JOURNAL OF AGRICULTURAL AND FOOD CHEMISTRY

ARTICLE

pubs.acs.org/JAFC

Impact of Physicochemical Parameters on the Decomposition of Deoxynivalenol during Extrusion Cooking of Wheat Grits

Qinghua Wu,[†,‡] Lilia Lohrey,[‡] Benedikt Cramer,[‡] Zonghui Yuan,[†] and Hans-Ulrich Humpf[*,‡]

[†]MOA Key Laboratory of Food Safety Evaluation/National Reference Laboratory of Veterinary Drug Residues (HZAU), Huazhong Agricultural University, Wuhan, Hubei 430070, People's Republic of China

[‡]Institute of Food Chemistry, University of Münster, Corrensstrasse 45, 48149 Münster, Germany

ABSTRACT: Deoxynivalenol (DON) is a toxic secondary metabolite produced by molds of the *Fusarium* genus and is known to cause a spectrum of diseases in animals such as vomiting and gastroenteritis. It is found in cereals and cereal products as most processing techniques lead only to a partial reduction of deoxynivalenol levels. One technique with a reported relatively high impact on deoxynivaleol decomposition is extrusion cooking. In the current work, systematic studies of a range of physicochemical parameters, such as temperature, moisture, compression, residence time in the extruder, pH value, and protein content, on their impact on deoxynivalenol decomposition during extrusion cooking were performed. The analysis of deoxynivalenol was made by high-performance liquid chromatography−tandem mass spectrometry using a quick, easy, cheap, effective, rugged, and safe-based cleanup with 15-d_1-deoxynivalenol as an internal standard. It could be shown that the reduction of deoxynivalenol levels is dependent on a set of parameters partially interacting with each other. Especially the moisture content and compression are key factors for the reduction of deoxynivalenol levels. A correlation between residence time of the mycotoxin in the extruder and deoxynivalenol degradation was also observed when screws without a compression factor were used. Generally, the reduction of deoxynivalenol levels was increased by the use of screws with a high compression factor. As known from cooking, deoxynivalenol could also be easily degraded by extrusion under alkaline conditions. Furthermore, an increase of the protein content of the starting material resulted in higher reduction rates of deoxynivalenol.

KEYWORDS: Deoxynivalenol, DON, extrusion, reduction, degradation, pH, protein, starch, moisture, compression, QuEChERS

INTRODUCTION

Deoxynivalenol (DON; vomitoxin; 3α,7α,15-trihydroxy-12,-13-epoxytrichothec-9-en-8-one) belongs to the group B trichothecenes and is considered to be one of the most important mycotoxins in cereal commodities due to the high occurrence rate (Figure 1).[1] DON can be produced by fungi such as *Fusarium graminearum*, *Fusarium culmorum*, and *Fusarium crookwellense*, which are common pathogens on cereal crops like wheat, barley, and maize in the field.[2] DON has been shown to cause a variety of toxic effects in vivo. Feed refusal, vomiting, and digestive disorders with subsequent losses of weight are the common toxic effects for animals.[3] Furthermore, the immunoglobulin A (IgA) nephropathy was observed in mice that ingested 25 ppm of DON for 24 weeks.[4] Among animals, pigs are the mostly susceptible to DON.[5] For humans, dizziness, throat irritation, nausea, vomiting, diarrhea, and blood in the stool are described as symptoms of DON intoxication.[2]

Extrusion cooking is a process of central importance as its application is widespread in the food industry. Several cereal products such as breakfast cereals, snacks, and animal feedstuffs are produced by extrusion. The extrusion process is carried out with either single or twin screw configuration and utilizes a combination of high temperature, high pressure, and severe shear forces. As a result, complex chemical changes and modifications including starch retrogradation polymer cross-linking and Maillard reactions can be observed for food components as well as contaminants.[6] Extrusion cooking can not only improve the quality by modifying the texture or increasing the digestibility of the processed products; it also has an effect on the levels of mycotoxins found in the final product. For instance, approximately 90−99% of fumonisin B_1 was lost by extruding alkali-cooked flour at 171 °C.[7]

The extrusion of DON in cereals has already been studied by different groups. Wolf-Hall et al.[8] investigated the extrusion of DON-contaminated corn grits and dry dog food at 100 °C. The levels of DON were reduced after processing in the extruded corn grits and dry dog food by 53 and 21%, respectively. Accerbi et al.[9] studied the effects of sodium bisulfite on DON levels in wheat grain and mill fractions during extrusion at 130−170 °C. The soaking treatment with sodium bisulfite solution lowered DON from 3.7 to 0.8 μg/g without extrusion and to 0.3 μg/g with an additional extrusion process. In another study by Cazzaniga et al., 95% of DON was lost in corn flour after the extrusion at 150 °C as well as 180 °C.[10] More recently, Scudamore et al.[11,12] reported that DON was quite stable during extrusion and that the moisture content has a stronger effect than temperature at 140−180 °C.

