THE ASSOCIATION BETWEEN SOME *FUSARIUM* SPP. AND SEED QUALITY IN MAIZE (*ZEA MAYS* L.)

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ABSTRACT

The effect of delayed harvest on the occurrence and incidence of seed-borne *Fusarium* spp. and their effects on seed quality was investigated using four maize cultivars (Pioneer 3551, 3591, 3709 and 3475) over two seasons (1989/90, 1990/91) at Massey University, Palmerston North. As harvest was delayed from April to July, the percentage of cobs showing *Fusarium* mould increased. Cultivar 3551 tended to develop *Fusarium* cob mould later in the season (June) than the other three cultivars.

In both seasons the percentage of seeds of all four cultivars infected with *Fusarium* spp. increased as harvest was delayed. However, there was a difference between the two seasons; in 1989/90 the mean percentage of seeds carrying *Fusarium* spp. was 26%, 39%, 70% and 82% for April, May, June and July harvests respectively, while the corresponding levels for 1990/91 were 1%, 9%, 31% and 40% respectively. Between season differences were ascribed to climatic differences, the former season being wetter and warmer than the latter. There were only minor differences among cultivars for the percentage of seeds carrying *Fusarium* spp. *F. graminearum* was the species most consistently detected in all cultivars in both seasons, being recorded from 16%, 31%, 53% and 72% of seeds from the 1989/90 April to July harvests respectively, and from 0%, 6%, 25% and 30% of seeds from the same harvest times in 1990/91. *F. subglutinans*, *F. poae* and other *Fusarium* spp. were also detected, but their incidence was generally low.

Seed-borne *Fusarium* did not significantly reduce seed germination or vigour. In both seasons germination was between 86-99% for all cultivars. However, any dead seeds bore evidence of *F. graminearum* mycelial growth. Mycotoxins were recorded in seeds from all harvests in both seasons and mycotoxin levels increased as harvest was delayed. However, there were differences between seasons, as mean levels of Zearalenone, αZearalenol, Nivalenol and Deoxynivalenol ranged from 0.06 - 1.42 mg/kg seed in 1989/90, but from 0.0 - 0.54 mg/kg seed in 1990/91. In all cultivars and at most harvests in both years, levels of αZearalenol and of Nivalenol increased earlier than those of Zearalenone and Deoxynivalenol. Mycotoxin

differences among cultivars and the precise nature of the relationship between specific *Fusarium* species and mycotoxin development urgently requires further study, because of the potential for human and animal health problems.

Fusarium spp. from seed-culture colony were initially identified macroscopically on Malt Agar (MA), with pure cultures later being verified by the International Mycological Institute (UK). Subsequently, cultures were studied on Potato Dextrose Agar (PDA), Malt Extract Agar (MEA) and on Carnation Leaf Agar (CLA), with the final identity of seed-culture colonies being verified on CLA.

Colony texture and colour (including agar pigmentation) were initially used to separate *Fusarium* species detected on MA from infected seeds after harvest into a series of groups, ie 'red and fluffy', 'red centre', 'red and lobed', 'cream and fluffy', and 'cream and lobed' for *F. graminearum*. *F. crookwellense* was also separated as a 'red centre' type of colony while *F. culmorum* was separated as 'cream and flat', *F. subglutinans* 'purple and strands' type, and *F. poae* as 'purple/white/cream and powdery' type. While it was possible to differentiate the five types of *F. graminearum* on MA, it was not possible to distinguish *F. graminearum* 'red centre' type from *F. crookwellense*, although *F. culmorum* was relatively easy to differentiate from *F. graminearum* and *F. crookwellense*. Use of PDA or MEA pure cultures to differentiate *F. graminearum* from *F. crookwellense* or *F. culmorum* was not successful because the colony morphology of these three species was similar. However, *F. subglutinans* and *F. poae* were readily identified macroscopically on MA and MEA.

F. graminearum seed-culture colonies which did not sporulate on MA or MEA in most cases readily formed perithecia of *Gibberella zeae* on CLA (in the presence of 40W NUV light) regardless of whether the cultures were initiated by single germinated spores or by mass transferred inoculum. Those colonies which did sporulate on MA or MEA formed abundant sporodochia on CLA but not perithecia. CLA was also used to identify *F. graminearum* (*G. zeae*) from maize seeds or

seedlings by direct plating of these structures after surface disinfection. Full descriptions of the *Fusarium* colonies on the various media used are presented.

Fusarium survival in seed during storage depended upon seed moisture content (SMC) and storage temperature. *F. graminearum* was eliminated from seed at 14% SMC stored at 30°C and 25°C after 3 or 6 months storage, respectively, but survived at low levels (1-5%), together with *F. subglutinans* (1-7%), *F. poae* (1-2%) at these temperatures and 10% SMC. *F. subglutinans* and *F. poae* in seeds at 14% SMC did not survive after 9 months storage at 30°C. In seed stored at 5°C, *Fusarium* spp. infection levels did not decline after 12 months of storage at both 10 and 14% SMC. These results suggest a possible control strategy for producing *Fusarium* free seed, providing seed moisture content is not greater than 10%. At a storage temperature of 30°C, the post-storage germination of seed at 14% SMC had dropped to under 10% within 3 months, but seed at 10% SMC maintained its germination (88-97%) throughout the storage trial. After 12 months seed storage at 5°C (sealed storage) or 25°C (open storage), mycotoxin levels were similar to pre-storage levels.

The requirements of Koch's postulates were fulfilled in demonstrating that seed-borne *F. graminearum* was transmitted from maize seeds to seedlings under aseptic conditions in a glasshouse maintained at a temperature of 14°C to 17°C. The mean transmission rate (48%) was similar to the original seed-borne inoculum which suggests that under favourable environmental conditions, the pathogen will be effectively transferred from the seed to seedlings. *F. graminearum* had little effect on seedling emergence or survival, but was associated with a high percentage of seedlings with scutellum-mesocotyl/scutellum-main root lesioning. In the field, *F. graminearum* was consistently isolated from seedlings, but seed transmission could not be confirmed because of the presence of soil-borne inoculum, ie the pathogen was isolated from up to 37% of seedlings from a seed lot which carried only 1% seed-borne inoculum.

F. subglutinans was also proved to be seed transmitted under the same glasshouse conditions as described for *F. graminearum*. The significance of surface-

borne inoculum of this pathogen was demonstrated in that the mean transmission rate for non-surface disinfected seed lots was 81%, whereas it was only 7% for surface disinfected seed lots. *F. subglutinans* was associated mainly with 'above sand level' seedling infection (coleoptile-node infection, leaf/shoot blight, shoot wilt and seedling stunting). However, *F. subglutinans* was rarely detected in seedlings from the field, possibly because of the antagonistic effects of mycopathogenic fungi such as *Gleocladium roseum*.

These results are discussed, particularly in relation to the significance of *F. graminearum* and *F. subglutinans* as seed-borne pathogens of maize, and the difficulties inherent in the identification of *Fusarium* spp. following seed health testing. It is likely that these seed-borne *Fusarium* spp. are more important because of their association with mycotoxins, than with any effects they have as an inoculum source for diseases of maize.

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