present in debris pieces collected from 75% of the fields in 1985 and from 100% in 1986.

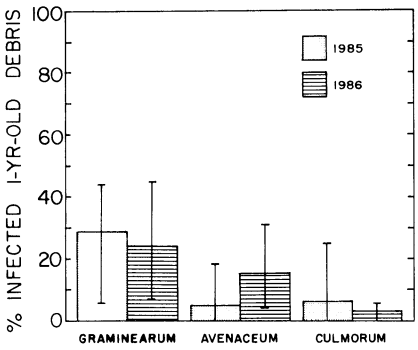

Fig. 3. Percent 1-yr-old stalk residues from which *Fusarium* species were recovered in early October 1985 and 1986; 224 pieces of 1-yr-old debris from eight fields were assayed in 1985 and 401 pieces from six fields in 1986. Line within each bar represents minimum and maximum occurrences among fields per year.

F. graminearum was isolated more frequently from debris than from cornstalks in 1985, but the opposite was true in 1986 (Fig. 4). Correlations between percentage of 1-yr-old debris and cornstalks colonized by *F. graminearum* were not statistically significant ($r = 0.35$ in 1985 and 0.57 in 1986). *F. avenaceum* and *F. culmorum* were isolated more frequently from debris than from cornstalks, but the overall recovery of both species was low.

Perithecial formation. Of the 674 cultures of *F. graminearum* isolated from cornstalks, 327 were tested for perithecial production. Perithecia of *G. zeae* formed in 99.5% of the cultures and bore mature ascospores. Of the 160 cultures of *F. graminearum* isolated from 1-yr-old corn debris, 154 were tested for perithecial production; all formed perithecia and bore mature ascospores.

DISCUSSION

A history of stalk rot of corn caused by *Fusarium* species as well as a history of the incidence of corn infected with *Fusarium* species lacking external and internal symptoms of stalk rot have been developed for southern Minnesota for more than a decade (14). These *Fusarium* species were obviously parasites, sometimes pathogens, but their role in corn plants as endophytes has not been clearly established. Moreover, corn plants of southern Minnesota were 105- to 115-day hybrids growing in an area where corn and soybeans were the predominant crops. Northwestern Minnesota, on the other hand, comprises areas of mainly small grains and other crops in which corn more recently has been introduced because of the development of 75- to 85-day hybrids. The prevalence of wheat scab caused by *F. graminearum* reported in the same area by Wilcoxson et al (12) suggests the possibility of a relationship between corn and small grains in this region with respect to *Fusarium* species as pathogens.

Because different ecological conditions and cropping systems prevail in southern and northwestern Minnesota, results of infection and pathogenicity as well as species of *Fusarium* isolated could be different. For example, *F. culmorum* and *F. avenaceum* were found in low percentages in stalks of northwestern Minnesota but were absent in corn of southern Minnesota. Also, *F. graminearum* appeared early in northwestern Minnesota (by early October) and in greater frequency (20–37% of stalks) compared with southern Minnesota, where it first appeared from mid-September to November and averaged 15% over an 11-yr period (14). At the time of sampling, stalks were greener and more succulent in northwestern Minnesota than they were in southern Minnesota at sampling time, yet *F. graminearum* was isolated more frequently from the corn in northwestern Minnesota, suggesting that infection occurs earlier there. Whether this is attributable to climate, inoculum, or hybrid was not established.

The results found earlier for southern Minnesota (14) concerning the incidence of stalks infected with *Fusarium* species, even when symptoms were absent, were in general similar in northwestern Minnesota despite the differences in climate, soils, and crops. Also, the recovery of *F. graminearum* in 1-yr-old corn debris is similar between the two locations in the state. Harvest delay is a considerable factor in incidence of stalk rot, as colonization by *F. graminearum* increases as tissues senesce (14). During 1983 through 1987, 50% of the corn crop in northwestern Minnesota was harvested between 13 October and 11 November; 1985 was so wet that 25% of the corn was standing by 24 November (Minnesota Agriculture Statistics Service, St. Paul). This delay of harvest in 1985 likely allowed more stalks to become rotted by *Fusarium* species. For example, a field sampled at Crookston on 28 October 1985 had a stalk rot lodging incidence of 35%; *F. graminearum* was isolated from 70% of the stalks, *F. subglutinans* from 62%, *F. proliferatum* from 18%, *F. moniliforme* from 6%, and *F. avenaceum*