However, to our knowledge, until now there are no comprehensive studies on the degradation of DON during the extrusion of wheat grits, dealing with a larger range of physicochemical parameters that can be varied to improve product quality. Furthermore, no reports are available on combinatorial effects such as moisture and compression or residence time in the

Received: September 22, 2011
Accepted: October 19, 2011
Revised: October 17, 2011
Published: October 19, 2011

dx.doi.org/10.1021/jf2038604 | *J. Agric. Food Chem.* 2011, 59, 12480–12485

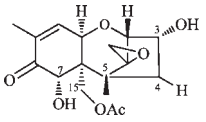

Figure 1. Chemical structure of DON.

extruder and temperature. Only Cazzaniga et al.[10] tried to evaluate the effects of combined variations in extrusion parameters in a larger scale. The group systematically changed moisture, temperature, screw speed, and compression while studying the degradation of DON in corn. However, under all conditions described, the group observed a complete degradation, which does not allow a comparison of different extrusion parameters and is contrary to observations by other groups.[8,11,12]

Therefore, the aim of this work is to study systematically the role of the parameters temperature, compression, residence time, and moisture on DON degradation during extrusion cooking. Additionally, the effects of the protein content and pH value of the starting material on the degradation of DON were studied. The analysis of DON in the extruded samples was performed by high-performance liquid chromatography−tandem mass spectrometry (HPLC-MS/MS) using a modified quick, easy, cheap, effective, rugged, and safe (QuEChERS) cleanup with 15-d_1-DON as and internal standard (IS).[13,14]

MATERIALS AND METHODS

Sampling. Wheat grits (particle size, 0.125−0.25 mm) were bought from a supermarket (Münster, Germany). Corn grits were kindly provided by the Kampffmeyer mill (Hameln, Germany). The moisture content was determined by weight loss after oven drying at 150 °C for 16 h and was measured to be approximately 14%. DON was prepared from *F. graminearum* according to previously described procedures,[15] and the DON standard solution was prepared by dissolving 20.0 mg of DON (purity >98%) in 20 mL of acetonitrile using a 20 mL volumetric flask. The wheat grits were spiked with DON by adding 400 mL of water containing 1 mL of DON standard solution (1000 μg/mL, in acetonitrile) to 2 kg of blank grits. The mixture was blended for 45 min using a Bosch Profi 67 mixer (Bosch, Stuttgart, Germany). The final concentration of DON and the moisture content were 500 μg/kg and 34% [dry weight basis (dw)], respectively. The samples were allowed to equilibrate overnight in polyethylene bags. To check whether the homogeneity is sufficient, 10 g of grits from four different parts of the bag was taken randomly from each batch and analyzed by HPLC-MS/MS for the DON content.

DON-spiked grits (500 μg/kg) were mixed with $NaHCO_3$ to adjust the concentration to be 0.1, 0.5, and 2.0% (w/w) (Merck, Darmstadt, Germany) and afforded pH values 7.3, 7.8, and 8.0, respectively. The pH value of the nontreated grits was measured to be 6.0. The pH was determined in the suspension of 5 g of samples in 25 mL of water.

The natural protein content in wheat grits is reported to be about 10.6%.[16] Ten and 20 g of gluten from wheat (Sigma-Aldrich, Taufkirchen, Germany) with approximately 80% of protein was added to 90 and 80 g of DON-spiked wheat grits (500 μg/kg), respectively, to adjust the gluten concentration in wheat to be 10 and 20% (w/w). In addition, 10 g of gluten was mixed with 90 g of starch (from corn) to prepare a 10% gluten starch sample. Pure starch (100 g) was used as the 0% protein sample. The DON standard solution (50 μL, 1000 μg/mL, in acetonitrile) mixed with 34 mL of water was spiked into a 100 g starch sample to obtain a DON content of 500 μg/kg. The mixture was blended for 45 min using a Bosch Profi 67 mixer (Bosch). Thus, different protein levels in samples were reached.

All experiments were conducted with samples having 34% moisture content and 500 μg/kg of DON unless otherwise mentioned. DON-spiked grits (400 g) were oven-dried at 50 °C for 24 h, and then, 68 mL of distilled water was added and mixed for 45 min to adjust the moisture content to 17% (dw) as the samples with lower moisture content.

