Influences of Production Inputs on Incidence of Infection by *Fusarium* Species on Cereal Seed

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ABSTRACT

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Production inputs of supplementary nitrogen, fungicides, and plant growth regulator were evaluated on wheat and triticale cultivars (four site years) and on two- and six-row barley cultivars (two site years each) for effect on the incidence of *Fusarium* infection of the harvested seed. Supplementary nitrogen and a plant growth regulator increased, by up to 125%, the incidence of infection by *Fusarium* species of the seed. The use of a fungicide spray program had no effect on incidence of *Fusarium* infection. Triticale lines were more susceptible than wheat to *Fusarium* infection. Cadette was the most susceptible six-row barley cultivar to *Fusarium*, and Rodeo and Birka were the most susceptible two-row barley cultivars. Over the 2 yr, four major species were isolated: *F. avenaceum, F. graminearum, F. sporotrichioides*, and *F. poae*.

Fusarium graminearum Schwabe, the anamorph of *Gibberella zeae* (Schwein.) Petch, is the major species associated with Fusarium head blight of cereals in Canada (16). A number of other species, including *F. culmorum* (Wm. G. Sm.) Sacc., *F. avenaceum* (Fr.:Fr.) Sacc., and *F. nivale* (Fr.) Ces., are also associated with Fusarium head blight of cereals in other areas (22). In addition to *F. graminearum*, approximately 20 other species of *Fusarium* have been isolated from cereal heads and seed (2,10). In Atlantic Canada, *F. graminearum, F. avenaceum, F. poae* (Peck) Wollenweb., and *F. sporotrichioides* Sherb. are the most commonly isolated species from wheat (*Triticum aestivum* L.) and barley (*Hordeum vulgare* L.) (*unpublished*). Other species may also be important, depending on location and environmental conditions. In Idaho, *F. culmorum* and *F. acuminatum* Ellis & Everh. were the major species isolated from wheat and barley seed (9).

Surveys of wheat fields in southern Ontario have shown that the incidence of Fusarium head blight and the concentration of the mycotoxin deoxynivalenol can be influenced by cultural practices (18,19). The incidence of Fusarium head blight was higher in fields of winter wheat that followed corn than in those that followed soybeans, barley, or mixed grain. The incidence of Fusarium head blight was reduced if residues of a previous corn crop were plowed down and/or when the seed was treated with a fungicide. Frequency of Fusarium head blight was very low in the surveys (<1%). Disease was not correlated with soil P, K, or pH and was not influenced significantly by amount of nitrogen applied, cultivar planted, presence of other diseases, herbicide used, or weed density (19).

The effect of nitrogen fertilization on the development of diseases has been noted for several *Fusarium*-incited diseases (3). Increased nitrogen fertilization of carnations resulted in a corresponding increase in infection by *G. zeae* (15). Similarly, foot rot of wheat was increased by a high rate of nitrogen (12,14). In a study on the role of nitrogen on Fusarium root rot of winter wheat seedlings, plants treated with NH_4^+ were more severely infected than those treated with NO_3^-, and an increase in nitrogen level increased root infection (13). A 1983 survey did indicate a significant management effect on the incidence of Fusarium head blight, where adequate levels of nitrogen fertilization resulted in a lower disease frequency than where nitrogen was felt to be limiting (19).

The utilization of added inputs to obtain high yields is part of intensive systems of cereal production. The objective of the current study was to determine the effects of production inputs—i.e., the use of supplementary nitrogen, a foliar fungicide program, and a plant growth regulator—on the incidence of *Fusarium* infection of the harvested seed of wheat, triticale (× *Triticosecale* Wittmack), and barley. A crop produced by use of management practices that increase the incidence of *Fusarium* infection of seed could impact on quality of the seed and have potential effects on the next crop. *Fusarium* infection of seed can be associated with mycotoxin contamination that reduces quality. When planted, infected seed can act as an inoculum source for *Fusarium* infection of the crop, including root rots and head blight (22).

