Czech Journal of Food Sciences, 39, 2021 (2): 131–139 *Original Paper*

https://doi.org/10.17221/247/2020-CJFS

Mycotoxins in oat flakes – changes during production and occurrence on the Czech market

Tomas Dropa[1]*, *Zbynek Dzuman*[2], *Petra Jonatova*[2]

[1]*National Institute for Nuclear, Biological and Chemical (NBC) Protection, Milín, Czech Republic*
[2]*Department of Food Analysis and Nutrition, University of Chemistry and Technology Prague, Prague, Czech Republic*

**Corresponding author: dropa@sujchbo.cz*

Citation: Dropa T., Dzuman Z., Jonatova P. (2021): Mycotoxins in oat flakes – changes during production and occurrence on the Czech market. Czech J. Food Sci., 39: 131–139.

Abstract: Raw oats are frequently contaminated by fungi producing mycotoxins, and as such, they can pose a health risk to humans regularly consuming oat products. To reveal the effect of particular processing steps on contamination levels in final oat products, two series of samples obtained from an oat flake manufacturer were analysed. Among oat fractions treated, the most significant mycotoxin decrease was demonstrated in cleaning and dehulling steps, where the observed mycotoxin reduction was in the range of 13–75% for monitored mycotoxins, enniatin B, enniatin B1, HT-2 toxin, T-2 toxin, beauvericin and tentoxin. The overall mycotoxin reduction found in final oat flakes was in the range of 65–100%. Furthermore, 20 commercially available oat flakes from the Czech market were analysed. We detected 16 mycotoxins, mainly trichothecenes and emerging enniatins with levels in the interval of 1–156 μg kg^{-1}. With regard to the current EU legislation, two analysed oat flake products exceeded the maximum limit for ochratoxin A.

Keywords: fungal pathogens; cereal contamination; oat processing

Oats have been grown for thousands of years. Until the 19th century, oats were mainly used for animal feeding and practically were not accepted as an ingredient of human diet. It is speculated that oat introduction into Europe may have been caused by contamination of the preferably grown species, wheat and barley, and consecutive oat plant adaptability (Zohary et al. 1988). To date, there are 27 known species of the genus *Avena* around the world, while *A. sativa* (hulled oat) and *A. nuda* (naked oat) are dominant oat species. *A. sativa* is the most economically important oat worldwide with average year production of 33 million metric tons (Strychar et al. 2011). Due to highly valuable nutrients such as proteins, non-saturated lipids, oils, minerals, or soluble fibre, oats have significant importance in the present human diet. Containing many bioactive compounds, oat products are considered to have notable health effects as well. Beta-glucans, for instance, have proven effectivity in preventing diabetes and cardiovascular diseases, anti-inflammatory as well as cholesterol lowering properties of oat antioxidants avenanthramides have also been reported previously (Peterson et al. 2002; Liu 2007; Wood 2011; Grundy et al. 2018).

Oat plants and grains, however, similarly like any other cereal crops, can be attacked by a variety of diseases and, consequently, mycotoxin contamination can occur. Besides yield reduction accompanied by economic losses, fungal contamination in oats can seriously affect consumers, either through acute food poisoning, induction of chronic disease, or systemic damage. Among the most important fungi attacking oats belong *Ustilago avenae* causing powdery smut, *Puccinia coronata* causing crown rust, or fungi of the genus *Fusarium* (Kiecana et al. 2005). All *Fusarium* species, depending on current environmental conditions, are able to produce one or more mycotoxins, while many of them produce trichothecenes (Edwards 2004). Transfer of the mycotoxins as well as subsequent contamination of cereal products have been reported in many studies during the last decades (Alshannaq and Yu 2017). According to an extensive literature search conducted by Palumbo et al. (2020) focusing on the occurrence of mycotoxins in cereals

https://doi.org/10.17221/247/2020-CJFS

and derived products from Europe, frequent contamination with type A trichothecenes, i.e. HT-2 and T-2 toxins (HT-2, T-2), was reported contrary to other cereals indicating a potential risk to consumers while other examined cereals, wheat and maise, were connected with common mycotoxin co-occurrence. One of the most comprehensive studies related to oats as well as retail oat products was focused on trichothecenes and other mycotoxins (Pettersson et al. 2011). The study reported frequent co-occurrence of T-2 and HT-2 at non-negligible levels in 243 raw oats, 529 oat flakes, 105 oatmeals, and 209 oat by-products.

With regard to the facts mentioned above, there is still a concern that mycotoxin contamination of oat products can endanger consumer health. The objective of the present study is *i*) to reveal the mycotoxin fate within oat flake production, focusing attention on initial raw oat contamination, as well as on mycotoxin levels in single fractions emerging through the flake production chain and *ii*) to provide a complex insight into mycotoxin occurrence within oat flake products available on the Czech retail market.

MATERIAL AND METHODS

Oat processing chain. During direct harvesting of oats in the field, the matured crop with the moisture content between 12% and 14% is cut and threshed in one processing step. The usual subsequent processing chain of raw oats is schematically summarised in Figure 1.