Extraction Method. The extraction of extruded samples was performed by a modified QuEChERS method[17] previously described by Trebstein et al.[13] Briefly, 5 g of extruded samples was weighted into a 50 mL polypropylene tube, and 20 mL of water was added. After the tube was shaken for 5 s, 20 mL of acetonitrile was added, and the suspension was vigorously shaken for 1 min. Afterward, 8 g of $MgSO_4$ (AppliChem, Darmstadt, Germany) and 2 g of NaCl were added, and the suspension was again shaken for 1 min. The samples were centrifuged at 3500*g* for 5 min, and 5 mL of the supernatant was transferred into a 15 mL single use centrifugation tube, which contained 125 mg of primary secondary amine (Agilent, Böblingen, Germany) and 750 mg of $MgSO_4$. The samples were shaken for 1 min and centrifuged at 3500*g* for 5 min. One milliliter of the sample extract was evaporated to dryness under a nitrogen stream (45 °C), dissolved with 500 μL of methanol/water solvent (1/9, v/v), and further analyzed by HPLC-MS/MS (MRM mode). For quantification, 15-d_1-DON was synthesized in our group[14] but is also commercially available (Sigma-Aldrich) and was used as an IS (100 ng/mL).

Calibration Curves and Determination of the Limit of Detection (LOD), Limit of Quantitation (LOQ), and Recovery Rate. The stock solution of DON (1 mg/mL, in acetonitrile) was diluted to a concentration of 1 μg/mL with acetonitrile (standard solution). Aliquots of the standard solutions were mixed in various concentration ratios (DON:15-d_1-DON 1:4 up to 5:1) with 15-d_1-DON (100 ng/mL, in methanol/water, 1/9, v/v) to afford DON concentrations ranging from 25 to 500 ng/mL. The mixtures were analyzed in the MRM mode with HPLC-MS/MS, and each concentration was injected twice. The resulting peak area ratios were plotted against the concentration ratios. The LOD was determined at a signal-to-noise ratio (S/N) higher than 3:1 and the LOQ > 10:1.

For the determination of the recovery rate, blank grits were extruded at 150 °C by screw 1/1 and ground. The extruded grits (5 g) were spiked with 250 (1 μg/mL), 125 (10 μg/mL), and 250 μL of DON (10 μg/mL) standard solution to obtain concentrations of 50, 250, and 500 μg/kg, respectively. Duplicate samples were cleaned up according to the procedure described above and analyzed by HPLC-MS/MS.

Extrusion Processing. A model KE19/25D 150 N m/150 rpm laboratory scale conical single-screw extruder (Brabender, Duisburg, Germany) was used with compression ratios screw 3/1 and 1/1 and a 5 mm diameter cylindrical die. The screw speed and feed speed was 35 and 10 rpm/min, respectively. The residence time of the wheat grits with 34% moisture in the heated zone at this speed is 2.0 min. Under these conditions at 150 °C, the feed rate is 21.5 g/min. The wheat grits (34% moisture) were extruded at 100, 110, 120, 130, 140, 150, 160, 170, and 180 °C by screw 3/1 and 1/1, respectively. Low moisture wheat grits (17%) were extruded at 120, 150, and 170 °C by screw 3/1 and 1/1, respectively. To make a longer residence time, the screw and feed speed was set up to be 20 and 5 rpm/min, respectively; thus, the residence time of wheat grits was extended to 4 min. In the low speed extrusion processing, wheat grits (17 and 34% moisture) were extruded at 120, 150, and 170 °C by screw 3/1 and 1/1, respectively. In addition, samples with different pH values and different protein contents were extruded at 150 °C using screw 1/1 with the screw/feed speed 35/10.

The extruder was neither stopped nor cleaned between samples; hence, a portion of clean corn grit samples were used to purge the extruder. For each extruded sample, 100 g was collected, ground and stored at −20 °C until analyzed.

Two samples were taken at each experimental set of conditions and analyzed independently. Data shown in this study are given as means for

