P. 3

Sep 15391

Blackwell Synergy

Email: ?
Password: GO

Empresa Brasileira de Pesquisa Agropecuaria

Register Forgotten Password Athens/Institution Login

Synergy Home | Browse | Search | My Synergy | Books Online | Resources | About | Help

Journal of Phytopathology

Journal Menu

Table of Contents
List of Issues

Tools

Email this article
Add to favorite articles
Export this citation
Alert me when this article is cited: Email | RSS
(What is this?)

View ISI citation
Related articles

Publication history

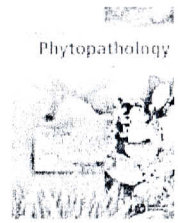
Published article online:
30 Jul 2007
Issue online:
10 Sep 2007

Received January 18, 2007; accepted March 18, 2007

Journal of Phytopathology

Volume 155 Issue 10 Page 577-581, October 2007

To cite this article: E. M. Del Ponte, J. M. C. Fernandes, G. C. Bergstrom (2007) Influence of Growth Stage on Fusarium Head Blight and Deoxynivalenol Production in Wheat
Journal of Phytopathology 155 (10) , 577–581
doi:10.1111/j.1439-0434.2007.01281.x



Next Article

You have full access rights to this content

Abstract

Influence of Growth Stage on Fusarium Head Blight and Deoxynivalenol Production in Wheat

E. M. Del Ponte[1], J. M. C. Fernandes[2] and G. C. Bergstrom[3]

Authors' addresses: [1]Department of Fitossanidade, Federal University of Rio Grande do Sul, 91540000, Porto Alegre, RS, Brasil; [2]Embrapa Trigo, Rodovia BR 285, km 174, Passo Fundo, RS, Brasil; [3]Department of Plant Pathology, Cornell University, Ithaca, NY 14853-5904, USA (correspondence to E. M. Del Ponte. E-mail: emerson.delponte@ufrgs.br)

Department of Plant Pathology, Cornell University, Ithaca, NY, USA

Abstract

Fusarium head blight is a major concern for wheat production worldwide. The fungi that cause the disease may infect head tissues from flowering to late stages of kernel development, but a better understanding of the influence of the time of infection on grain weight reduction and mycotoxin accumulation resulting from the infection process is needed. We investigated the influence of wheat reproductive stage at the time of inoculation on disease and grain quality parameters, especially production of deoxynivalenol (DON) in mature grains. Heads of Norm wheat were spray inoculated with a macroconidial suspension of a DON-producing isolate of *Fusarium graminearum* at each of six reproductive stages from flowering to hard dough. Plants were incubated in a mist chamber for 48 h and then moved to the greenhouse until maturity. Norm wheat was susceptible at all stages inoculated but the highest grain weight reduction and DON accumulation occurred in plants inoculated past flowering to late milk stages. However, high incidences of kernel infection and significant levels of DON accumulation resulted from inoculations as late as the hard dough stage, even though there was no corresponding reduction in grain weight compared to non-inoculated plants. The occurrence of commercially significant levels of DON in plump, high-yielding wheat may result from infections that occur during favourable environments well after the flowering stages. Late infection and

P. 3

J. Phytopathology, **155**, 577–581 (2007)
doi:10.1111/j.1439-0434.2007.01281.x
Journal compilation © 2007 Blackwell Verlag, Berlin
No claim to original US government works

Department of Plant Pathology, Cornell University, Ithaca, NY, USA

Influence of Growth Stage on Fusarium Head Blight and Deoxynivalenol Production in Wheat

E. M. Del Ponte[1], J. M. C. Fernandes[2] and G. C. Bergstrom[3]

Authors' addresses: [1]Department of Fitossanidade, Federal University of Rio Grande do Sul, 91540000, Porto Alegre, RS, Brasil; [2]Embrapa Trigo, Rodovia BR 285, km 174, Passo Fundo, RS, Brasil; [3]Department of Plant Pathology, Cornell University, Ithaca, NY 14853-5904, USA (correspondence to E. M. Del Ponte. E-mail: emerson.delponte@ufrgs.br)

Received January 18, 2007; accepted March 18, 2007

Keywords: *Gibberella zeae*, tricothecenes, fungal infection, toxigenic fungi, food safety