A standardised procedure of oat flake production employed in the production facility consists of oat sieve cleaning followed by grain dehulling and scouring. The material obtained passes through a kilning machine and after steaming the flaking step follows, which results in the final product (Webster and Wood 2011; Menon et al. 2016). The Czech oat flake manufacturer involved in the study employs the corresponding technology of oat processing.

Reagents, analytical standards. Chemicals used in this study comprising acetonitrile, methanol, formic acid, ammonium formate, and ammonium acetate [all liquid chromatography-mass spectrometry (LC-MS) grade; Merck, Germany], sodium chloride and magnesium sulphate (both 99.9%) were purchased from Merck (Germany). Deionised water (18 MΩ) was produced by a Milli-Q system (Millipore, USA). Analytical standards of 57 *Fusarium*, *Aspergillus*, *Penicillium*, *Alternaria*, *Claviceps*, *Stachybotrys*, and *Phomopsis* mycotoxins (purity ≥ 96%) were obtained from Merck (Germany) and Romer Labs (Austria). A composite mixture of 1 μg mL^{-1} in acetonitrile was prepared and used for the method validation and quantification of mycotoxins in samples.

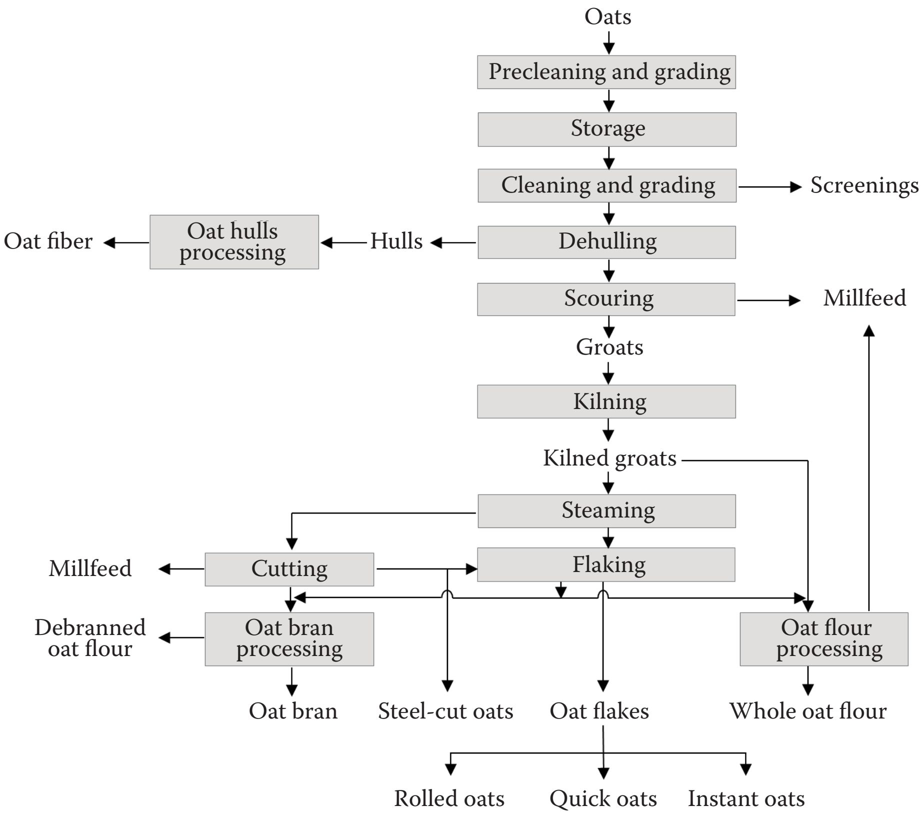

Figure 1. A schematic of the raw oat processing chain

Samples. The samples of two batches from the oat flake production (5 samples each) were obtained from a local Czech manufacturer together with information regarding the process mass balance and quantity of production and analysed for the presence of 57 mycotoxins. Apart from that, a set of oat flake samples (*n* = 20) purchased on the Czech retail market was examined to evaluate the degree of mycotoxin contamination.

Sample preparation. The employed QuEChERS-like (quick, easy, cheap, effective, rugged, safe) procedure according to Dzuman et al. (2014) comprised the following steps: *i*) sample weighing (2.00 g), *ii*) addition of acidified water (0.2% formic acid), *iii*) matrix soaking (30 min), *iv*) addition of acetonitrile (10 mL), *v*) shaking on a lab shaker (240 RPM, 30 min, IKA KS 130; IKA Labortechnik, Germany), *vi*) addition of NaCl (1 g) and $MgSO_4$ (4 g), *vii*) vigorous handshaking (1 min), *viii*) centrifugation (13 081 g, 5 min; Rotina R380, Hettich, Germany), and *ix*) extract removal into an amber glass vial.

Method validation and calibration. Quantification of analytes was performed using matrix-matched calibration standards (*n* = 11) prepared in the range of 0.1–200 ng mL^{-1} corresponding to 0.5–1 000 μg kg^{-1} (calibration points 0.5; 1; 2.5; 5; 10; 25; 50; 100; 250; 500; and 1 000 μg kg^{-1}) using internal blank oat flake sample extract (mycotoxin-free). Method recovery and repeatability were determined using a spiked mycotoxin-free sample at 250 μg kg^{-1} (corresponding to 50 ng mL^{-1}; *n* = 6).