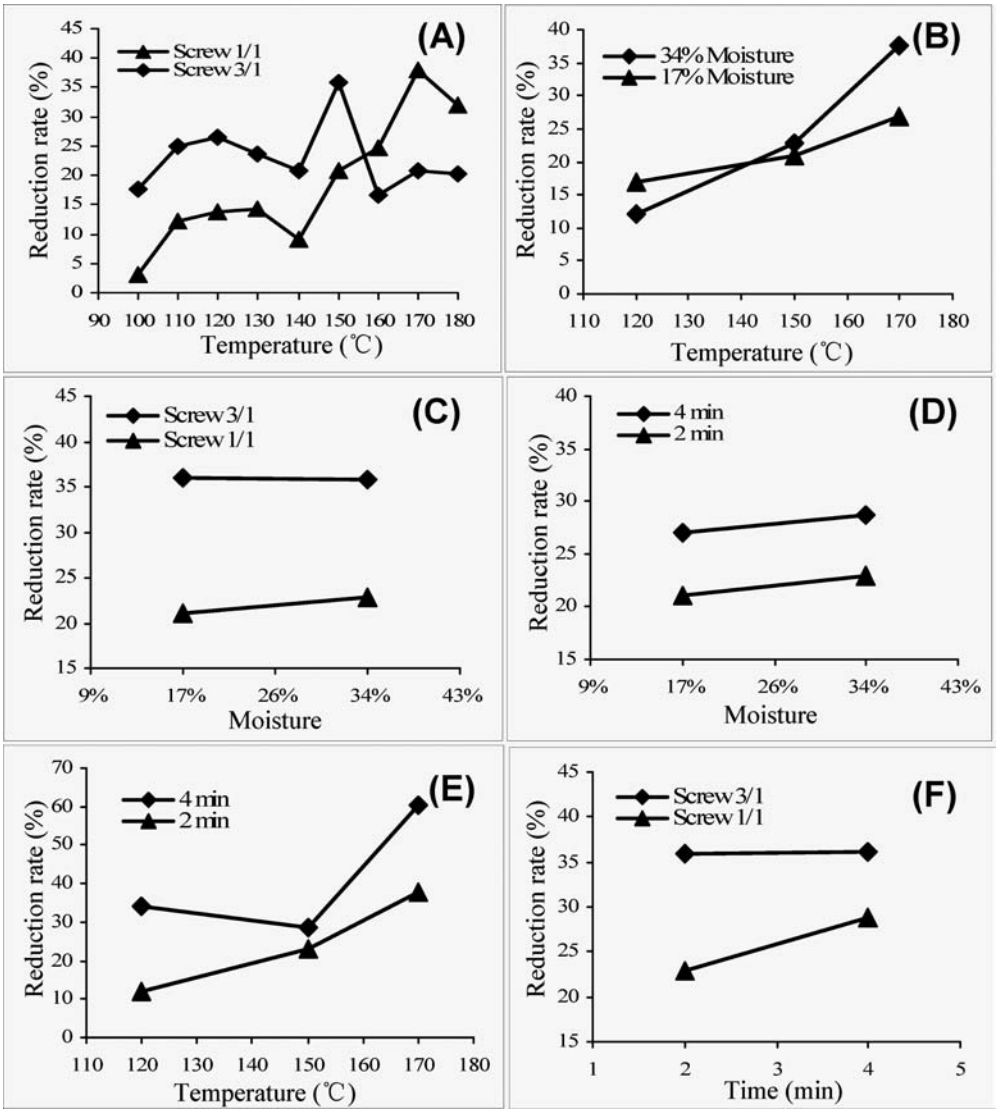

Figure 2. Reduction of DON in wheat grits by the combinatorial effects of temperature, moisture, compression, and residence time. Panels C, D, and F were extruded at 150 °C. Each bar represents the mean of DON reduction values of extruded samples (n = 2).

this duplicate analysis. The reduction rate (%) is (DON concentration before extrusion − DON concentration after extrusion)/DON concentration before extrusion × 100%.

Diameter Detection. The radial expansion ratios of extrusion-cooked samples were calculated by dividing the cross-sectional area of the extrudates by the cross-sectional area of the die orifice. The radial expansion ratio will be referred in the text simply as the expansion ratio. A vernier caliper was used to measure the diameter of the extrudates. The average of 10 measurements of extrudate diameter was used to calculate radial expansion.

HPLC-MS/MS Analysis. An Agilent 1100 series HPLC (Agilent Technologies) linked to an API 4000 QTRAP mass spectrometer (ABSciex, Darmstadt, Germany) was used for HPLC-ESI-MS/MS analysis. Data acquisition was performed with the Analysis 1.4.2 software (ABSciex). The column used for chromatographic separation was a 150 mm × 2.0 mm i.d., 5 μm, Gemini C-18 (Phenomenex, Aschaffenburg, Germany) at 40 °C. The mobile phase was methanol (A) and water (B). The injection volume was 10 μL, and the flow rate was 250 μL/min. The gradient was as follows: 0−1 min, 10% A; 1−10 min, 10−100% A. For HPLC-MS/MS, the mass spectrometer was operated in the multiple reaction monitoring mode (MRM), detecting negative ions $[M - H]^-$. Zero-grade air served as nebulizer gas (35 psi) and, heated at 300 °C, as the turbo gas for solvent drying (50 psi). The following transition reactions were monitored for durations of 150 ms each. Declustering potential (DP), collision energy (CE), and collision cell exit potential (CXP) are given in brackets: DON: m/z 295. MS/MS: 265 (DP, −80 V; CE, −16 V; and CXP, −7 V). 15-d_1-DON: m/z 296. MS/MS: 265 (DP, −80 V; CE, −16 V; and CXP, −7 V).

