

Fusarium spp. in Wheat Grain in the Czech Republic Analysed by PCR Method

JAN NEDĚLNÍK¹, HANA MORAVCOVÁ¹, JANA HAJŠLOVÁ², KATEŘINA LANCOVÁ²,
MARIE VÁŇOVÁ³ and JAROSLAV SALAVA⁴

¹Research Institute for Fodder Crops, Ltd., Troubsko, Czech Republic; ²Department of Food Chemistry and Analysis, Faculty of Food and Biochemical Technology, Institute of Chemical Technology Prague, Prague, Czech Republic; ³Agricultural Research Institute, Ltd., Kroměříž, Czech Republic; ⁴Crop Research Institute, Prague-Ruzyně, Czech Republic

Abstract

NEDĚLNÍK J., MORAVCOVÁ H., HAJŠLOVÁ J., LANCOVÁ K., VÁŇOVÁ M., SALAVA J. (2007): *Fusarium* spp. in wheat grain in the Czech Republic analysed by PCR method. Plant Protect. Sci., 43: 135–137.

The frequency of occurrence of four *Fusarium* spp. on wheat in the Moravia region, Czech Republic, was determined by polymerase chain reaction (PCR). Grain samples were collected during 2003–2006 at grain purchase centres. The dominant species was *F. graminearum*, which was recorded in all samples of the first 3 years of the study and in 88% of them in 2006. The previously more frequent *F. culmorum* was detected in 100% of the samples only in 2005; in the preceding two years the frequency of its detection was lower, 84% and 60%, and in 2006 it was detected in 55% of the samples. *Fusarium avenaceum* had a very low occurrence in the years 2003–2004, but in 2005 it was recorded in 100% of the samples. In 2006 it was the opposite – total absence of this species. A quite different situation was found in the occurrence of the fourth species – *F. poae*. In the years 2005 and 2006 it was only detected in 10%, resp. 2% of the samples, compared to markedly higher occurrences in the previous years. A comparison of the current weather development with the long-term mean at the Troubsko locality suggests that years with a relatively long, wet and cold start of the growing season and warmer end of vegetation (late May–July) will favour *F. graminearum*.

Keywords: *Fusarium* spp.; PCR; detection; wheat

A complex of *Fusarium* spp. causes Fusarium head blight (FHB) on wheat and also on barley. Infection with FHB results not only in yield loss, but also causes depreciation of the harvested product due to the accumulation of toxins such as deoxynivalenol produced by *Fusarium* spp. The flowering time is a very susceptible period for primary infection. One reason might be that during this period spores can get into the opened wheat florets where they may later cause infection.

Initial symptoms of infection of a wheat ear are visible whitening and drying of individual spikelets or entire parts of the ear. If the florets became infected shortly after blooming, grains are often not formed, while other infected spikelets have grains with various deformities, shrunken and with pink-white coloration (KLEM & TVARŮŽEK 2005).

The main aim of experiments was detection of *Fusarium* species on wheat grains by using of the PCR methods.

Partly supported by the Ministry of Agriculture of the Czech Republic, Project No. QF 3121.

This paper summarises the results of a survey for four *Fusarium* species in collaboration with the laboratory of molecular genetics of the Crop Research Institute, Prague-Ruzyně. The species were detected by using one of the methods of PCR; for *Fusarium avenaceum* the method of TURNER *et al.* (1998) was used, *F. culmorum* and *F. graminearum* were detected according to SCHILLING *et al.* (1996), while *F. poae* was detected using the method of PARRY and NICHOLSON (1996). The grain samples of different varieties of wheat (total number 160 samples) came from 3 grain purchase centres in southern Moravia and were collected in the years 2003–2006. The representative samples were collected in compliance with the directive on sampling. As the next step the grain was milled and stored in a deep freeze (Table 1).

PCR confirmed the dominance of the species *F. graminearum*, which was recorded in more than 88% of samples in all the years of study. The previous more frequent species *F. culmorum* was only detected in 100% of samples in the year 2005; in the preceding two years the frequency of its detection was lower, being in the range of 60–84%. In 2006 *F. culmorum* was detected in 55% samples. In the years 2003–2004 there was a very low occurrence of *F. avenaceum*, but in the year 2005 this species was recorded in 100% of samples. An opposite situation was in 2006 – total absence of this species. A quite different situation was found in the occurrence of the fourth species – *F. poae*. In the years 2005 and 2006 it was only detected in 10%, resp. 2% of samples, compared to a markedly higher occurrence in the previous years.

Although wheat samples for PCR analysis came from various localities of southern and central Moravia, it is possible to reveal indirectly some relationships by analysing weather development. The comparison of the current weather development with the long-term mean in the Troubsko locality suggests that years with a relatively long, wet and cold start of the growing season and

warmer end of vegetation (late May–July) will favour *F. graminearum*.

Europe has recently witnessed considerable changes in the proportions of incidence of different species of the genus *Fusarium*. However, the effect of some factors favourable for FHB may result in changes of the species profile from region to region (BOTTALICO & PERRONE 2002). In colder maritime areas of NW Europe the most widespread species causing FHB were *F. culmorum*, *F. graminearum*, *F. avenaceum* and *F. poae*. Some observations, however, signalled an increase in the frequency of the occurrence of *F. culmorum* and higher importance of the species *F. poae* and *F. avenaceum*, mainly in the years less favourable for infection by *F. graminearum* (PARRY *et al.* 1995). In Poland the dominant species were *F. poae* and *F. tritinctum* (GOLINSKI *et al.* 1997). The occurrence of *F. avenaceum*, which earlier had been uncommon in colder areas, is confirmed by analyses from the Netherlands (WAALWIJK & CHELKOWSKI 2003). Detailed information is available from Italy. *Fusarium graminearum* and *F. culmorum* were the dominant species if at the time of blooming the weather was warmer and wetter, whereas in drier seasons *F. culmorum* occurred more frequently (BOTTALICO & PERRONE 2002). At some Slovak localities the dominant and most frequent species causing FHB on wheat were *F. graminearum*, *F. culmorum* and *F. avenaceum*, less frequent were *M. nivale* and *F. poae* (HUDEC 2006).