Abstract

Fusarium head blight is a major concern for wheat production worldwide. The fungi that cause the disease may infect head tissues from flowering to late stages of kernel development, but a better understanding of the influence of the time of infection on grain weight reduction and mycotoxin accumulation resulting from the infection process is needed. We investigated the influence of wheat reproductive stage at the time of inoculation on disease and grain quality parameters, especially production of deoxynivalenol (DON) in mature grains. Heads of Norm wheat were spray inoculated with a macroconidial suspension of a DON-producing isolate of *Fusarium graminearum* at each of six reproductive stages from flowering to hard dough. Plants were incubated in a mist chamber for 48 h and then moved to the greenhouse until maturity. Norm wheat was susceptible at all stages inoculated but the highest grain weight reduction and DON accumulation occurred in plants inoculated past flowering to late milk stages. However, high incidences of kernel infection and significant levels of DON accumulation resulted from inoculations as late as the hard dough stage, even though there was no corresponding reduction in grain weight compared to non-inoculated plants. The occurrence of commercially significant levels of DON in plump, high-yielding wheat may result from infections that occur during favourable environments well after the flowering stages. Late infection and DON production should therefore be a future research focus for wheat breeding and integrated management of FHB and an important consideration for grading systems that employ the presence of visibly damaged kernels as a means of estimating DON content of wheat.

Introduction

Fusarium head blight (FHB), also known as scab, is a destructive disease of cereals that has reached epidemic proportions in North America and several other parts of the world in the past decades (McMullen et al., 1997). The major causal organism of this disease worldwide is *Gibberella zeae* (Schwein) Petch (anamorph: *Fusarium graminearum* Schwabe) (Schmale and Bergstrom, 2003). The disease affects wheat, barley, and other small grains both in temperate and semi-tropical areas causing reduction in grain yield and quality. Costs to farmers include increased management costs, reduced income from outright food or feed losses, and lower selling prices for commodities contaminated with mycotoxin (Jones and Mirocha, 1999). The occurrence of DON is of concern for human and animal health, as this toxin is known to cause food refusal, vomiting, and depressed immune function (World Health Organization, 2001). Although progress is being made in disease reduction through chemical control, cultural control, and the development of resistant cultivars, satisfactory levels of control have yet to be attained (Bai and Shaner, 1994; Cromey et al., 2001). The fungus overwinters as a saprophyte in debris of infected cereals and grasses, including corn and wheat (Dill-Macky and Jones, 2000). Ascospores and macroconidia of the fungus are produced on overwintered residues (Khonga and Sutton, 1988) and both spore types are able to infect wheat any time from flowering through kernel filling stages if conditions are conducive (Shaner, 2003). However, there is lack of consensus about the exact size of the phenological window of vulnerability to infection, and especially for the accumulation of DON in the kernels. Some authors demonstrated that infection occurred principally at anthesis with the anthers as the main infection site (Atanasoff, 1920; Pearce et al., 1976). However, others have observed an extended window of infection with a peak after anthesis (Andersen, 1948; Schroeder and Christensen, 1963).

DON accumulation in the kernels is influenced by many factors including host resistance, chemotype,

aggressiveness of the prevalent fungal species, and environment (mainly temperature and moisture) during disease development (Hart et al., 1984; Mesterházy et al., 1991). The dynamics of toxin production in the field seem to be of a complex nature. Miller and Young (1985) reported that DON levels in heads inoculated at flowering increased for six weeks, then declined. This study was conducted to determine the influence of growth stage on disease and grain quality parameters. A brief, preliminary report of a portion of these findings was published (Del Ponte et al., 2003).

Materials and Methods

Inoculum preparation

Cultures of a single-conidium isolate of *G. zeae* (GZ014NY1998), with documented virulence and ability to produce DON in wheat, were grown in Petri dishes containing 1/4 strength potato-dextrose-agar (PDA) at a constant 25°C and a photoperiod of 12 h fluorescent light for 7 to 10 days. Macroconidia were harvested by washing the culture surface with sterile, distilled water. The resulting inoculum suspensions were adjusted with water to a concentration of 10^5 conidia/ml. Inoculum was used within 3 h after preparation.

