Think about **food**
Care about **safety**

Vol. 6
**Mycotoxins**
Focus on mycotoxins

The global production and trading of food and feed products makes monitoring and control difficult. Only analysis of food and raw materials for the production of food can ensure safe and edible food. Food safety is an essential part of our quality of life.

**MN shows examples for food analysis that will help you with your work and make our food safer!**

**Vol. 6: Mycotoxins in food infested with mold**

Mycotoxins are secondary metabolites of fungi/molds. In contrast to environmental contaminants (pesticides, PCBs, PAHs, etc.) mycotoxins are inevitable. More than 300 mycotoxins are chemically characterized [Food and Agriculture Organization of the United Nations (FAO) – http://www.fao.org/docrep/005/y1390e/y1390e0b.htm]. The following mycotoxins can be considered as the most common mycotoxins in feed and food:

Aflatoxins (aflatoxin B₁, B₂, G₁ and G₂, aflatoxin M₁), ochratoxin A (OTA), trichothecenes (Deoxynivalenol – DON, nivalenol, T-2 toxin, HT-2 toxin), patulin, ergot alkaloids, zearalenone – ZEA, fumonisins B₁ and B₂

The presence of mycotoxins in food and feed may affect the health of humans and animals. Many adverse health effects are suspected, such as causing of cancer and mutations and excess estrogens, gastrointestinal and kidney disorders [Food and Agriculture Organization of the United Nations (FAO) – http://www.fao.org/wairdocs/x5008e/x5008e01.htm].

**Toxicological properties of selected mycotoxins**

<table>
<thead>
<tr>
<th>Mycotoxin</th>
<th>Possible effects</th>
</tr>
</thead>
<tbody>
<tr>
<td>Aflatoxins</td>
<td>Liver diseases (hepatotoxic, hepatocarcinogenic); carcinogenic and teratogenic effects; hemorrhages (intestinal tract, kidneys); reduced growth rate; diminution of performance; immune suppression</td>
</tr>
<tr>
<td>Ochratoxins</td>
<td>Nephrotoxic; carcinogenic; mild liver damage; enteritis; teratogenic effects; poor feed conversion; reduced growth rate; immune suppression</td>
</tr>
<tr>
<td>Fumonisins</td>
<td>Pulmonary edema; equine leukoencephalomalacia; nephro- and hepatotoxic; immune suppression</td>
</tr>
<tr>
<td>Trichothecenes</td>
<td>Digestive disorders (vomiting, diarrhea, feed refusal); reduced weight gain; hemorrhages (stomach, heart, intestine, lung, bladder, kidney); edema; oral lesions; dermatitis; blood disorders; infertility; degeneration of bone marrow; slow growth; immune suppression</td>
</tr>
<tr>
<td>Zearalenone</td>
<td>Estrogenic effects; edema of vulva; prolapse of vagina; enlargement of uterus; atrophy of testicles; atrophy of ovaries; enlargement of mammary glands; infertility; abortion</td>
</tr>
<tr>
<td>Ergot alkaloids</td>
<td>Nervous or gangrenous syndromes; Digestive disorders (vomiting, diarrhea, feed refusal); reduced weight gain; convulsion; abortion</td>
</tr>
<tr>
<td>Citrinin</td>
<td>Nephrotoxic; teratogenic; hepatotoxic</td>
</tr>
<tr>
<td>Patulin</td>
<td>Mutagenic; genotoxic; neurotoxic; immune suppression</td>
</tr>
</tbody>
</table>

Compounds of interest

Chemical structures of selected mycotoxins

Aflatoxin B<sub>1</sub>  
\[
\begin{align*}
\text{Aflatoxin B}_{1} & : \text{Chemical Structure} \\
\end{align*}
\]

Aflatoxin B<sub>2</sub>  
\[
\begin{align*}
\text{Aflatoxin B}_{2} & : \text{Chemical Structure} \\
\end{align*}
\]

Aflatoxin M<sub>1</sub>  
\[
\begin{align*}
\text{Aflatoxin M}_{1} & : \text{Chemical Structure} \\
\end{align*}
\]

Aflatoxin G<sub>1</sub>  
\[
\begin{align*}
\text{Aflatoxin G}_{1} & : \text{Chemical Structure} \\
\end{align*}
\]

Aflatoxin G<sub>2</sub>  
\[
\begin{align*}
\text{Aflatoxin G}_{2} & : \text{Chemical Structure} \\
\end{align*}
\]

Aflatoxin M<sub>2</sub>  
\[
\begin{align*}
\text{Aflatoxin M}_{2} & : \text{Chemical Structure} \\
\end{align*}
\]

Ochratoxin A  
\[
\begin{align*}
\text{Ochratoxin A} & : \text{Chemical Structure} \\
\end{align*}
\]

Ochratoxin B  
\[
\begin{align*}
\text{Ochratoxin B} & : \text{Chemical Structure} \\
\end{align*}
\]

Zearalenone  
\[
\begin{align*}
\text{Zearalenone} & : \text{Chemical Structure} \\
\end{align*}
\]

Nivalenol  
\[
\begin{align*}
\text{Nivalenol} & : \text{Chemical Structure} \\
\end{align*}
\]

HT-2-Toxin  
\[
\begin{align*}
\text{HT-2-Toxin} & : \text{Chemical Structure} \\
\end{align*}
\]

T-2-Toxin  
\[
\begin{align*}
\text{T-2-Toxin} & : \text{Chemical Structure} \\
\end{align*}
\]

Patulin  
\[
\begin{align*}
\text{Patulin} & : \text{Chemical Structure} \\
\end{align*}
\]