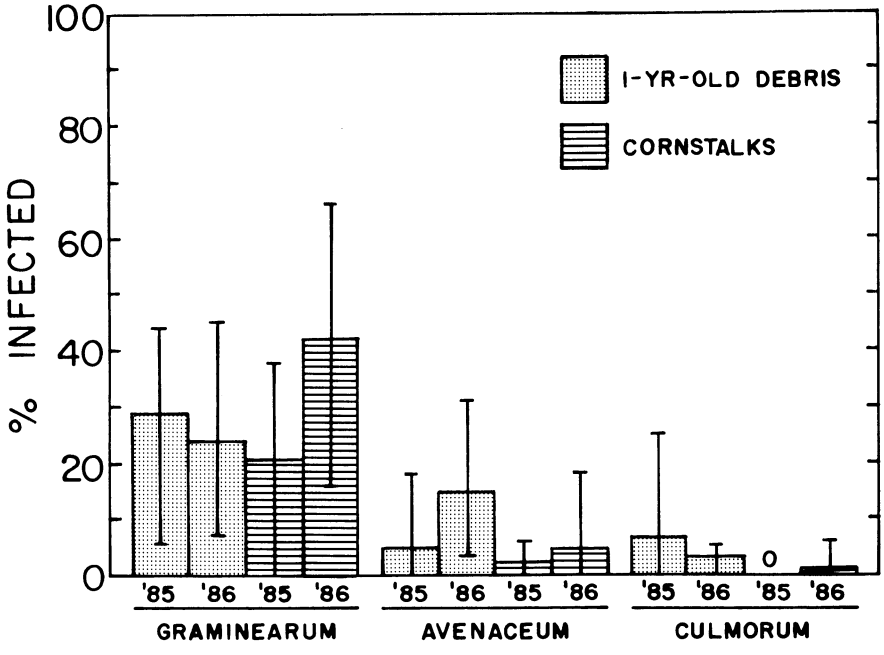

Fig. 4. Recovery of three *Fusarium* species from 1-yr-old debris and cornstalks collected from the same eight fields in 1985 and from the same six in 1986. Line within each bar represents minimum and maximum occurrences among fields per year.

from 2% (C. E. Windels, *unpublished*). This also means that debris carried over to the next season will harbor high concentrations of inoculum.

The Liseola section was revised by Nelson et al (9) in 1983 from one species (*F. moniliforme*) to four species (*F. anthophilum* (A. Braun) Wollenw., *F. moniliforme, F. proliferatum,* and *F. subglutinans*). Identification of *Fusarium* species in this study is based on these revisions, and thus direct comparison to our previous studies (5,6) cannot be made for species in the Liseola section.

The severe epidemic in scab of wheat reported in recent years in northwestern Minnesota attributable to *F. graminearum* (12) together with the increasing culture of corn has raised concerns about both stalk rot and Fusarium scab of wheat, especially when reduced tillage is a growing practice. Presumably, the ascospores of *G. zeae* or the conidia of *F. graminearum* produced on corn residues serve as a source of inoculum (3), although inoculum probably originates also in residues of small grains and grasses. In northwestern Minnesota, crowns of spring wheat (13) and barley (C. E. Windels, *unpublished*) have been found to be colonized by *F. graminearum* group 2.

Our experience at Minnesota in isolating *Fusarium* species from stalks and roots of corn and from heads and grains of wheat consistently yields group 2 isolates of *F. graminearum* (12,14). Pathogenicity tests on corn and wheat seedlings have shown that seedling blight can be induced with these group 2 isolates in the field (T. Kommedahl et al, *unpublished*). It is likely that all *F. graminearum* isolates in Minnesota consist of group 2 isolates.

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