RESULTS AND DISCUSSION

The analysis of all extruded wheat samples was carried out by HPLC-MS/MS with 15-d_1-DON added as the isotope-labeled IS.[11] The addition was necessary as it is known that the food extracts following the QuEChRS procedure contain large amounts of matrix and thus can lead to matrix-dependent signal suppression or enhancement (matrix effects). The recovery rates were 107.9 ± 6.66, 97.1 ± 1.40, and 92.4 ± 2.05% for 50, 250, and 500 μg/kg of DON, respectively. The calibration curve in the range of 50−1000 ng/g DON showed a good

Figure 3. Appearance of the extruded products from wheat grits by screw 1/1 at 110−180 °C.

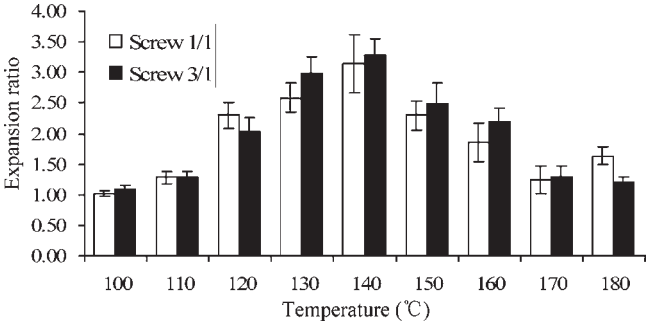

Figure 4. Expansion ratio of the wheat extrudates at 100−180 °C.

correlation (r = 0.9976). The LOD and LOQ were 5.8 and 19.4 ng/g, respectively.

The temperature dependency of the degradation of DON was studied by almost all groups investigating the impact of extrusion cooking on this mycotoxin. However, the different groups yielded a broad spectrum of results ranging between no changes in DON concentration to complete loss.[10,11] Thus, to compare our extruder setup and analytical system with those published, we recorded the temperature-dependent DON degradation using a 1/1 screw. All other factors like moisture, screw speed, and dye size were kept constant. As shown in Figure 2A, an increase of the DON degradation rate from 3.1 ± 0.30% at 100 °C up to 37.9 ± 2.15% at 170 °C was observed; at 180 °C, the degradation rate slightly decreased to 31.9 ± 3.80%. The degradation rates observed in these experiments are essentially lower than those described by Cazzaniga et al.,[10] who observed nearly complete degradation but higher than the results reported by Scudamore et al.[11] Explanations for the observed differences compared to Scudamore et al.[11] are the temperature gradient and moisture content applied by that group. Their experimental setup started with a moisture content of 17% and run a temperature gradient along the different extruder segments with the final temperatures of 140−180 °C being reached only in the last zone of the system. In the setup used in our experiments, all temperature zones were set to the respective temperature given in Figure 2, resulting in a longer exposure of the DON-containing material to the maximum heat and pressure. Additionally, in these experiments, the moisture content was adjusted to 34%. Thus, the higher degradation rates in this basic experiment, especially at high temperatures, can be explained by generally harsher extrusion parameters applied in our system. To allow a comparison of the extrusion products obtained by our setup and those from other groups, pictures of the extrudates are shown in Figure 3, and the temperature-dependent radial expansions are given in Figure 4. The radial expansion of the extrudates can reflect the overall expansion[18] and allows us to monitor the progress of extrusion cooking. Figure 4 shows that the expansion rate initially increases with the rising barrel temperature from 100 to 140 °C but declines gradually with further rising temperatures. The expansion ratio of cereals depends mainly on its degree of gelatinization, which is determined by temperature, shear rate, and moisture content of the feed material.[19−21] By elevating the temperature, the degree of gelatinization increases until at a specific temperature (140 °C in this study), the gelatinization reaches a maximum. Above this temperature, expansion reduces again, and smaller diameters of the extrudates are measured. In addition, the shear rate is inversely proportional to the expansion ratios, and the shear rate is initially declined and then increases with the rising temperature during the extrusion.[20,22,23]

After this basic set of extrusion experiment, the screw type (1/1 and 3/1) was changed, followed by the parameters moisture (17 and 34%), temperature (120, 150, and 170 °C), and residence time (2 and 4 min), which were studied in combination (see Figure 2B−F).