LC-MS analysis. Mycotoxin analyses were performed using an ultra-high performance liquid chromatograph Acquity UPLC™ System (Waters, USA) coupled to tandem mass spectrometer (U-HPLC-MS/MS) QTRAP® 5500 (Sciex, Canada) equipped with quadrupole-linear ion trap mass analysers, electrospray ionisation (ESI) interface and Acquity UPLC™ HSS T3 analytical column (100 × 2.1 mm, 1.8 μm; Waters, USA). Separate instrumental methods in positive and negative ionisation mode (ESI+/−) in multiple reaction monitoring mode (MRM) were used for analyte detection (Dzuman et al. 2014). Injection volume was 4 μL, temperature of autosampler and analytical column was set to 10 °C and 40 °C, respectively. As mobile phases, 5 mM ammonium formate and 0.2% formic acid in water (A1) and methanol (B1) were used in ESI(+) and 5 mM ammonium acetate in water (A2) and methanol (B2) in ESI(−). The elution gradient was identical for both instrumental methods (12 min run time); it started at 10% B1/B2 (0.35 mL min^{-1}) with linear increase to 50% of B1/B2 in 1 min, lower gradient to 8 min followed by another linear increase of mobile phase composition to 100% of B1/B2 and flow rate (0.50 mL min^{-1}). The column was then washed for 2 min (0.50 mL min^{-1}) and reconditioned to initial conditions for 2 min.

RESULTS AND DISCUSSION

Method validation. In the first phase, the employed analytical method combining QuEChERS-like analyte isolation and U-HPLC-MS/MS detection was validated for oat flakes (Dzuman et al. 2014). The validation resulted in obtaining good performance parameters of the method which are summarised in Table 1. Recoveries were in the range of 68–108% [with the exception of 37% for highly polar deoxynivalenol-3-glucoside (D3G)], repeatabilities

Table 1. Method performance characteristics of the employed U-HPLC-MS/MS method

No.	Mycotoxin	Abbreviation	Recovery (%)	RSD (%)	Limit of quantification (μg kg^{-1})
1	15-acetyldeoxynivalenol	15-ADON	86	7	25.0
2	3-acetyldeoxynivalenol	3-ADON	96	2	10.0
3	aflatoxin B1	AFB1	78	9	0.5
4	aflatoxin B2	AFB2	77	1	0.5
5	aflatoxin G1	AFG1	81	4	0.5
6	aflatoxin G2	AFG2	82	5	1.0
7	agroclavine	A-clavine	77	5	5.0
8	alpha-zearalenol	α-ZOL	92	2	5.0
9	alternariol	AOH	97	2	0.5
10	alternariol-methylether	AME	93	3	0.5
11	beauvericin	BEA	82	6	1.0
12	beta-zearalenol	β-ZOL	95	1	5.0
13	citrinin	CIT	91	1	10.0
14	cyclopiazonic acid	CPA	77	8	100.0

https://doi.org/10.17221/247/2020-CJFS

Table 1 to be continued

No.	Mycotoxin	Abbreviation	Recovery (%)	RSD (%)	Limit of quantification (µg kg^{-1})
15	deoxynivalenol	DON	92	3	10.0
16	deoxynivalenol-3-glucoside	D3G	37	7	10.0
17	diacetoxyscirpenol	DAS	83	7	10.0
18	enniatin A	EnnA	83	4	0.5
19	enniatin A1	EnnA1	86	4	0.5
20	enniatin B	EnnB	84	7	0.5
21	enniatin B1	EnnB1	83	9	0.5
22	ergocornine	E-cornine	81	9	25.0
23	ergocorninine	E-corninine	86	9	25.0
24	ergocristine	E-cristine	86	10	25.0
25	ergocristinine	E-cristinine	89	12	25.0
26	ergocryptine	E-cryptine	74	1	25.0
27	ergocryptinine	E-cryptinine	76	3	25.0
28	ergometrine	E-metrine	82	2	5.0
29	ergosine	E-sine	77	6	25.0
30	ergosinine	E-sinine	78	2	25.0
31	ergotamine	E-amine	92	2	25.0
32	ergotaminine	E-aminine	83	5	25.0
33	fumonisin B1	FB1	78	3	50.0
34	fumonisin B2	FB2	80	2	50.0
35	fumonisin B3	FB3	76	4	50.0
36	fusarenon X	FUS-X	84	4	50.0
37	gliotoxin	GLIO	74	4	50.0
38	HT-2 toxin	HT-2	93	4	5.0
39	meleagrin	MEL	83	1	1.0
40	mycophenolic acid	MPA	75	4	5.0
41	neosolaniol	NEO	89	3	10.0
42	nivalenol	NIV	72	4	100.0
43	ochratoxin A	OTA	91	8	2.5
44	patulin	PAT	73	2	100.0
45	paxilline	PAX	96	4	25.0
46	penicillic acid	PenAc	89	5	50.0
47	penitrem A	PEN-A	71	1	50.0
48	phomopsin A	Phom A	68	1	100.0
49	roquefortine C	ROQ-C	88	5	5.0
50	stachybotrylactam	STACH	84	9	25.0
51	sterigmatocystin	STER	75	5	1.0
52	T-2 toxin	T-2	91	8	1.0
53	tentoxin	TEN	90	7	0.5
54	tenuazonic acid	TEA	74	5	250.0
55	verrucarol	VER	94	6	100.0
56	verruculogen	Verruc	89	7	100.0
57	zearalenone	ZEA	99	3	0.5

U-HPLC-MS/MS – ultra-high-performance liquid chromatography tandem mass spectrometry; RSD – relative standard deviation in the range of 1–12% and limits of quantification were in a relatively wide range of 0.5–250 μg kg^{-1} due to a considerable variability of mycotoxins with regard to their physicochemical properties.

