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THE MYCOTOXIN OCCURRENCE IN DIFFERENT GENOTYPES OF *TRITICUM MONOCOCCUM* AND *TRITICUM DICOCCUM* FROM ORGANIC FARMING

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Abstract

The presence of mycotoxins: aflatoxins, OTA, T-2 toxin and DON was determined with enzyme-linked immunosorbent assay (ELISA) in *Triticum monococcum* and *T. dicoccum* genotypes. Among three of the examined cultivars of *T. monococcum,* DON was identified in all samples (up to 0.31 mg/kg) and T-2 toxin was identified in one sample (179.6 μg/kg). Among three of the examined spring *T. dicoccum* cultivars DON was identified in two samples (up to 0.3 mg/kg), while T-2 toxin, aflatoxins and ochratoxin A were not found. Among five of the examined winter *T. dicoccum* cultivars only DON was identified in three samples (up to 0.3 mg/kg), while aflatoxins, OTA or T-2 toxin were not found.

Key words: *Triticum monococcum*, *Triticum dicoccum,* deoxynivalenol, T-2 toxin, aflatoxins, ochratoxin A

Introduction

The increasing interest in organic farming and healthy food products led to a comeback of hulled wheat species growing such as emmer (*Triticum dicoccum*) and *Triticum monococcum* (De Vita et al. 2006). Knowledge of quality traits, as content of mycotoxins, in these cereals is required by consumers. Mycotoxins are secondary metabolites produced by different species of fungi and they contaminate cereals grain. Most frequent mycotoxins occurring in cereals are trichothecenes produced by *Fusarium* spp. but also aflatoxins and ochratoxins produced by the genera *Penicillium* and *Aspergillus*.

The aim of the study was determination of mycotoxins (aflatoxin B1 and B2, ochratoxin A, T-2 toxin and deoxynivalenol) in kernels of *Triticum monococcum* and *Triticum dicoccum* genotypes.

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Material and methods

Samples

Three *T. monococcum* cultivars, three *T. dicoccum* spring cultivars and five *T. dicoccum* winter cultivars collected in 2008 from the organic experimental farm Chwałowice were examined for aflatoxins, ochratoxin A, deoxynivalenol, and T-2 toxin content with enzyme-linked immunosorbent assay (ELISA).

ELISA kits

Commercial ELISA kits: AgraQuant Total Aflatoxin Assay, AgraQuant T-2 Toxin Assay, AgraQuant Ochratoxin Assay, AgraQuant DON Assay were used in the study. These ELISA assays kits are direct competitive immunoassays with horseradish peroxidase conjugate. Kits basic characteristics are presented in Table 1.

Table 1

Basic characteristic of AgraQuant ELISA kits

AgraQuant® Test Kit	Quantitation range	Limit of detection
Total aflatoxin	1–20 ppb	1 ppb
Deoxynivalenol	0.25–5 ppm	0.2 ppm
Ochratoxin	2–40 ppb	2 ppb
T-2 toxin	75–500 ppb	35 ppb

The assays were performed according to the procedure described in the AgraQuant Assay kit manual. Ground samples (20 g) were extracted with 100 ml of 70% methanol in a ratio of 1:5 (w:v) for T-2 toxin, ochratoxin A and aflatoxins and with 100 ml of distilled water for deoxynivalenol. Samples were shaken for 3 min and filtered through Whatman No. 1 paper. After that samples were diluted in a ratio of 1:10 with distilled water for T-2 toxin and 1:5 with distilled water for deoxynivalenol. According to the procedure there is no need for dilution of samples for aflatoxins and ochratoxin A determination.

100 μl of diluted extracts and 100 μl of extracts without dilution were further used in the procedure. Absorption in microwells was measured with a Tecan Sunrise microwell reader using a 450 nm absorbance filter.