The installation of a 3/1 screw revealed a different profile as compared to the 1/1 screw with an almost constant degradation of DON ranging between 15 and 25% (Figure 2A). Only at 150 °C, an exceptional reduction of DON levels by 36% was detected. Generally, this curve shows that with higher compression, temperature plays a minor role regarding the reduction of DON levels.

The effect of moisture content on DON degradation was studied with two different moisture levels of 17 and 34% and in combination with three different temperatures (120, 150, and 170 °C), two screw types (1/1 and 3/1), and two residence times (2 and 4 min) (Figure 2B−D). As shown in Figure 2B, moisture and temperature strongly interact on the DON degradation. At the low temperature of 120 °C, less moisture supports DON degradation, while at 150 °C, the reduction of DON levels is slightly stronger with a high moisture content. At a temperature of 170 °C, the impact of high moisture on the DON degradation is strongly increased with 37.7 ± 0.62% loss of DON at 34% moisture as compared to only 27.0 ± 0.01% reduction at 17% moisture.

The observations for low temperature are in good agreement with the results published by Scudamore et al.[11] who reported that loss of DON during extrusion in 15% moisture wheat (18.9% loss of DON) is much higher than that in 21% moisture. However, at higher temperatures, we see a different tendency with high moisture content strongly supporting DON degradation. A possible explanation for this difference could again be the different extruder setup and temperature profile used by Scudamore et al.[11] described above. This group used a temperature gradient for extrusion cooking with no heating, 50 and 70 °C for the first, second, and third segment, respectively. Thus, as compared to our setup with constant heating of all extruder segments to the desired temperature, less thermal energy is brought into the system.

The reduction of DON by screw 3/1 at 150 °C was higher than that by screw 1/1 at both moisture levels (Figure 2C). Like Figure 2A, this clearly shows that compression supports the reduction of DON at a temperature of 150 °C. For both screw types, no combinatorial effect with moisture could be observed, as the degradation rates are nearly constant for both moisture levels.

When the residence time was extended from 2 to 4 min, the degradation of DON was strongly increased from 21.1 ± 0.21 to 26.9 ± 2.54% and from 22.9 ± 0.04 to 28.8 ± 2.05% for low and high moisture grits, respectively (Figure 2D). The increase of DON degradation with residence time is similar for both moisture levels; thus, there is no combinatorial effect between these parameters.

The amount of DON degradation due to increased residence time is temperature dependent as shown in Figure 2E. At 120 °C, the reduction rate for DON changed from 12.1 ± 0.38 to 34.1 ± 2.62%. At 170 °C, the degradation was increased from 37.7 ± 0.62 up to 60.4 ± 0.61% with increasing residence time, while at 150 °C, only a moderate change from 22.9 ± 0.04 to 28.8 ± 2.05% could be observed. The impact of the residence time is also dependent on the screw type applied in the experiments (Figure 2F). While, as described above, for a 1/1 screw, a change from 22.9 ± 0.04 to 28.8 ± 2.05% was measured, nearly no influence of the residence time on a 3/1 screw was detected with a constant reduction rate of DON at approximately 36% (Figure 2F). In summary, this suggests that the increase of the residence time can be a critical factor for efficient DON degradation.

The results shown in Figure 2 generally demonstrate that there is a complex interaction of different parameters on the reduction of DON levels during extrusion. Thus, before simply elevating the extrusion temperature when trying to lower DON levels, the combinatorial effects shown here should be considered. As an example, extension of the residence time while lowering temperature might be an approach to reduce the DON levels without major changes in the general product quality.

To examine the effect of pH on the reduction of DON levels, $NaHCO_3$ was added to DON-spiked wheat grits (pH 6.0, 7.3, 7.8, and 8.0) prior to extrusion at 150 °C using a 1/1 screw and 34% moisture. By increasing the pH value, we observed a greater reduction of DON levels. At pH 6, the reduction rate of DON was 20.7 ± 4.03%, which increased up to 79.2 ± 1.17% at pH 8.0. At pH 7.3 and 7.8, moderate reduction rates of 40.6 ± 3.98 and 45.1 ± 1.62% were observed. This demonstrates that DON is easily degraded under alkaline conditions. Our results are in good agreement with the data provided by Wolf and Bullerman.[24] They reported that DON is totally destroyed after 15 min when heated at 170 °C in an aqueous environment at pH 10. Another study showed that DON is reduced by 72−82% during the traditional process of preparing tortillas (110−120 °C) from corn by adding calcium hydroxide.[25] However, without heating, DON remains also nearly stable under alkaline conditions. Lauren and Smith[26] studied the stability of DON in aqueous $NaHCO_3$ solutions at pH 1−12. DON was found to be susceptible to alkaline conditions, but these conditions need to be quite harsh (prolonged exposure and heat). However, as shown above, a moderate increase of the pH to 7.3 could already double the degradation of DON from 20.7 ± 4.03 to 40.6 ± 3.98%, making a moderate pH adjustment a potential tool for improving the quality of highly contaminated wheat.