Experimental design

Ten seeds of hard red spring wheat cv. Norm were sown in clay pots (2.5 l) containing a soil substrate and grown in the greenhouse at temperatures ranging from 20–23°C. By the beginning of anthesis, groups of six pots (each pot was one replicate) of homogeneous phenology were established. In each pot, 7 to 10 main tillers were maintained and secondary tillers were consistently eliminated during the experiment. Each treatment consisted of single-time spray-inoculations at one distinct growth stage, as follows, according to Zadoks scale (Zadoks et al., 1974): 65 – Anthesis half-complete; 71 – kernel watery ripe; 73 – early milk; 77 – late milk; 83 – early dough; and 87 – hard dough. Control treatment consisted of a group of wheat heads sprayed with sterile water. A hand-sprayer was used to apply water and the spore suspension; wheat heads were sprayed to saturation. After applying the treatments, plants were moved into a mist chamber for 48 h under continuous moisture and temperatures ranging from 21 to 23°C. Thereafter, plants were moved back to the greenhouse and grown until maturity. The experiment was repeated once with six replications (a potted plant with 7 to 10 main tillers) of treatments in each experiment.

Disease and grain quality parameters evaluated

Wheat heads were inspected at the sixth day following inoculation and the percentage of spikelets which were symptomatic (RDS - relative disease severity) was recorded. A spikelet was recorded as visually infected when showing premature bleaching and discolouration. At maturity, all kernels of each replicate were hand-harvested, hand-threshed and dried to a uniform moisture content of 13% by weight. Five sub-samples of 50 kernels were taken from each treatment to determine the mean percentage of visually scabby kernels (VSK), e.g. discoloured, shriveled or pinkish white kernels. Mean percentage of kernel infection (KI) was estimated by plating 10 sub-samples of 100 kernels in 10 plates with a selective medium – SNAW (Mandanhar and Cunfer, 1991). The fungal isolate inoculated previously was identified based on cultural and morphological characteristics. Three sub-samples of 100 kernels were weighed and the average used to estimate 1000-kernel weight (KW). DON levels were determined by direct competitive enzyme immunoassay RIDA-SCREEN® DON (Digen Ltd., Oxford, UK) according to manufacturer's instructions. For each sample, 5-g grain samples were ground in a coffee mill for 12–15 s, and then stored in sealed plastic containers before analysing. One hundred millimetres of extraction buffer was added to the 5-g ground sample. Samples were shaken for 3 minutes and filtered into a collection tube. When toxin levels exceeded the test range, the test was repeated after the extract was diluted with buffer. The DON concentration was calculated accordingly.

Data analysis

Data from the two experiments were pooled due to lack of a significant difference between them (data not shown), and statistical analysis was based on all 12 combined replicates. DON analysis was an exception in that there were three replicates per experiment (total of six replicates for both experiments). Summary statistics and Pearson correlations were made in R software (R Foundation for Statistical Computing, Vienna, Austria). For the correlations of DON × KW and DON × VSK, averages for two pairs of the 12 replicates were used, in order to form six pair values of KW and VSK to correlate with six DON replicate values and to make the graphs.

Results

Norm wheat was susceptible to infection from flowering through all stages of kernel development. Disease severity could not be evaluated for heads inoculated at hard dough stage, due to natural senescence. The time from inoculation to appearance of symptoms decreased from flowering until early dough stage inoculations. Typical disease symptoms were first observed 3 to 4 days following inoculation, except in heads inoculated at flowering, in which the incubation period ranged from 7 to 10 days. Hence, RDS value at 6 days for inoculations at anthesis was recorded as zero, although it increased dramatically afterwards (data not shown). Mean values for disease and grain quality parameters evaluated are presented in Table 1. The incidence of visually scabby kernels and the degree of damage was highest for the earliest inoculations and least for inoculations at hard dough, which did not affect grain weight compared to the control. A higher incidence of visual symptoms was observed in kernels

Table 1
Effect of *Fusarium graminearum* inoculated at six growth stages on disease and grain quality parameters of wheat

