Seedborne *Fusarium moniliforme* and Seedling Infection in Hybrid Sweet Corn

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Research supported in part by the Idaho State Sweet Corn Breeders Association. We thank them for this assistance.
Approved as an Idaho Seed Institute contribution (Research Paper 80721) by the director of the Idaho Agricultural Experiment Station.
Accepted for publication 10 March 1981.

ABSTRACT

Anderegg, J., and Guthrie, J. W. 1981. Seedborne *Fusarium moniliforme* and seedling infection in hybrid sweet corn. Phytopathology 71:1196-1198.

In greenhouse studies, a highly significant positive correlation was found between the percentage of hybrid sweet corn seeds with seedborne *Fusarium moniliforme* and the percentage of crowns of resulting plants containing *F. moniliforme*. Under field conditions at Caldwell, ID, seedling infection was independent of the level of seedborne inoculum, suggesting that infection originated primarily from soilborne inoculum at this location. Under field conditions at Moscow, ID, however, *F. moniliforme* was isolated from seedling tissues more frequently when seed was coated with inoculum of the pathogen than when seed was surface disinfected. Other soil fusaria, as well as the inoculum level, influenced the number and kind of *Fusarium* species isolated from young seedlings.

Fusarium moniliforme Sheld. is a serious pathogen of rice, causing bakanae disease and foot rot, and has been reported to cause foot rot, root rot, ear and cob rots, and seedling blight of many cereals and other crops (5). Seedborne infection by *F. moniliforme* has been reported for 28 host species throughout the world. As early as 1920, Valleau (6) concluded that *F. moniliforme* could be carried within the corn seed; he found a high incidence of seedborne *F. moniliforme* in numerous field corn cultivars, and a high percentage of apparently healthy seeds still contained *F. moniliforme* after surface disinfection.

Some reports suggest that seed infection, unless severe, has little effect on germination or on the vigor of young seedlings (6,7). Kingsland and Wernham (3) were unable to obtain seedling blight under greenhouse conditions when seeds were planted in soil infested with *F. moniliforme*. However, Valleau (6) observed root rot in young seedlings originating from infected kernels, and Foley (2) found that *F. moniliforme* induced root rot of corn when seeds were grown in nutrient solutions. Foley isolated *F. moniliforme* from the epicotyls of young plants. We attempted to determine the relative importance of seedborne and soilborne *F. moniliforme* in early seedling infection.

MATERIALS AND METHODS

Seeds of two commercial sweet corn hybrids, cultivars Iochief and Earlivee, were provided by Crookham Seed Company (Caldwell, ID 83605). The same seed lots were used for all greenhouse and field tests. Fusarium-selective Nash and Snyder medium (4) was used to detect seed infection before and after surface disinfection for 5 min in 10% Clorox (0.5% available chlorine). Seventy-three percent of the Iochief seed was infested with *F. moniliforme* (73 of each 100 seeds tested carried *F. moniliforme* either internally or externally), and 3% contained the fungus after surface disinfection. *F. moniliforme* was isolated from 67% of the Earlivee seed before the Clorox treatment and from 4% after surface disinfection. All seed was stored at −10 C, and neither germination nor percentage of kernels containing *F. moniliforme* changed significantly during the test period.

0031-949X/81/11119603/$03.00/0

F. moniliforme (both externally and internally borne) was assayed by plating seeds on Nash and Snyder medium, 10 seeds per 100 × 15 mm petri dish. After incubation for 7 days at 20 C with 12 hr of near-ultraviolet light and 12 hr of darkness per day, dishes were examined for colonies of *Fusarium* spp.

In addition, presence of the pathogen within the seed was detected by the soak-freeze method: seed subsamples obtained with a Precision divider (Gamet Mfg. Co., Minneapolis, MN 55423) were pretreated for 5 min in 10% Clorox, soaked for 24 hr in 20 ml of distilled water in a petri dish with five pieces of filter paper (Easton-Dikeman grade 641; Van Water and Rogers, Seattle, WA 98124), frozen for 24 hr, and then placed 10 seeds per dish on potato-dextrose agar (PDA) (Difco Laboratories, Detroit, MI 48201). Dishes were incubated at 20–22 C with 12 hr of near-ultraviolet light per day in a Stults germinator (Stults Scientific, Springfield, IL 62703) for 5 days. On the seventh day, dishes were examined for colonies of *Fusarium* spp.