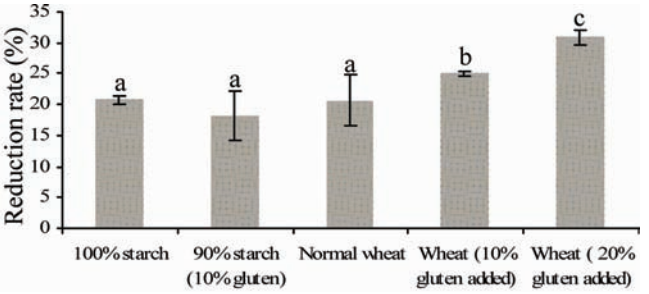

Figure 5. Reduction of DON in wheat grits and starch with a different content of gluten at 150 °C by single screw 1/1 extrusion. Each bar represents the mean of DON reduction values of extruded samples ($n = 2$), and bars with different letters are significantly different ($p \leq 0.05$).

The impact of the protein content on the reduction of DON levels by extrusion at 150 °C using a 1/1 screw and 34% moisture is shown in Figure 5. Grits with higher protein content showed greater reduction of DON. The highest loss of DON was found in grits with 20% gluten added (30.9 ± 1.30%). To study the reduction of DON with lower protein content, 100% of starch (as the 0% protein sample) as well as 90% starch mixed with 10% gluten with the same concentration of DON were investigated. The reductions rates of DON were lower than those obtained with higher protein content. Therefore, we conclude that some degradation of DON could be based on reactions with the amino acid side chains of proteins.

In summary, to find the optimal condition for DON reduction during extrusion, the effects of a range of physicochemical parameters such as temperature, moisture, compression, residence time, pH value, and protein content on the reduction of DON levels in wheat were studied. Because of the observed interactions of different parameters, the degradation curves shown in Figure 2 are intended to help optimize extrusion processes to lower the DON levels. However, as the fate of DON during extrusion cooking is not clear, the use of Good Agricultural and Manufacturing Practices (GAP/GMP) on the field to reduce DON levels is strongly recommended as the first approach to control this contaminant in cereal products.

SAFETY

Wheat grits spiked with DON standard solutions should be handled with proper caution, and contact with skin should be avoided.

AUTHOR INFORMATION

Corresponding Author

*Tel: +49 251 83 333 91. Fax: +49 251 83 333 96. E-mail: humpf@uni-muenster.de.

Funding Sources

This study was supported by a fellowship to Q.W. by China Scholarship Council. We also thank the state of North-Rhine Westphalia and the European Union in the Ziel 2-Programm NRW for funding.

REFERENCES

(1) Moss, M. O. Mycotoxin review-2. *Fusarium*. *Mycologist* **2002**, *16*, 158–161.

(2) SCF (Scientific Committee on Food). Opinion of the scientific committee on *Fusarium* toxins. Part 1: Deoxynivalenol (DON), 1999; http://ec.europa.eu/food/fs/sc/scf/out44_en.pdf (accessed 17/08/2011).

(3) Eriksen, G. S.; Pettersson, H. Toxicological evaluation of trichothecenes in animal feed. *Anim. Feed Sci. Technol.* **2004**, *114*, 205–239.

(4) Dong, W.; Pestk, A. J. Persistent dysregulation of IgA production and IgA nephropathy in the B6C3F1 mouse following withdrawal of dietary vomitoxin (deoxynivalenol). *Fundam. Appl. Toxicol.* **1993**, *20*, 38–47.

(5) Conkova, E.; Laciakova, A.; Kovac, G.; Seidel, H. Fusarial toxins and their role in animal diseases. *Vet. J.* **2003**, *165*, 214–220.

(6) Bullerman, L. B.; Bianchini, A.; Hanna, M. A.; Jackson, L. S.; Jablonski, J.; Ryu, D. Reduction of fumonisin B in corn grits by single-screw extrusion. *J. Agric. Food Chem.* **2008**, *56*, 2400–2405.

(7) Cortez-Rocha, M. O.; Trigo-Stockli, D. M.; Wetzel, D. L.; Reed, C. R. Effect of extrusion processing on fumonisin B_1 and hydrolyzed Fumonisin B_1 in contaminated alkali-cooked corn. *Bull. Environ. Contam. Toxicol.* **2002**, *69*, 471–478.