Zadoks stage at inoculation[a]	Mean (standard error) for disease parameters			Mean (standard error) for grain quality parameters	
	RDS[b] (%)	VSK[c] (%)	KI[d] (%)	KW[e] (g)	DON[f] (mg/kg)
65	0.0	94.0 (1.2)	52.0 (2.3)	23.4 (0.1)	24.7 (5.7)
71	31.3 (3.4)	97.3 (0.7)	72.2 (2.5)	11.4 (0.1)	98.0 (3.2)
73	40.6 (5.1)	95.2 (1.6)	94.0 (1.4)	17.8 (0.1)	89.7 (5.5)
77	67.0 (6.5)	99.6 (0.2)	99.3 (0.5)	26.9 (0.0)	49.0 (3.0)
83	82.4 (3.6)	72.3 (3.7)	96.2 (1.3)	37.8 (0.1)	13.3 (1.5)
87	na	23.4 (1.8)	71.7 (3.6)	42.9 (0.1)	1.2 (0.2)
Non-inoculated control	0.0	0.0	0.0	41.0 (0.1)	0.0

[a]65 - Anthesis half-complete; 71 – Kernel watery ripe; 73 – Early milk; 77 – Late milk; 83 – Early dough; 87 – Hard dough (Zadoks et al., 1974).
[b]Relative disease severity – % of diseased spikelets per head at 6 days after inoculation ($n = 12$).
[c]Visually scabby kernels ($n = 12$).
[d]Kernel infection by *Fusarium graminearum* on culture media ($n = 12$).
[e]1000-kernel weight.
[f]Deoxynivalenol concentration ($n = 6$).

inoculated at early dough (72%) but the impact on grain weight was not so evident. Nearly one hundred percent of kernels from inoculations at early milk to early dough stages were colonized by the fungus.

DON levels ranged from 1.2 to 98 mg/kg with the highest toxin levels detected in kernels from inoculations at watery ripe or early milk stages. These values were around three and two times higher than toxin levels in kernels from earlier (anthesis) or later (late milk) inoculations, respectively. A high amount (13.3 mg/kg) of DON was detected in kernels from heads inoculated at early dough, and lowest levels (1.2 mg/kg) were detected in kernels inoculated at hard dough (Table 1). Kernel weight was greatly reduced (two to four times lower than the control) by inoculations at anthesis up to late milk stages. Inoculations at dough stages resulted in 1000-kernel weights similar to the non-inoculated control. Both positive and negative correlations among the parameters were observed by analysing combined data from all six replicates in each of the six inoculation times ($n = 36$). A significant negative correlation ($P < 0.001$) was observed between kernel weight and DON ($r = -0.88$) and kernel weight and VSK ($r = -0.77$). A significant positive correlation ($P < 0.001$) was observed between DON and VSK ($r = 0.64$). Correlation between KI and DON was not significant ($r = 0.09$). Relationships for DON and VSK and between DON and KW are presented in Fig. 1. DON content in the kernels ranged from around 1 to 100 mg/kg. Deoxynivalenol decreased linearly with increasing kernel weight from inoculations performed at later stages of kernel development, yet a significant amount of toxin was observed in some kernels that were nearly as plump as kernels from non-inoculated plants.