All tissue sections (crowns, stems, and roots) collected from seedlings were surface disinfected for 5 min in Clorox, ethanol, and distilled water (1:1:8) and plated on Nash and Snyder medium. All *Fusarium* colonies were transferred to PDA before species identification. Representative cultures were identified or confirmed by the Fusarium Research Center (Department of Plant Pathology, The Pennsylvania State University).

In greenhouse tests, seeds of Iochief and Earlivee were treated and planted in 38 × 60 × 8 cm flats of Idaho soil mix #1 (1) containing no resident *Fusarium* population. Any resident inoculum was eradicated from the seed surface by soaking samples, each containing 100 seeds, in 50 ml of 10% Clorox for 5 min. Seeds were then soaked for 4 hr in a suspension of *F. moniliforme* (5.0 × 10^7 cells per milliliter) prepared by blending the contents of three petri dishes of rapidly growing colonies with 250 ml of sterile, distilled water. Seeds were drained, air-dried, and planted after 24 hr. Seedlings were counted after 9 days, and crowns were assayed for *F. moniliforme* after 16 days.

For field trials, 12,000 Iochief seeds were surface disinfected for 5 min in 10% Clorox. Half of the seed lot became treatment A. The other half, treatment B, was soaked for 20 min in a conidial suspension of *F. moniliforme* (5.0 × 10^7 cells per milliliter), drained, and air-dried. When plated on Nash and Snyder medium and PDA without additional surface disinfection, 1% of the seeds of treatment A and 100% of the seeds of treatment B contained *F. moniliforme*.

Field plots were located in two areas of Idaho where management practices had resulted in different resident levels of *F. moniliforme*. One set of plots was in the Crookham Seed Company genetic nursery in Caldwell, which was specifically designed for disease resistance selection. Corn is grown every year, and all residues are incorporated into the soil to increase pathogen inoculum (S. Marshall, Director of Research, Crookham Seed Company, *personal communication*). On the Moscow site at the Plant Science Farm, corn had not been grown, and soil dilution plates indicated a level of *F. moniliforme* of less than 10^{-3} viable propagules per gram of soil.

The experiment was designed as a randomized complete block with 60 seeds per 23-m row and six replicates of four rows per treatment at each site. Stand counts were taken at the eight-leaf stage on 22 June. Four seedlings about 20 cm tall were chosen at random from each row, and tissue sections of roots, crowns, and stems were surface disinfected and incubated on Nash and Snyder

TABLE 1. Stand count and *Fusarium moniliforme* infection in two hybrids sweet corn cultivars grown in the greenhouse

Treatment number[t]	Treatment: Seed surface disinfected[u]	Treatment: Infested seed[v]	Treatment: Infested soil[w]	Stand count[x,y]: Cultivar Iochief	Stand count[x,y]: Cultivar Earlivee	Crowns from which *Fusarium moniliforme* was isolated[y,z]: Cultivar Iochief (%)	Crowns from which *Fusarium moniliforme* was isolated[y,z]: Cultivar Earlivee (%)
I	no	no	no	97.5 a	91.5 abc	56.5 b	56.5 b
II	yes	no	no	97.5 a	88.0 bc	7.5 c	3.0 c
III	yes	yes	no	94.0 abc	76.5 de	99.5 a	97.5 a
IV	yes	yes	yes	95.0 a	75.5 e	98.0 a	99.5 a
V	yes	no	yes	99.0 a	85.0 cd	99.0 a	99.0 a