Processing chain influence on the present mycotoxin levels. For the experiments performed, ten oat-based materials, representing two batches of initial oats as well as respective intermediates (fractions) coming from oat flake production, were obtained from a local manufacturer. Considering technical possibilities as well as restrictions on the manufacturer's site, sampling of materials was carried out *i*) in input tray, raw oats; *ii*) after oat cleaning and dehulling; *iii*) after oat steaming and cutting; *iv*) in the middle of the processing line and *v*) in output tray, oat flakes. The overview of mycotoxin levels obtained from analyses of the oat-based materials and products is summarised in Table 2. Mycotoxin analyses were carried out in four replicates to avoid obtaining outlying data which may result in mass balance misinterpretation. It should be noted that relative standard deviation (RSD) < 15% was observed in all analyte-matrix combinations and therefore average data are presented (Table 2). According to the conclusions of previously performed studies (Marchelli and Suman 2010; Kostelanska et al. 2011; De Angelis et al. 2013; Tibola et al. 2015), processing steps have a significant potential to influence levels of mycotoxins in cereal products intended for direct human consumption as well as for other food production. Balance of mycotoxin levels in the actual oats and oat materials undergoing single processing steps in the production line is shown in Figure 2 (batch A) and Figure 3 (batch B), respectively. A certain reduction of the mycotoxin content as a result of particular processing steps was observed in the study. Compared to initial oats (100% in the mycotoxin balance), the levels of enniatin B (EnnB), enniatin B1 (EnnB1), HT-2, T-2, beauvericin (BEA), and tentoxin (TEN) in batch A were reduced by 70, 75, 48, 58, 35, and 13%, respectively, within the first processing step of cleaning and dehulling. Oat materials after a subsequent operation of steaming and cutting showed reduced levels of EnnB, EnnB1, T-2, BEA, and TEN by 61, 72, 72, 65, and 35%, respectively, whereas HT-2 was detected only in the input material. Mycotoxin levels in the final flake product in batch A were significantly lower than the initial ones. Compared to the original contents in raw oats, the levels of EnnB, EnnB1, HT-2, T-2, BEA, and TEN in the final product were reduced by 80, 87, 100, 80, 65, and 65%, respectively. Similar results were obtained for batch B. The most significant reduction of mycotoxins levels was observed after cleaning and dehulling procedure. Compared to initial raw oat levels, the reduction was by 67, 59, 48, 28, and 40% for EnnB, EnnB1, T-2, BEA, and TEN, respectively. After steaming and cutting, decrease of mycotoxins levels was by 60, 52, 49, 37, and 42%, respectively. Compared to original levels in batch B, mycotoxin levels in the final product were reduced by 68, 62, 64, 32, and 49% for EnnB, EnnB1, T-2, BEA, and TEN, respectively. Results of our study are consistent with conclusions of previous studies (Marchelli and Suman 2010; Kostelanska et al. 2011; De Angelis et al. 2013; Tibola et al. 2015). In the survey of T-2/HT-2 occurrence in European oats and oat products, Pettersson et al. (2011) confirmed significant importance of processing technology on overall mycotoxin content. The mean levels of T-2 and HT-2 in the analysed materials, oat flakes and oatmeals, were

Table 2. Mycotoxin levels in oat materials supplied from a food plant

Batch	Sample	Sample description	Mycotoxin levels (μg kg^{-1})					
			EnnB	EnnB1	HT-2	T-2	BEA	TEN
Batch A	OBM-A1	initial oat	11.5	3.2	19.5	10.3	4.3	2.3
	OBM-A2	oat after cleaning and dehulling	3.3	0.8	10.2	4.3	2.8	2.0
	OBM-A3	oat after steaming and cutting	4.5	0.9	< 5.0	2.9	1.5	1.5
	OBM-A4	sample from middle line	4.2	0.8	< 5.0	2.0	1.5	1.2
	OBM-A5	oat flakes	2.3	0.5	< 5.0	2.1	1.5	0.8
Batch B	OBM-B1	initial oat	8.5	2.9	< 5.0	8.3	8.7	4.5
	OBM-B2	oat after cleaning and dehulling	2.8	1.2	< 5.0	4.3	6.3	2.7
	OBM-B3	oat after steaming and cutting	3.4	1.4	< 5.0	4.2	5.5	2.6
	OBM-B4	sample from middle line	3.6	1.2	< 5.0	3.5	5.3	2.2
	OBM-B5	oat flakes	2.7	1.1	< 5.0	3.0	5.9	2.3