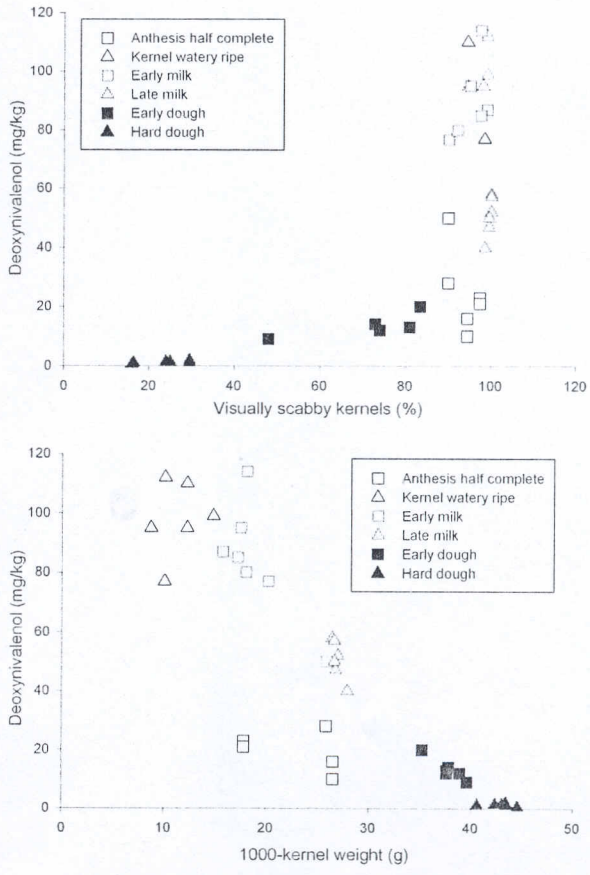


Fig. 1 Relationships between visually scabby kernel and deoxynivalenol concentration (top) and between 1000-kernel weight and deoxynivalenol concentration (bottom) resulting from single-time inoculations at six distinct stages of kernel development ($n = 36$). Mean kernel weight in the non-inoculated control was 41 g

Discussion

In the present study we demonstrated that Norm, a cultivar already known as highly susceptible to FHB, has a wide window of vulnerability to infection by *F. graminearum*. A high incidence of fungal colonization of kernels was observed following anthesis through the dough stages of kernel development. This is in agreement with Andersen (1948) who reported

that wheat heads were most susceptible at postanthesis stages. Strange et al. (1974) suggested that anthers play an important role in infection based on the presence in anthers of the compounds choline and betaine that stimulated fungal growth. Some have suggested that susceptibility increases after flowering precisely because senescent anthers adhere to glumes (Takegami, 1957). However, Engle et al. (2002) found no effect of choline and betaine on germination of conidia and ascospores, suggesting that endogenous compounds of flowers do not promote colonization by *F. graminearum*. In our study, a significant proportion of kernels became infected after inoculation at soft and hard dough stages, although visual symptoms on kernels and toxin content were lower than in kernels inoculated earlier in development. Susceptibility of wheat inoculated at different growth stages was previously documented in a field study by Fernando et al. (1997) who exposed wheat heads to field inoculum (ascospores) at specific times during kernel development. In that study, the most susceptible period was anthesis (70% of kernels were symptomatic) to early milk; whereas only 20% of kernels symptomatic were symptomatic in heads exposed at late milk stage and 5% of kernels symptomatic in heads exposed at hard dough stage. In our controlled inoculation study, a high incidence of kernel infection was recorded at all reproductive stages, although DON levels declined steadily between kernel watery ripe and hard dough. Early dough stage inoculations resulted in a slightly lower percentage of visually scabby kernels than did earlier inoculations. It is possible that the consistently high level of kernel infections we observed at the late stages, in contrast with results by Fernando et al. (1997), might be due to differences in the methodology, i.e. inoculum type and dosage and wetness duration, since our study was conducted with high inoculum and a highly favourable environment. The lack of a relationship between kernel infection incidence and DON was reported by Argyris et al. (2003) who collected wheat kernels at 5-day intervals during kernel filling. They observed that although kernel infection increased in later stages, DON was present at significant levels as early as 10 days after anthesis and it remained within a narrow range through seed development. Those results, however, conflict with a previous paper (Trigo-Stockli et al., 1995) that reported significant correlation between kernel infection and DON, when analysing mature kernels and not kernels harvested during filling stages. Mesterházy et al. (2005) found strong correlations between FDK and DON contamination resulting from inoculations at flowering, suggesting that scabby or tombstone kernels are the most important carriers of the toxins, and selections for low FDK at a reasonably high infection pressure would lead to lower toxin levels in the grain. In our study, we found a significant percentage of kernels (23%) with visually lighter damage in kernels inoculated at hard dough stages that did not impact kernel weight yet may have contributed to the DON levels observed.