Results

In all examined *T. monococcum* cultivars deoxynivalenol was identified in quantities from 0.3 to 0.31 mg/kg, T-2 toxin was identified in one sample (up to 179.6 μg/kg), while aflatoxins and ochratoxin A were not found. In the tested samples deoxynivalenol was identified in two of three examined samples of flour prepared

from cultivars of spring *T. dicoccum* at the concentration of 0.3 mg/kg each. T-2 toxin, aflatoxins and ochratoxin A were not found in these samples (Table 2). Among the five examined cultivars of winter *T. dicoccum* deoxynivalenol was identified in three samples (0.28–0.3 mg/kg), T-2 toxin, aflatoxins and ochratoxin A were not found in the flour prepared from *T. dicoccum* grain (Table 2).

Table 2

The occurrence of mycotoxins in flour prepared from cultivars of *Triticum dicoccum* and *Triticum monococcum*

Cultivar	DON (mg/kg)	T-2 (μg/kg)	Aflatoxins (μg/kg)	OTA (μg/kg)
Triticum monococcum				
T. monococcum b6 1195	0.3	0	0	0
T. monococcum l. *hornemanii* 5040	0.31	0	0	0
T. monococcum l. *hornemanii* b6 5007	0.3	179.6	0	0
Triticum dicoccum, spring				
PL 20757	0.3	0	0	0
PL 21799 IHAR	0	0	0	0
PL 20762	0.3	0	0	0
Triticum dicoccum, winter				
T. dicoccum B6 5029	0.3	0	0	0
T. dicoccum B6 1306	0	0	0	0
T. dicoccum B6 5028	0.29	0	0	0
T. dicoccum B6 5049	0.28	0	0	0
T. dicoccum B6	0	0	0	0

Discussion

Trichothecenes are ubiquitous natural contaminants occurring in all cereals cultivated in the temperate climate zone. These compounds are potent inhibitors of eukaryotic protein synthesis (Beyer et al. 2009). Deoxynivalenol, representing trichothecene B group, was a dominating *Fusarium* mycotoxin found in most of *T. dicoccum* and *T. monococcum* genotypes examined in the study, nevertheless, its levels were fairly below maximum limit established in the EU for this toxin in cereals. The presence of T-2 toxin, belonging to trichothecene A group, was documented only in one *T. monococcum* genotype but its level was very high. The occurrence of T-2 and HT-2 toxins in cereals implies health risk for consumers (Beyer et al. 2009). Among the different factors that are relevant for reduction of trichothecenes accumulation, choice of cultivar can be the determinant (Bountigny et al. 2008). Among the eight spring and winter *T. diccocum* genotypes tested, there were differences in contamination with DON. Three of them were free of the mycotoxin. Besides, none of the *T. diccocum* genotypes was contaminated by any other tested mycotoxin, i.e. T-2 toxin, aflatoxins or ochratoxin A. In the study on the occur-

rence of nine mycotoxins and on contamination by pre- and postharvest fungal pathogens of cereals in samples of stored *T. monococcum*, *T. dicoccum*, and *T. spelta* (spelt) different mycotoxins content or lack of them was found in all species, which collectively referred to as *faro* (Castoria et al. 2005). Differences of *T. dicoccum* cultivars in mycotoxin contamination resulted from different reaction to Fusarium head blight. Among the evaluated genotypes of *T. dicoccum* significant genetic diversity concerning reaction to fungal spread was found (Buerstmayr et al. 2003, Oliver et al. 2007). The results obtained here showed, that the choice of *T. dicoccum* cultivars without the presence of main trichothecenes is possible, as well as *T. monococcum* cultivars only without T-2 toxin contamination. These cultivars should be recommended for cultivation in organic farming.

Some authors claimed that mycotoxins produced by postharvest fungi are particularly dangerous to organic cereals. A serious threat to cereals in temperate climate zone constitutes ochratoxin A (Elmholt and Rasmussen 2005). The lack of either ochratoxin A or aflatoxins in grain samples of *T. monococcum* and *T. dicoccum* cultivars could be an evidence of good postharvest practices.

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