EnnB – enniatin B; EnnB1 – enniatin B1; HT-2 – HT-2 toxin; T-2 – T-2 toxin; BEA – beauvericin; TEN – tentoxin

https://doi.org/10.17221/247/2020-CJFS

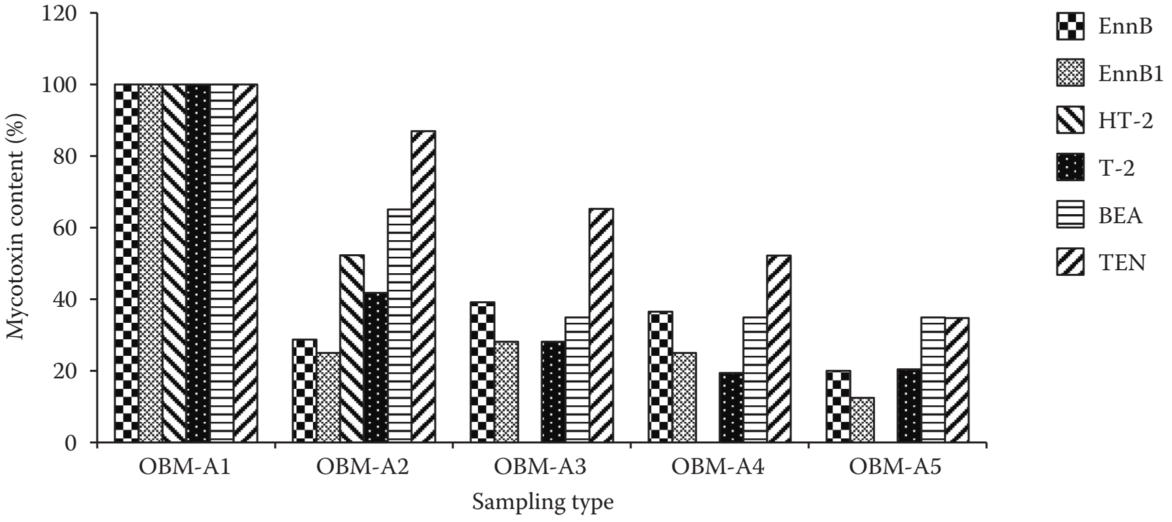

Figure 2. Mycotoxin balance in batch A

OBM-A1 – initial oats; OBM-A2 – oat after cleaning and dehulling; OBM-A3 – oat after steaming and cutting; OBM-A4 – sample in middle line; OBM-A5 – oat flakes; EnnB – enniatin B; EnnB1 – enniatin B1; HT-2 – HT-2 toxin; T-2 – T-2 toxin; BEA – beauvericin; TEN – tentoxin

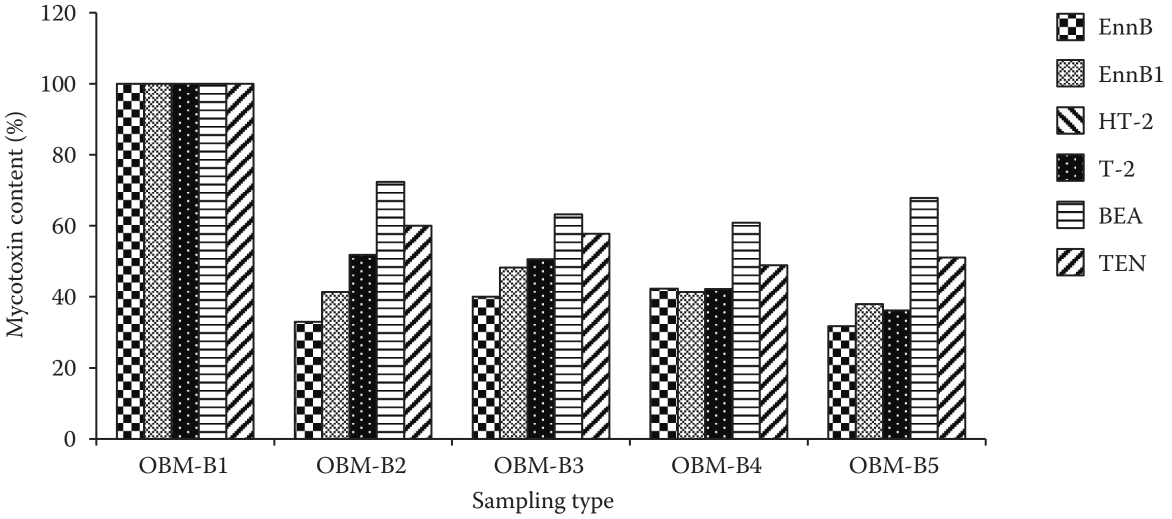

Figure 3. Mycotoxin balance in batch B

OBM-B1 – initial oats; OBM-B2 – oat after cleaning and dehulling; OBM-B3 – oat after steaming and cutting; OBM-B4 – sample from middle line; OBM-B5 – oat flakes; EnnB – enniatin B; EnnB1 – enniatin B1; HT-2 – HT-2 toxin; T-2 – T-2 toxin; BEA – beauvericin; TEN – tentoxin

much lower, by 82% and 88%, respectively, than the levels found in corresponding raw oats. On the other hand, the authors demonstrated high mycotoxin contamination in oat by-products where the mean T-2 and HT-2 levels were 3- and 4-fold higher compared to the levels found in raw oats.