Our findings also agree with a previous study by Hart et al. (1984) who observed that production of DON in wheat kernels varied according to the duration of wetness, regardless of the stage of kernel development, after they were filled. Nevertheless, we have observed a very high accumulation (49 to 98 mg/kg) of mean DON in kernels inoculated from watery ripe through milk stages and commercially significant mean toxin levels (1–13 mg/kg) in kernels inoculated at dough stages. Infections during flowering directly affect kernel development resulting in floret abortion or the development of small, shriveled and lightweight kernels that may be lost during harvest and cleaning operations (Schaafsma et al., 2001). In the present study, toxin-contaminated kernels resulting from inoculations at dough stages were similar in weight to non-inoculated kernels, suggesting that late infected kernels may also contribute to lower grain quality, i.e., mycotoxin contamination, even if they have a negligible effect on grain yield. Depending on the country and intended grain use, wheat may be substantially discounted or even refused by buyers at DON levels as low as 1 mg/kg, the mean level of contamination found in this study for kernels inoculated at late dough stage. Empirical evidence exists for the potential role of late infections in commercially significant toxin accumulation in mature grains. In several recent years in New York State, USA, an apparent uncoupling of DON contamination in soft winter wheat from the occurrence of FHB symptoms and grain weight reduction has been observed. That is, plump and high-yielding wheat has been contaminated with DON above 2 mg/kg (Bergstrom, unpublished). Similarly, Cowger and Sutton (2005) reported that severity of FHB in a portion of the south-eastern USA in 2003 was better correlated with weather parameters after wheat flowering than with those before or at the time of flowering, thus suggesting an important role for postflowering infections. Moreover, accuracy of prediction models for disease severity and DON increased when they included weather variables from periods past flowering (Schaafsma and Hooker, 2003; Del Ponte et al., 2005). A possible explanation suggested by the current findings as well as the previous findings by Hart et al. (1984) is that DON can be produced from infections late in kernel development without producing dramatic reductions in grain weight, especially if multiple infection events occur during grain filling. This biological fact complicates the already challenging task of timely prediction and control of FHB and toxin contamination. If, in fact, the results of this study and others further confirm an extended phenological window of wheat vulnerability to FHB, especially for DON accumulation, then (i) wheat breeders would need to consider screening methods for resistance to DON production resulting from late infections; (ii) farmers would need to consider management strategies aimed to protect wheat heads from infection for several weeks rather than several days around flowering; and (iii) practices which use damaged kernels as a predictor

of DON may be less reliable in years when conditions allow for infection late in the growing season.

Acknowledgements

This research was conducted at Cornell University and was supported by Cornell Hatch Project NYC153433 and a fellowship from CNPq-Brazil that allowed Emerson Del Ponte, a former graduate student at the Federal University of Pelotas, to conduct research at Cornell. We gratefully acknowledge John Cianchetti and Stanley Kawamoto of Cornell for technical assistance; and Linda Geiger and Francois Lachance of Star of the West Milling Company in Churchville, NY for conducting the DON analyses reported in this paper.

References

Andersen AL. (1948) The development of *Gibberella zeae* head blight of wheat. *Phytopathology* **38**:595–611.

Argyris J, Sanford, DV, TeKrony D. (2003) *Fusarium graminearum* infection during wheat seed development and its effect on seed quality. *Crop Sci* **43**:1782–1788.

Atanasoff D. (1920) Fusarium-blight (scab) of wheat and other cereals. *J Agric Res* **20**:1–40.

Bai G, Shaner G. (1994) Scab of wheat: Prospects for control. *Plant Dis* **78**:760–766.

Cowger C, Sutton AL. (2005) *The Southeastern U.S. Fusarium Head Blight Epidemic of 2003*. Plant Health Progress online, doi:10.1094/PHP-2005-1026-01-RS.

Cromey MG, Lauren DR, Parkes RA, Sinclair KI, Shorter SC, Wallace AR. (2001) Control of Fusarium head blight of wheat with fungicides. *Australian J Plant Pathol* **30**:301–308.

Del Ponte EM, Fernandes JMC, Bergstrom GC. Fusarium head blight and eoxynivalenol accumulation in wheat inoculated at developmental stages from flowering through grain maturation. In: *Proceedings 2003 National Fusarium Head Blight Forum*. Bloomington, MN, East Lansing, Michigan State University 2003, pp. 129–132.

Del Ponte EM, Fernandes JMC, Pavan W. (2005) A risk infection simulation model for Fusarium head blight in wheat. *Fitopatol Bras* **30**:634–642.

Dill-Macky R, Jones RK. (2000) The effect of previous crop residues and tillage on Fusarium head blight of wheat. *Plant Dis* **84**:71–76.