(8) Wolf-Hall, C.; Hanna, M.; Bullerman, L. B. Stability of deoxynivalenol in heat-treated foods. *J. Food Protect.* **1999**, *62*, 962–964.

(9) Accerbi, M.; Rinaldi, V. E. A.; Ng, P. K. W. Utilization of highly deoxynivalenol-contaminated wheat via extrusion processing. *J. Food Protect.* **1999**, *62*, 1485–1487.

(10) Cazzaniga, D.; Basilico, J. C.; Gonzalez, R. J.; Torres, R. L.; de Greef, D. M. Mycotoxins inactivation by extrusion cooking of corn flour. *Lett. Appl. Microbiol.* **2001**, *33*, 144–147.

(11) Scudamore, K.; Guy, R. C. E.; Kelleher, B.; MacDonald, S. J. Fate of the *Fusarium* mycotoxins, deoxynivalenol, nivalenol and zearalenone, during extrusion of wholemeal wheat grain. *Food Addit. Contam. A* **2008**, *25*, 331–337.

(12) Scudamore, K.; Guy, R. C. E.; Kelleher, B.; MacDonald, S. J. Fate of *Fusarium* mycotoxins in maize flour and grits during extrusion cooking. *Food Addit. Contam. A* **2008**, *25*, 1374–1384.

(13) Trebstein, A.; Lauber, U.; Humpf, H. U. Analysis of *Fusarium* toxins via HPLC-MS/MS multimethods: Matrix effects and strategies for compensation. *Mycotoxin Res.* **2009**, *25*, 201–213.

(14) Bretz, M.; Beyer, M.; Cramer, B.; Humpf, H. U. Stable isotope dilution analysis of the *Fusarium* mycotoxins deoxynivalenol and 3-acetyldeoxynivalenol. *Mol. Nutr. Food Res.* **2006**, *50*, 251–260.

(15) Altpeter, F.; Posselt, U. K. Production of high quantities of 3-acetyldeoxynivale-nol and deoxynivalenol. *Appl. Microbiol. Biotechnol.* **1994**, *41*, 384–387.

(16) Souci, S. W.; Fachmann, W.; Kraut, H. Cereals. In *Food Composition and Nutrition Tables*, 7th ed.; Souci, S. W., Fachmann, W., Kraut, H., Eds.; MedPharm Scientific Publishers: Stuttgart, Germany, 2008; pp 619−741.

(17) Anastassiades, M.; Lehotay, S. J.; Stajnbaher, D.; Schenck, F. J. Fast and easy multiresidue method employing acetonitrile extraction/partitioning and 'dispersive solid phase extraction' for the determination of pesticide residues in produce. *J. AOAC Int.* **2003**, *86*, 412–431.

(18) Ali, Y.; Hanna, M. A.; Chinnaswamy, R. Expansion characteristics of extruded corn grits. *Food Sci. Technol.* **1996**, *29*, 702–707.

(19) Chinnaswamy, R.; Bhattacharya, K. R. Studies on expanded rice. Optimum processing conditions. *J. Food Sci.* **1983**, *48*, 1604–1608.

(20) Chinnaswamy, R.; Hanna, M. A. Relationship between amylase content and extrusion-expansion properties of corn starches. *Cereal Chem.* **1988**, *65*, 138–143.

(21) Chiang, B. Y.; Johnson, J. A. Gelatinization of starch in extruded products. *Cereal Chem.* **1977**, *54*, 436–443.

(22) Davidson, V. J.; Paton, D.; Diosady, L. L.; Rubin, L. J. A model for mechanical degradation of wheat starch in a single-screw extruder. *J. Food Sci.* **1984**, *49*, 1154–1157.

(23) Chinnaswamy, R.; Hanna, M. A. Optimum extrusion-cooking conditions for maximum expansion of corn starch. *J. Food Sci.* **1988**, *53*, 834–836.

(24) Wolf, C. E.; Bullerman, L. B. Heat and pH alter the concentration of deoxynivalenol in an aqueous environment. *J. Food Protect.* **1998**, *61*, 365–367.

(25) Abbas, H. K.; Mirocha, C. J.; Rosiles, R.; Carvajal, M. Decomposition of zearalenone and deoxynivalenol in the process of making tortillas from corn. *Cereal Chem.* **1988**, *65*, 15–19.

(26) Lauren, D. R.; Smith, W. A. Stability of the Fusarium mycotoxins nivalenol, deoxynivalenol and zearalenole in ground maize under typical cooking environments. *Food Addit. Contam. A* **2001**, *18*, 1011–1016.