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MATERIALS AND METHODS

Management trials of barley and of wheat and triticale were established at various locations in the Atlantic provinces to evaluate cultivar response to a series of production inputs. The incidence of *Fusarium* infection of seed was determined at sites in Charlottetown, Prince Edward Island, and in Truro and Nappan, Nova Scotia (Table 1). Table 2 lists the cultivars used in the trials. Seeding rates were 350 and 450 viable seeds per square meter for the barley and wheat trials, respectively. For wheat and triticale tests, the management variables were: 1) standard management that included a single application of 70 kg/ha of nitrogen at seeding; 2) standard management plus supplementary nitrogen (50 kg/ha of ammonium nitrate) at Zadoks's growth stages (ZGS) 30 (pseudostem erection) and 45 (boot swollen) (23); 3) the supplementary nitrogen program plus a fungicide program of propiconazole (Tilt), 125 g a.i./ha, plus chlorothalonil (Bravo), 808 g a.i./ha, applied at ZGS 45–50 (boot to first spikelets visible); and 4) the supplementary nitrogen and fungicide program plus ethephon (Cerone), 280 g a.i./ha, at ZGS 37–39 (flag leaf visible). For the two- and six-row barley trials, the same basic management variables were used, but only a single supplementary nitrogen application of 50 kg/ha was made at ZGS 30 and chlorothalonil was omitted. The experimental design was a split plot with four replications, with management treatments as the main plots and cultivars as the subplots. Plots were at least eight rows wide by 5 m long and were harvested with small plot combines.

Samples of harvested seed (84–105 seeds per plot) were surface-sterilized for 10 min in 10% Javex (6% sodium hypochlorite) containing 0.01% Tween 20, rinsed in distilled water, and plated on potato-dextrose agar containing 0.67 g a.i./L of pentachloronitrobenzene, 125 ppm of chlortetracycline, and 125 ppm of dihydrostreptomycin sulfate. After incubation at room temperature for approximately 10 days, the incidence of *Fusarium* colonies was determined. Subcultures of 10 randomly selected colonies per plot were transferred to potato-dextrose agar plus antibiotics and, after incubation, were identified according to Nelson et al (11). Data were analyzed and presented as arcsine transformations of the percent incidence of *Fusarium* infection.

RESULTS

Eight site years were used in the evaluation of the effects of production inputs on the incidence of *Fusarium* infection of cereal seed—four for wheat and triticale and two each for two- and six-row barley cultivars (Table 1). With the exception of one test, the addition of supplementary nitrogen, foliar fungicide, and plant growth regulator resulted in a significant increase in the incidence of *Fusarium* infection of the harvested seed. The incidence increased significantly at five locations after the addition of supplementary nitrogen alone. The effect of foliar fungicide plus supplementary nitrogen treatment was not appreciable when compared with the effect of the supplementary nitrogen alone. Supplementary nitrogen plus foliar fungicides plus plant growth regulator generally resulted in the highest incidences of *Fusarium* infection of the seed. All wheat and triticale sites showed a significant increase in *Fusarium* incidence, to a maximum of 47.9%, as a result of the inputs. In both 1986 and 1987, the addition of production inputs resulted in a significant increase (up to 92.1%) in the incidence of *Fusarium* on six-row barley cultivars. In 1987, the two-row barley trial showed a similar effect, with an increase of 125.9% in incidence of *Fusarium*.

A wide range of *Fusarium* species were identified on the seed, including: *F. avenaceum, F. graminearum, F. poae, F. sporotrichioides, F. culmorum, F. sambucinum* Fuckel, *F. subglutinans* (Wollenweb. & Reinking) P.E. Nelson, T.A. Toussoun, & Marasas, *F. moniliforme* J. Sheld., *F. equiseti* (Corda) Sacc., *F. semitectum* Berk. & Ravenel., *F. crookwellense* L.W. Burgess, P.E. Nelson, & T.A. Toussoun, and *F. acuminatum*, plus a number of other species. The most common species was *F. avenaceum*, which accounted for a mean infection incidence in the seed of 21.8 and 15.8% in 1986 and 1987, respectively. Mean infection incidences for *F. graminearum, F. poae*, and *F. sporotrichioides* were 17.7, 4.3, and 4.8%, respectively, in 1986, and 3.1, 5.8, and 1.6%, respectively, in 1987. Other *Fusarium* species accounted for 4.0 and 4.3% in 1986 and 1987, respectively.