Engle JS, Lipps PE, Graham TL, Bohem MJ. Effect of floral structure extracts and endogenous compounds on the growth of *Fusarium graminearum*. In: *Proceedings 2002 National Fusarium Head Blight Forum*. KY, USA, Erlanger, 2002, pp. 151–153.

Fernando WGD, Paulitz TC, Seaman WL, Martin RA. (1997) Fusarium head blight susceptibility of wheat inoculated at different growth stages. *Phytopathology* **87**:S30.

Hart LP, Pestka JJ, Liu MT. (1984) Effect of kernel development and wet periods on production of deoxynivalenol in wheat infected with *Gibberella zeae*. *Phytopathology* **74**:1415–1418.

Jones R, Mirocha CJ. (1999) Quality parameters in small grains from Minnesota affected by Fusarium head blight. *Plant Dis* **83**:506–511.

Khonga EB, Sutton JC. (1988) Inoculum production and survival of *Gibberella zeae* in maize and wheat residues. *Canadian J Plant Pathol* **10**:232–239.

Mandanhar JB, Cunfer BM. (1991) An improved selective medium for the assay of *Septoria nodorum* from wheat seed. *Phytopathology* **81**:771–773.

McMullen M, Jones R, Gallenberg D. (1997) Scab of wheat and barley: A re-emerging disease of devastating impact. *Plant Dis* **81**:1340–1348.

Mesterházy A, Bartók T, Téren J, Korom A. (1991) Resistance level and toxin contamination in wheat and corn after artificial inoculation. *Mycotoxin Research* **7**:64–67

Mesterházy A, Bartók T, Kászonyi G, Varga M, Tóth B, Varga J. (2005) Common resistance to different *Fusarium* spp. causing Fusarium head blight in wheat. *European J Plant Pathol* **112**:267–281.

Miller JD, Young JC. (1985) Deoxynivalenol in an experimental *F. graminearum* infection of wheat. *Can J Plant Pathol* **7**:132–135.

Pearce RB, Strange RN, Smith H. (1976) Glycinebetaine and choline in wheat: Distribution and relation to infection by *Fusarium graminearum*. *Phytochemistry* **15**:953.

Schaafsma AW, Hooker D. Predicting deoxynivalenol in wheat for Ontario. In: *Proceedings 2003 National Fusarium Head Blight Forum*. St Paul, MN, East Lansing, Michigan State University 2003, pp. 174.

Schaafsma AW, Tamburinc-Illincic L, Miller JD, Hooker DC. (2001) Agronomic considerations for reducing deoxynivalenol in wheat grain. *Can J Plant Pathol* **23**:279–285.

Schmale DG, Bergstrom GC. (2003) *Fusarium Head Blight. The Plant Health Instructor*. doi:10.1094/PHI-I-2003-0612-01.

Schroeder HW, Christensen JJ. (1963) Factors affecting resistance to wheat scab caused by *Gibberella zeae*. *Phytopathology* **53**:831–838.

Shaner GE. Epidemiology of Fusarium head blight of small grain cereals in North America. In: Leonard KL, Bushenell WR (eds), *Fusarium Head Blight of Wheat and Barley*. East Lansing, Michigan State University: APS Press, pp. 2003, 84–119.

Strange RN, Majer JR, Smith H. (1974) The isolation and identification of choline and betaine as the two major components in anthers and wheat germ that stimulate *Fusarium graminearum in vitro*. *Physiol Plant Pathol* **4**:277–290.

Takegami S. (1957) Studies on the resistance of wheat varieties to *Gibberella zeae* (Schw.) Petch. (head blight) and mechanism. I. Varietial difference of the position of florets in the wheat spikelet attacked by head blight incipiently and the relationships between existence of anther corpses in florets and the infection by head blights. *Sci Rep Agric Okayama Univ* **10**:33–42.

Trigo-Stockli DM, Curran SP, Pederson JR. (1995) Distribution and occurrence of mycotoxins in 1993 Kansas wheat. *Cereal Chem* **72**:470–474.

World Health Organization. (2001) Deoxynivalenol. In: *WHO Food Additives Series 47. FAO Food Nutr Pap* **74**:419–556.

Zadoks JC, Chang TT, Konzak CF. (1974) A decimal code for the growth stages of cereals. *Weed Res* **14**:415–421.