Mycotoxin occurrence in oat products available on Czech retail market. In the second part of the study, twenty samples of different oat flake products commercially available on the Czech retail market were collected whereas the analyses were realised prior to the product expiration date. The mycotoxin levels obtained from analyses of the oat flake products are summarised in Table 3.

A broad spectrum of 16 different mycotoxins has been detected in the analysed oat flake products, more specifically toxic compounds produced by *i*) *Fusarium* fungi – deoxynivalenol (DON), D3G, 3-acetyldeoxynivalenol (3-ADON), HT-2, T-2, zearalenone (ZEA),

Table 3. Mycotoxin levels in oat flake products commercially available on the Czech retail market

Sample	Mycotoxin levels (μg kg^{-1})															
	DON	D3G	3ADON	HT-2	T-2	ZEA	EnnA	EnnA1	EnnB	EnnB1	BEA	AOH	AME	TEN	OTA	MPA
OF-1	< 10	< 10	< 10	10	5	< 1	< 1	1	13	3	5	1	1	2	< 1	< 1
OF-2	< 10	< 10	< 10	9	1	< 1	< 1	< 1	7	2	2	< 1	< 1	< 1	< 1	< 1
OF-3	< 10	< 10	< 10	15	1	< 1	< 1	1	5	3	15	1	< 1	< 1	< 1	11
OF-4	< 10	< 10	< 10	15	1	< 1	< 1	< 1	7	2	2	< 1	< 1	< 1	7	< 1
OF-5	< 10	< 10	< 10	< 10	1	< 1	< 1	2	10	5	3	< 1	< 1	< 1	< 1	< 1
OF-6	13	< 10	< 10	< 10	2	< 1	< 1	< 1	6	2	2	< 1	< 1	< 1	< 1	< 1
OF-7	< 10	< 10	< 10	< 10	< 1	< 1	1	3	111	20	2	< 1	< 1	< 1	< 1	< 1
OF-8	< 10	< 10	< 10	< 10	2	< 1	1	5	74	21	2	< 1	< 1	< 1	< 1	< 1
OF-9	< 10	< 10	< 10	< 10	1	< 1	< 1	1	23	6	5	1	< 1	1	< 1	< 1
OF-10	< 10	< 10	< 10	34	16	< 1	< 1	2	12	6	3	< 1	< 1	< 1	< 1	< 1
OF-11	< 10	< 10	< 10	< 10	< 1	< 1	< 1	1	47	9	1	< 1	< 1	2	< 1	< 1
OF-12	< 10	< 10	< 10	< 10	3	< 1	< 1	1	6	3	6	5	2	< 1	< 1	< 1
OF-13	38	22	< 10	10	3	3	3	13	132	42	4	2	< 1	< 1	< 1	< 1
OF-14	< 10	< 10	< 10	< 10	5	< 1	< 1	2	18	6	1	< 1	< 1	< 1	< 1	< 1
OF-15	< 10	< 10	< 10	12	3	< 1	< 1	1	9	3	3	< 1	< 1	2	< 1	< 1
OF-16	< 10	< 10	< 10	< 10	< 1	< 1	1	5	98	19	3	< 1	< 1	< 1	< 1	< 1
OF-17	< 10	< 10	< 10	< 10	2	< 1	1	7	66	20	4	< 1	< 1	< 1	< 1	18
OF-18	156	53	12	52	15	< 1	5	11	73	29	3	< 1	< 1	< 1	< 1	< 1
OF-19	< 10	< 10	< 10	< 10	< 1	< 1	< 1	< 1	1	1	1	< 1	< 1	< 1	< 1	< 1
OF-20	< 10	< 10	< 10	14	7	< 1	< 1	< 1	1	1	2	< 1	< 1	< 1	4	< 1

DON – deoxynivalenol; D3G – deoxynivalenol-3-glucoside; 3ADON – 3-acetyldeoxynivalenol; HT-2 – HT-2 toxin; T-2 – T-2 toxin; ZEA – zearalenone; EnnA – enniatin A; EnnA1 – enniatin A1; EnnB – enniatin B; EnnB1 – enniatin B1; BEA – beauvericin; AOH – alternariol; AME – alternariol-methylether; TEN – tentoxin; OTA – ochratoxin A; MPA – mycophenolic acid

enniatin A (EnnA), enniatin A1 (EnnA1), EnnB, EnnB1, and BEA; *ii*) fungi of the genus *Alternaria* – alternariol (AOH), alternariol monomethyl ether (AME), and TEN; and *iii*) mycotoxins of the genus *Aspergillus* and/or *Penicillium* – ochratoxin A (OTA) and mycophenolic acid (MPA). Three mycotoxins were detected in all monitored samples, EnnB, EnnB1, and BEA, with average contents of 36, 10, and 4 μg kg^{-1}, respectively. In addition, we also observed the notable incidence of T-2, EnnA1, HT-2, and EnnA, being identified in 80, 75, 45, and 30% of the analysed samples, with average contents of 4, 4, 19, and 2 μg kg^{-1}, respectively. The occurrence of the other monitored mycotoxins was relatively low, fluctuating between 5% and 25% within analysed samples. The findings of our study correspond with conclusions of Pettersson et al. (2011). In their extensive survey, T-2 and HT-2 toxins were simultaneously detected in raw oats, oat flakes, oatmeals, and by-products with the incidence of 93, 77, 34, and 99%, respectively.