There were significant cultivar differences at all test sites (Table 2). Within the wheat and triticale trial, the two triticale lines, Beaguelita and Trit 4, had the highest mean incidences of *Fusarium* infection; Messier registered the highest incidences among the wheat cultivars. The semidwarf cultivar Cadette had the highest incidence among the six-row barleys, and Birka and Rodeo, among the two-row barleys.

Response of *F. avenaceum* to production inputs for wheat and triticale indicated that the addition of inputs to the standard level increased incidences of infection (Table 3). At Charlottetown, cultivar response between years was similar, with the triticale lines and Messier wheat having the highest incidences of infection by *F. avenaceum*. Changes in incidence of *F. graminearum* were significant for management inputs at only one site, with a maximum 98% increase for the highest level of input. Three sites had significant cultivar effects relative to *F. graminearum*, with the triticale lines having the highest incidence levels. Production inputs had no significant impact on the incidence of *F. poae*

Table 1. Effect of crop management practices on incidence of *Fusarium* species isolated from cereal seed at various test locations in Canada, 1986 and 1987

Crop Location, year	Incidence per treatment[a] No. 1	No. 2	No. 3	No. 4	SEM	LSD ($P = 0.05$)
Wheat and triticale						
Charlottetown, P.E.I., 1986	19.0	24.3	27.2	28.1	1.216	3.9
Charlottetown, 1987	19.2	21.7	20.1	24.2	0.486	1.6
Nappan, N.S., 1987	12.3	12.6	13.5	16.5	0.441	1.4
Truro, N.S., 1987	20.8	25.6	26.5	29.7	0.856	2.7
Two-row barley						
Charlottetown, 1986	34.3	41.4	39.2	38.5	NS[b]	
Charlottetown, 1987	11.6	12.3	19.4	26.2	1.121	3.6
Six-row barley						
Charlottetown, 1986	25.3	29.1	30.5	31.8	1.030	3.3
Charlottetown, 1987	12.7	17.4	16.9	24.4	0.736	2.6

[a] Arcsine transformation of percent data. No. 1 = standard treatment, No. 2 = No. 1 + ammonium nitrate, No. 3 = No. 2 + foliar fungicide, No. 4 = No. 3 + plant growth regulator.
[b] Not significant at $P = 0.10$.

Table 2. Response of wheat (W), triticale (T), and barley cultivars to *Fusarium* seed infections

Crop Cultivar	Incidence[a] Charlottetown 1986	Charlottetown 1987	Nappan 1987	Truro 1987	Mean
Wheat and triticale					
Milton (W)	18.8	19.7	12.6	18.6	16.0
Max (W)	19.4	17.9	13.1	19.2	15.9
Messier (W)	27.0	24.4	14.9	26.2	20.0
Belvedere (W)	18.1	18.1	11.9	24.5	17.0
Sinton (W)	22.0	19.5	12.0	24.4	18.0
Beaguelita (T)	33.7	27.3	16.8	32.0	23.7
Trit 4 (T)	33.7	22.3	14.7	34.4	23.3
SEM	0.896	0.759	0.733	1.157	
LSD	2.5*[b]	2.1*	2.1*	3.3*	
Six-row barley					
Bruce	28.9	···	···	···	
Cadette	32.2	26.6	···	···	29.4
Laurier	29.2	15.2	···	···	22.2
Leger	28.5	13.7	···	···	21.1
OAC Kippen	26.2	···	···	···	
Joly	29.9	15.8	···	···	22.9
SEM	1.280	1.056			
LSD	3.0**	3.0*			
Two-row barley					
Albany	35.0	15.8	···	···	25.4
AB78-1	34.2	···	···	···	
AB80-1	41.4	···	···	···	
Birka	41.3	20.9	···	···	31.2
KGSB84-6	37.1	···	···	···	
Micmac	35.4	14.0	···	···	24.7
Rodeo	43.3	18.6	···	···	31.0
SEM	1.870	1.348			
LSD	5.3*	3.9*			

[a] Arcsine transformation of percent data.
[b] * = $P = 0.05$, ** = $P = 0.10$.