[t] Treatment I seed contained 73% (Iochief) and 67% (Earlivee) *F. moniliforme*. *F. moniliforme* was isolated from 3% of surface-disinfected Iochief seed and 4% of surface-disinfected Earlivee seed in treatments II–V.
[u] Surface-disinfected 5 min in 10% Clorox (0.5% available chlorine).
[v] Soaked for 4 hr in *F. moniliforme* suspension, after which seeds were 100% infested.
[w] Flats of Idaho soil mix #1 (1) infested with a suspension of *F. moniliforme*.
[x] Number of plants after 9 days per 100 seeds planted. Data are averages of two replicates of 100 seeds each.
[y] Values within a column followed by the same letter are not significantly different from each other at the 5% level.
[z] Percentage of crowns from seedlings surviving after 16 days from which *F. moniliforme* was isolated when tissues were incubated on Nash and Snyder medium. Data are averages of two replicates of 73–93 seedlings remaining from 100 seeds planted.

TABLE 2. Isolations of *Fusarium moniliforme* and other fusaria from tissue sections[a] of 14-day-old corn plants growing in two locations in Idaho[b]

Location and seed treatment[c]	Crown: *F. moniliforme* (%)	Crown: Other *Fusarium* spp. (%)	Stem: *F. moniliforme* (%)	Stem: Other *Fusarium* spp. (%)	Roots: *F. moniliforme* (%)	Roots: Other *Fusarium* spp. (%)
Caldwell						
A	94	5[d]	87	5[d]	64	8
B	83	4	77	5	52	11
Moscow[e]						
A	34	45	26	21	18	24
B	98	2	91	2	72	6

[a] Isolations from 16 seedlings per treatment in six replicates. Averages represent isolation frequencies for one crown section, one stem section, and three root sections per plant. Data are percentages of seedlings from which *F. moniliforme* or other *Fusarium* spp. were isolated.
[b] Field plots at Caldwell were heavily infested with *F. moniliforme*. Inoculum density at Moscow site was less than 10^{-3} propagules per gram of soil.
[c] In treatment A, seeds were surface disinfected; *F. moniliforme* was isolated from 3% of surface-disinfected seeds. In treatment B, seeds were surface disinfected and then soaked in suspensions of *F. moniliforme*, so that all seeds were infested.
[d] In seedlings from five of the six replicates, no *Fusarium* spp. other than *F. moniliforme* were isolated.
[e] All pairs of values for treatments A and B from the Moscow site are significantly different from each other ($P = 0.01$).

medium as described above.

RESULTS AND DISCUSSION

A highly significant positive correlation ($r = 0.9885$, eight degrees of freedom, $P < 0.01$) was found between the percentage of seeds with seedborne *F. moniliforme* and the percentage of crowns of resulting plants with *F. moniliforme* when cultivars Iochief and Earlivee were grown in the greenhouse (Table 1). In the field tests at the Caldwell location, no significant differences were found in the percentage of seedling tissues (crowns, stems, or roots) from which *F. moniliforme* was isolated whether 3% (treatment A) or 100% (treatment B) of the seed contained the fungus internally (Table 2). That is, infection did not appear to be associated with the level of seedborne inoculum, which suggests that seedling infection in treatment A originated primarily from soilborne inoculum at this location.

In field tests at the Moscow location, highly significant differences were observed between treatments A and B in the proportions of seedling tissues (roots, stems, and crowns) from which *F. moniliforme* was isolated (Table 2). *F. moniliforme* was isolated much more often from seedlings grown from seeds coated with inoculum of *F. moniliforme* than from seedlings grown from surface-disinfected seeds.

Other *Fusarium* spp. besides *F. moniliforme* were isolated from young seedlings originating from surface-disinfected seeds. When levels of seed or soil inoculum were low (as in treatment A at the Moscow site), other *Fusarium* spp., including *F. roseum* 'Equiseti', 'Graminearum', and 'Culmorum' and *F. tricinctum*, were commonly isolated.

These studies suggest that seedborne *F. moniliforme* can be a source of inoculum for seedling infection in the sweet corn cultivars Iochief and Earlivee. However, when seeds did not carry inoculum, soilborne inoculum was equally effective in infecting seedlings. Because so many crops are susceptible to *F. moniliforme*, inoculum carried over from one crop to the next in the soil and seeds is cause for added concern, because it may introduce the pathogen into new areas.

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