With regard to the maximum levels of contaminants in foodstuffs laid down in the relevant European legislation [Commission regulation (EC) No. 1881/2006; Commission recommendation 2013/165/EU], in the group of 16 mycotoxins identified in our study, OTA exceeded the set limit, i.e. 3 μg kg^{-1}. The incidence of OTA among analysed samples was 10% with average content of 6 μg kg^{-1}; neither of the samples positive for OTA met the legislative requirements. Other mycotoxin levels identified in the samples were proved to be below the set limits.

Considering a possible health risk to consumers, we compared our findings with the limits recommended by the European Food Safety Authority (EFSA), referred to as (temporary) tolerable daily intake (TDI) and tolerable weekly intake (TWI). TDI for DON (including D3G and 3-ADON) was set down by EFSA to 1 μg kg^{-1} of body weight (bw), combined temporary TDI for T-2 and HT-2 toxins to 0.02 μg kg^{-1} bw, and temporary TDI for ZEA to 0.25 μg kg^{-1} bw (EFSA 2017a, b, 2020). TWI for OTA was set down to 0.12 μg kg^{-1} bw (EFSA 2020). Considering a common daily portion of 50 g of oat flakes, a theoretical risk to a consumer of 70 kg

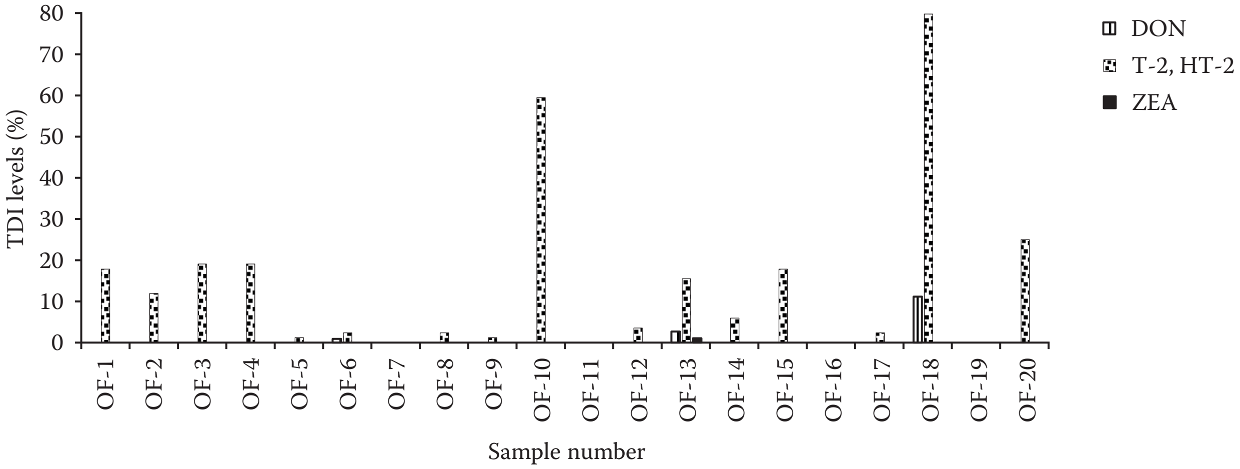

Figure 4. Oat flake contamination detected in samples *vs.* TDI levels reached for DONs (i.e. combined DON/D3G/3--ADON), combined T-2/HT-2 toxins, ZEA and TWI levels reached for OTA (calculated for the intake of 50 g of oat flakes consumed by a man of 70 kg)

TDI – tolerable daily intake; TWI – tolerable weekly intake; DON – deoxynivalenol; D3G – deoxynivalenol-3-glucoside; 3ADON – 3-acetyldeoxynivalenol; ZEA – zearalenone

could arise from the intake of sample OF-18 and OF-10, where such amount corresponds to 240% and 180% of combined TDI for T-2 and HT-2 toxins. The intake of the same amount of samples OF-4 or OF-20 may pose a potential risk to consumers with regard to the presence of OTA. TWI of the toxin reached 29% and 17% for a man of 70 kg in sample OF-4 and OF-20, respectively. Figure 4 illustrates actual TDI/TWI levels for DON, T-2/HT-2 toxins, ZEA, and OTA corresponding to the intake of 50 g of particular oat flake samples involved in our study by a human of 70 kg in weight.

CONCLUSION

Results of the present study showed that particular oat flake fractions obtained from a real production line contained generally low overall mycotoxin contamination. At the same time, we demonstrated a significant influence of the processing steps. Especially oat cleaning and dehulling induced a notable decrease of mycotoxin levels. The subsequent processing steps showed a negligible impact on the final product contamination, whereas the mycotoxin levels remained unchanged.