Table 3. Influence of production input and wheat (W) or triticale (T) cultivar on incidence of *Fusarium* species in seed, where cultivar or production input was significant

	Incidence[a]										
	F. avenaceum				*F. graminearum*			*F. poae*			*F. sporotrichioides*
	Charlottetown		Nappan	Truro	Charlottetown	Nappan	Truro	Charlottetown		Nappan	Charlottetown
	1986	1987	1987	1987	1986	1987	1987	1986	1987	1987	1986
Treatment[b]											
No. 1	14.8	17.8	9.2	13.9	9.2	1.8	15.3	1.2	7.3	5.6	1.1
No. 2	18.1	20.6	10.4	18.2	14.5	1.1	17.2	1.6	7.2	3.4	3.5
No. 3	21.5	18.7	10.6	18.9	13.1	1.1	17.8	0.5	6.7	6.1	0.5
No. 4	21.0	23.0	13.8	21.3	18.2	1.1	19.5	0.3	8.8	7.1	2.8
SEM	1.225	0.743	0.739	1.185	1.864	0.582	1.035	0.613	0.900	1.122	0.866
LSD	3.9*[c]	2.4*	2.4*	3.8*	6.0*	NS[d]	NS	NS	NS	NS	NS
Cultivar											
Milton (W)	12.7	18.8	10.6	15.4	12.0	0.9	9.6	2.0	7.9	3.2	0.6
Max (W)	15.6	17.2	10.8	15.0	11.2	0.4	11.4	0.0	4.3	3.5	2.4
Messier (W)	22.2	24.1	13.0	18.5	14.4	1.2	18.4	0.0	5.8	4.4	1.4
Belvedere (W)	13.8	16.0	9.4	21.7	11.0	0.0	11.8	1.1	7.7	6.1	0.6
Sinton (W)	15.7	18.5	10.9	19.0	12.0	0.4	14.9	2.4	4.7	4.1	4.7
Beaguelita (T)	29.3	25.0	11.9	20.3	18.6	3.4	25.2	0.0	12.2	9.6	0.0
Trit 4 (T)	22.9	20.8	10.3	16.7	17.0	2.8	30.7	0.7	9.7	8.0	4.1
SEM	1.632	0.910	0.947	1.172	1.751	0.695	1.555	0.636	1.200	1.019	1.099
LSD	4.6*	2.6*	NS	3.3*	4.9*	2.0*	5.0*	1.8	3.4*	2.9*	3.1

[a] Arcsine transformation of percent data.
[b] No. 1 = standard treatment, No. 2 = No. 1 + ammonium nitrate, No. 3 = No. 2 + foliar fungicide, No. 4 = No. 3 + plant growth regulator.
[c] * = $P = 0.05$.
[d] Not significant at $P = 0.10$.

Table 4. Influence of production inputs on incidence of *Fusarium* species in seed from six-row barley trials with a significant cultivar or production input effect

	Incidence[a]				
	F. avenaceum		*F. graminearum*		*F. poae*
	1986	1987	1986	1987	1987
Treatment[b]					
No. 1	18.0	10.4	15.3	0.4	4.6
No. 2	20.4	15.2	18.6	0.5	6.0
No. 3	19.3	15.8	19.5	0.4	3.2
No. 4	21.8	22.2	20.0	1.7	8.0
SEM	1.060	0.725	1.200	0.664	1.150
LSD	NS[c]	2.3*[d]	3.1**	NS	2.9**
Cultivar					
Bruce	21.2	...	16.6	...	...
Cadette	22.1	24.8	18.4	0.0	8.2
Laurier	21.5	10.6	16.3	1.2	7.5
Leger	16.6	13.2	20.4	0.0	3.1
OAC Kippen	15.8	...	19.1	...	...
Joly	22.0	15.0	19.5	1.8	3.1
SEM	1.550	1.227	1.610	0.536	1.207
LSD	4.4*	3.5*	NS	1.3**	3.5*