The occurrence of fusaria is significantly affected by environmental factors (temperature, precipitation, air humidity). According to the results from many studies it seems that *F. graminearum* is the dominant species in moist and warmer continental conditions, the other two often isolated species *F. culmorum* and *F. avenaceum* are more frequent in colder seasons. The same trend in the shift of the species spectrum was also recorded in the Czech Republic. This conclusion is supported by some results presented at the 9th European

Table 1. Occurrence of *Fusarium* spp. on wheat grain

Number of samples	<i>F. avenaceum</i>				<i>F. culmorum</i>				<i>F. graminearum</i>				<i>F. poae</i>			
	2003	2004	2005	2006	2003	2004	2005	2006	2003	2004	2005	2006	2003	2004	2005	2006
Tested	32	42	41	45	32	42	41	45	32	42	41	45	32	42	41	45
Positive	8	8	41	0	27	25	41	25	32	42	41	40	26	14	4	1

Fusarium Seminar, which was held in the year 2006 in Wageningen, the Netherlands. This is, however, a generalisation of many findings so it is important to note that the species spectrum may also be affected by a number of other factors, including species composition. The breeding of more resistant cultivars is the most important task to decrease the safety risk of food and feed. Resistance to *Fusarium* is race non-specific, effective against all *Fusarium* species and to all members of the *F. graminearum* clade that represents at least nine different species (lineages; MESTERHÁZY *et al.* 2005, 2006), and lineages have also been observed in *F. culmorum* (TÓTH *et al.* 2004).

References

- BOTTALICO A., PERRONE G. (2002): Toxigenic *Fusarium* species and mycotoxins associated with head blight in small-grain cereals in Europe. *European Journal of Plant Pathology*, **108**: 611–624.
- GOLINSKI P., PERKOWSKI J., KOSTECKI M., GRABARKIEWICZ-SZCESNA J., CHELKOWSKI J. (1997): *Fusarium* species and *Fusarium* toxins in wheat in Poland – a comparison with neighbour countries. *Sydowia* (Special Issue March 1997), **48**: 12–22.
- HUDEK K. (2006): Influence of locality and year on *Fusarium* species occurrence in *Fusarium* head blight and stalk rot of winter wheat. *Agriculture*, **52**: 69–76.
- KLEM K., TVARŮŽEK L. (2005): Klasové fuzariózy na ozimé pšenici – rizika infekce a možnosti ochrany. *Obilnářské listy*, **8**: 59–61.
- MESTERHÁZY A., BARTÓK T., KÁSZONYI G., VARGA M.T., TÓTH B., VARGA J. (2005): Common resistance to different *Fusarium* spp. causing *Fusarium* head blight in wheat. *European Journal of Plant Pathology*, **112**: 267–281.
- MESTERHÁZY A., TÓTH B., KÁSZONYI G., SZÁBO-HEVÉR A., BARTÓK T., SZEKERES A. (2006): Methodological background, QTLs and breeding for resistance for FHB resistance in wheat. In: Book of Abstracts 9th European *Fusarium* Seminar, 19.–22. 9. 2006, Wageningen: 86.
- PARRY D.W., JENKINSON P., MC LEOD L. (1995): *Fusarium* ear blight (scab) in small-grain cereals – a review. *Plant Pathology*, **44**: 207–238.
- PARRY D.W., NICHOLSON P. (1996): Development of a PCR assay to detect *Fusarium poae* in wheat. *Plant Pathology*, **45**: 383–391.
- SCHILLING A.G., MÖLLER E.M., GEIGER H.H. (1996): Polymerase chain reaction-based assays for species-specific detection of *Fusarium culmorum*, *F. graminearum* and *F. avenaceum*. *Molecular Plant Pathology*, **86**: 515–522.
- TÓTH B., MESTERHÁZY A., NICHOLSON P., TÉREN J., VARGA J. (2004): Mycotoxin production and molecular variability of European and American *Fusarium culmorum* isolates. *European Journal of Plant Pathology*, **110**: 587–599.
- TURNER A.S., LEES A.K., REZANOOR H.N., NICHOLSON P. (1998): Refinement of PCR-detection of *Fusarium avenaceum* and evidence from DNA marker studies for phenetic relatedness to *Fusarium tricinctum*. *Plant Pathology*, **47**: 278–288.
- WAALWIJK C., CHELKOWSKI J. (2003): Major changes in *Fusarium* spp. in wheat in the Netherlands. *European Journal of Plant Pathology*, **109**: 743–754.

Received for publication March 26, 2007

Accepted after corrections October 29, 2007

Corresponding author:

RNDr. JAN NEDĚLNÍK, Ph.D., Výzkumný ústav pícninářský, spol. s r. o., Troubsko, Zahradní 1, 664 41 Troubsko, Česká republika
tel.: + 420 547 227 380, fax: + 420 547 227 385, e-mail: nedelnik@vupt.cz