Oat flakes commercially available on the Czech retail market were proved to be contaminated by mycotoxins of *Fusarium*, *Alternaria*, and *Aspergillus*, and/or *Penicillium* origin. We detected 16 different toxins in 20 analysed oat flake products, whereas the most frequent were EnnB, EnnB1, and BEA. Furthermore, T-2, EnnA1, HT-2, and EnnA occurred also in many analysed samples. Two samples exceeded the current maximum limits for OTA and TDI would be exceeded for an average consumer and sum of HT-2 and T-2. Nevertheless, with regard to the risk to consumers, considering the set TDI/TWI as well as ordinary oat flakes intake, we concluded that the consumption of oat flake products does not pose a serious health risk to consumers.

REFERENCES

Alshannaq A., Yu J.H. (2017): Occurrence, toxicity and analysis of major mycotoxins in food. International Journal of Environmental Research and Public Health, 14: 632.

De Angelis E., Monaci L., Pascale M., Visconti A. (2013): Fate of deoxynivalenol, T-2 and HT-2 toxins and their glucoside conjugates from flour to bread: An investigation by high-performance liquid chromatography high-resolution mass spectrometry. Food Additives and Contaminants: Part A, 30: 345–355.

Džuman Z., Zachariášová M., Lacina O., Vepříková Z., Slavíková P., Hajšlová J. (2014): A rugged high-throughput analytical approach for the determination and quantification of multiple mycotoxins in complex feed matrices. Talanta, 121: 263–272.

Edwards S.G. (2004): Influence of agricultural practices on *Fusarium* infection of cereals and subsequent contamination of grain by trichothecene mycotoxins. Toxicology Letters, 153: 29–35.

EFSA (2017a): Human and animal dietary exposure to T-2 and HT-2 toxin. European Food Safety Authority. EFSA Journal, 15: 4972.

EFSA (2017b): Risks to human and animal health related to the presence of deoxynivalenol and its acetylated and modified forms in food and feed. European Food Safety Authority. EFSA Journal, 15: 4718.

EFSA (2020): Risk assessment of ochratoxin A in food. European Food Safety Authority. EFSA Journal, 18: 6113.

Grundy M.M.L., Fardet A., Tosh S.M., Rich G.T., Wilde P.J. (2018): Processing of oat: The impact on oat's cholesterol lowering effect. Food & Function, 9: 1328–1343.

Kiecana I., Mielniczuk E., Perkowski J., Golinski P. (2005): Infection of panicles with *Fusarium poae* (Peck) Wollenw. and mycotoxin content in oat grain. Acta Agrobotanica, 58: 91–102.

Kostelanska M., Dzuman Z., Malachova A., Capouchova I., Prokinova E., Skerikova A., Hajslova J. (2011): Effects of milling and baking technologies on levels of deoxynivalenol and its masked form deoxynivalenol-3-glucoside. Journal of Agricultural and Food Chemistry, 59: 9303–9312.

Liu R.H. (2007): Whole grain phytochemicals and health. Journal of Cereal Science, 46: 207–219.

Marchelli R., Suman M. (2010): Fate of *Fusarium* mycotoxins in the cereal product supply chain: The deoxynivalenol (DON) case within industrial bread-making technology. Food Additives and Contaminants: Part A, 27: 677–687.

Menon R., Gonzalez T., Ferruzzi M., Jackson E., Winderl D., Watson J. (2016): Oats – From farm to fork. In: Henry J. (ed.): Advances in Food and Nutrition Research. USA, Academic Press: 1–56.

Palumbo R., Crisci A., Venancio A., Abrahantes J.C., Dorne J.L., Battilani P., Toscano P. (2020): Occurrence and co-occurrence of mycotoxins in cereal-based feed and food. Microorganisms, 8: 74.

Peterson D.M., Hahn M.J., Emmons C.L. (2002): Oat avenanthramides exhibit antioxidant activities *in vitro*. Food Chemistry, 79: 473–478.

Pettersson H., Brown C., Hauk J., Hoth S., Meyer J., Wessels D. (2011): Survey of T-2 and HT-2 toxins by LC-MS/MS in oats and oat products from European oat mills in 2005–2009. Food Additives and Contaminants: Part B, 4: 110–115.

Strychar R., Webster F.H., Wood P.J. (2011): World oat production, trade, and usage. In: Webster F.H., Wood P.J. (eds.): Oats: Chemistry and technology. 2nd Ed. Minnesota, USA, American Association of Cereal Chemists, Inc.: 1–10.

Tibola C.S., Fernandes J.M.C., Guarienti E.M., Nicolau M. (2015): Distribution of *Fusarium* mycotoxins in wheat milling process. Food Control, 53: 91–95.

Webster F.H., Wood P.J. (2011): Oats: Chemistry and technology. 2nd Ed. Saint Paul, Minnesota, USA, American Association of Cereal Chemists, Inc.: 1–376.

Wood P.J. (2011): Oat beta-glucan: Properties and function. In: Webster F.H., Wood P.J. (eds.): Oats: Chemistry and Technology. 2nd Ed. St. Paul, Minnesota, USA, American Association of Cereal Chemistry, Inc.: 219–254.

Zohary D., Hopf M., Weiss E. (1988): Domestication of Plants in the Old World: The Origin and Spread of Cultivated Plants in West Asia, Europe, and the Nile Valley. 4th Ed. United Kingdom, Oxford University Press: 1–249.

Received: October 8, 2020
Accepted: March 15, 2021