[a] Arcsine transformation of percent data.
[b] No. 1 = standard treatment, No. 2 = No. 1 + ammonium nitrate, No. 3 = No. 2 + foliar fungicide, No. 4 = No. 3 + plant growth regulator.
[c] Not significant at $P = 0.10$.
[d] * = $P = 0.05$, ** = $P = 0.10$.

or *F. sporotrichioides*. In general, the triticale lines had higher incidences of infection by *F. poae* than did the wheat cultivars.

There were no significant effects from *F. sporotrichioides* in the six-row barley trial, and there were no significant effects in 1986 on *F. poae* (Table 4). Incidence of *F. avenaceum* was significantly influenced by treatment in 1987 at the maximum level of production inputs. Incidence of *F. graminearum* was increased by 30.7% at the maximum input level ($P = 0.10$). Infection incidences for *F. avenaceum* and *F. poae* were highest in Cadette. *F. graminearum* showed no significant cultivar responses in 1986, and levels were too low in 1987 to obtain meaningful comparisons.

In the two-row barley trials, with the exception of *F. sporotrichioides*, the incidence of infection by the major species increased as production inputs increased (Table 5). *F. sporotrichioides* showed that moderate inputs were more conducive to infection. Cultivar responses were significant in some trials but varied between years.

DISCUSSION

The use of high-yielding production inputs in this study, while having a beneficial impact on crop yield (*unpublished*), favored *Fusarium* infection of the seed. This impact was principally related to the supplementary nitrogen topdressing and the plant growth regulator. The foliar fungicide program had no effect on incidence of *Fusarium* infection of the seed.

Effect of nitrogen on *Fusarium*-incited diseases of cereals has been noted and includes both Fusarium root rot and Fusarium head blight (13,15,20,21). Our study indicates that supplementary nitrogen, in the form of ammonium nitrate, can have a significant impact on incidence of *Fusarium* infection of cereal seed. The role that supplementary nitrogen may play in *Fusarium* infection of seed is unknown, although changes in resistance may be involved. There is evidence that seedling resistance to *Fusarium* can be linked to resistance to head blight (8). Thus, evidence of nitrogen effects on seedling infection (12,14) may give some indication of a possible effect on response of Fusarium head blight. It has also been noted, however, that high rates of nitrogen fertilizers, unbalanced with phosphorus, decreased resistance to Fusarium head blight (20).

The form of nitrogen appears to play a role in Fusarium head blight response, with the incidence being lower in wheat fertilized with urea than in wheat fertilized with ammonium nitrate (17). Ammonium nitrate was used as the supplementary nitrogen source in our study, and another form may have produced a different response.

The application of foliar fungicides had no effect on the incidence of *Fusarium* infection of the seed. In other studies, however, propiconazole has had a significant effect in reducing disease severity of Fusarium head blight (7), as has the use of chlorothalonil (6) and of thiophanate-methyl (21). We should note that in the current study, fungicides were applied before full head emergence. A different application timing, relative to growth stage and infection periods, could have had a positive effect on disease control.

The maximum incidence of *Fusarium* infection of seed was observed with supplementary nitrogen plus foliar fungicide plus plant growth regulator. Plant growth regulators in a high-yield production system are used to reduce lodging, which is often enhanced by high levels of nitrogen fertilization. The plant growth regulator may increase incidence of *Fusarium* infection by changing crop characteristics, notably by reducing plant height, thereby increasing density and altering the microclimate of the canopy. Changes in canopy characteristics resulting from use of growth regulators may also enhance inoculum production on crop debris or even prolong periods when the crop is susceptible to infection. The fact that plant growth regulators reduce crop height would indicate that the plant is subjected to abnormal stress conditions that in turn may affect, either directly or indirectly, crop susceptibility to *Fusarium*. Weed density can also play a role in Fusarium head blight, either by increasing nutrient or water stress on the plant or by modifying the crop environment (19).

Some of the treatment effect could be related to the physical characteristics of the seed in relation to the method of harvest. Treatments used in this study tend to increase the seed size as measured by thousand kernel weight and hectoliter weight. In addition, the use of mechanical combining can result in the loss of severely infected kernels, which are light and shriveled. Thus, the low level of *Fusarium* infection of seed with the standard, or base, treatment should not be construed as meaning that the level of Fusarium head blight was lowest in these plots. High disease incidence could result in low incidence on the harvested seed if combining removes the lighter, infected seeds. If treatment results in heavier seeds, infected kernels are less likely to be removed during combining, thus giving an apparently high rate of seed infection. Although seed size selection, and not incidence of Fusarium head blight in the field, could account for some of the higher incidence on seed from cereals receiving supplementary nitrogen or growth regulator, the harvested seed is the final measure of disease impact on the current crop and potential effects on the next crop.

Table 5. Influence of production inputs on incidence of *Fusarium* species in seed from two-row barley trials with a significant cultivar or production input effect

	Incidence[a]						
	F. avenaceum		*F. graminearum*		*F. poae*	*F. sporotrichioides*	
	1986	1987	1986	1987	1987	1986	1987
Treatment[b]							
No. 1	21.5	9.2	20.6	0.4	5.5	7.8	0.0
No. 2	24.6	10.5	24.0	0.4	4.9	11.4	0.9
No. 3	28.2	18.2	21.1	1.7	6.5	5.9	1.7
No. 4	28.9	22.1	18.0	3.7	10.7	4.5	0.0
SEM	2.010	1.507	2.290	0.979	1.132	0.790	0.405
LSD	5.2**[c]	4.8*	NS[d]	3.1*	3.6*	2.5*	1.3*
Cultivar							
Albany	25.4	14.4	19.5	1.9	5.5	3.2	0.9
AB78-1	19.3	...	22.8	...	...	5.5	...
AB80-1	26.1	...	24.6	...	...	9.8	...
Birka	23.4	16.7	25.5	0.1	4.9	12.1	1.3
KGSB84-6	28.7	...	17.7	...	...	2.3	...
Micmac	25.7	12.6	19.7	1.3	6.5	5.6	0.0
Rodeo	31.8	16.3	16.6	3.0	10.7	13.2	0.4
SEM	2.060	1.445	2.480	0.797	1.495	1.840	0.564
LSD	5.8*	NS	5.9**	1.9**	3.6**	5.2*	1.6*

[a] Arcsine transformation of percent data.
[b] No. 1 = standard treatment, No. 2 = No. 1 + ammonium nitrate, No. 3 = No. 2 + foliar fungicide, No. 4 = No. 3 + plant growth regulator.
[c] * = $P = 0.05$, ** = $P = 0.10$.
[d] Not significant at $P = 0.10$.

The incidence of *Fusarium* infection of seed was higher in the triticale lines than in wheat, a fact noted in other studies of Fusarium head blight (4) and root rot (5). Higher incidence in the triticales may have been related to differences in inherent resistance between wheat and triticale but could also have been enhanced by other factors. The triticales may have been more susceptible to *Fusarium* infection in part as a result of the length of time during which infection could take place. The time between heading and ripening was approximately 43.5 days for wheat and 50 days for the triticale lines. *Fusarium* may have had more time, therefore, to infect the heads and progress into the seed of triticale.

The role the various *Fusarium* species play in pathogenicity of developing seedlings may be influenced by the effect intensive management has on seed production. Of the four major species isolated in the current study, all but *F. poae* are considered to be pathogenic to cereal seedlings, causing foot and root rots (1,22). Thus, when management inputs are being determined, the reactions of the cultivar and treatments selected to each pathogenic species should be considered. *Fusarium* infection of seed enhanced by management practices could not only increase the incidence of seedling infections but also be a source of inoculum for head infections.

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