Fumonisins  
\[
\begin{align*}
\text{Fumonisins} & : \text{Chemical Structure} \\
\end{align*}
\]

B<sub>1</sub>: R = OH  
\[
\begin{align*}
\text{B}_{1} : \text{Chemical Structure} \\
\end{align*}
\]

B<sub>2</sub>: R = H  
\[
\begin{align*}
\text{B}_{2} : \text{Chemical Structure} \\
\end{align*}
\]
Typical samples, preparation

T-2 toxin and HT-2 toxin from oat products in accordance with § 64 LFGB (German Regulation for Food Analysis)

Sample pretreatment
25 ± 0.1 g of a ground homogenized sample (particle size < 1 mm) are weighed into an Erlenmeyer flask or centrifuge tube. 100 mL acetonitrile – water (84:16, v/v) is added. The sample is mixed in a laboratory mixer (1 h, RT). After extraction 10 mL of solution is filtered through a folded filter or centrifuged (2500 g, RT).

Solid phase extraction

**Column type:**
CHROMABOND® Carbon/Alox/Celite®, 6 mL, 500 mg REF 730584 (on request)
No conditioning or equilibration of the SPE column is necessary.

**Sample aspiration:**
5 mL of prepared sample is passed through the column by slight vacuum (1–2 drops/s) into a silanized glass vial.

**Washing:**
2 x 5 mL acetonitrile – water (84:16, v/v); these eluates are passed into the same silanized vial, i.e., all eluates are combined.

Transfer into a sample vial, e.g., 1.5 mL screw neck vials N 9, amber, flat bottom, label and scale, wide opening, REF 702284 (silanized version REF 702079) and ready assembled screw closures N 9, blue, center hole with septum silicone / PTFE, REF 702287.1

It is recommended to filter the prepared sample through disposable filters CHROMAFIL® Xtra PVDF-45/25 REF 729219

For other applications see [www.mn-net.com/apps](http://www.mn-net.com/apps)
**Analysis**

**Subsequent analysis: HPLC**

**Mycotoxin exposure in Germany**

*MN Appl. No. 127240*

**Column:** EC 150/2 NUCLEODUR® C18 Pyramid, 3 μm

REF 760261.20

<table>
<thead>
<tr>
<th>Peaks</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>DON</td>
<td>Deoxynivalenol</td>
</tr>
<tr>
<td>DON-GlcA</td>
<td>Deoxynivalenol-3-glucuronide /</td>
</tr>
<tr>
<td></td>
<td>Deoxynivalenol-15-glucuronide</td>
</tr>
<tr>
<td>T-2</td>
<td>T-2 toxin</td>
</tr>
<tr>
<td>HT-2</td>
<td>HT-2 toxin</td>
</tr>
<tr>
<td>HT-2-GlcA</td>
<td>HT-2-toxin-4-glucuronide</td>
</tr>
<tr>
<td>FB1</td>
<td>Fumonisin B₁</td>
</tr>
<tr>
<td>FB2</td>
<td>Fumonisins B₂</td>
</tr>
<tr>
<td>AFB1</td>
<td>Aflatoxin B₁</td>
</tr>
<tr>
<td>AFB2</td>
<td>Aflatoxin B₂</td>
</tr>
<tr>
<td>AFG2</td>
<td>Aflatoxin G₂</td>
</tr>
<tr>
<td>AFM1</td>
<td>Aflatoxin M₁</td>
</tr>
<tr>
<td>ZAN</td>
<td>Zearalanone</td>
</tr>
<tr>
<td>ZEN</td>
<td>Zearalenone</td>
</tr>
<tr>
<td>α-ZEL</td>
<td>α-Zearalenol</td>
</tr>
<tr>
<td>β-ZEL</td>
<td>β-Zearalenol</td>
</tr>
<tr>
<td>ZEN-14-GlcA</td>
<td>Zearalenone-14-O-glucuronide</td>
</tr>
<tr>
<td>ZAN-14-GlcA</td>
<td>Zearalanone-14-O-glucuronide</td>
</tr>
<tr>
<td>α-ZEL-14-GlcA</td>
<td>α-Zearalenol-14-O-glucuronide</td>
</tr>
<tr>
<td>β-ZEL-14-GlcA</td>
<td>β-Zearalenol-14-O-glucuronide</td>
</tr>
<tr>
<td>OTA</td>
<td>Ochratoxin A</td>
</tr>
<tr>
<td>OTα</td>
<td>Ochratoxin αα</td>
</tr>
<tr>
<td>EnB</td>
<td>Enniatin B</td>
</tr>
<tr>
<td>DH-CIT</td>
<td>Dihydrocitrinone</td>
</tr>
</tbody>
</table>

**Patulin and hydroxymethylfurfural from apple juice**

*MN Appl. No. 121800*

**Column:** EC 250/4 NUCLEODUR® C18 Gravity, 5 μm

REF 760101.40

<table>
<thead>
<tr>
<th>Peaks</th>
<th>Description</th>
</tr>
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<tbody>
<tr>
<td>1</td>
<td>Hydroxymethylfurfural</td>
</tr>
<tr>
<td>2</td>
<td>Patulin</td>
</tr>
</tbody>
</table>

Detailed conditions are available online at [www.mn-net.com/apps](http://www.mn-net.com/apps)
Subsequent analysis: HPLC

Fast separation of aflatoxins

MN Appl. No. 125600

Column: EC 50/2 NUCLEOSHELL® PFP, 2.7 μm

REF 763532.20

MN suggests using guard columns to protect your main column and significantly increase column life.

The Column Protection System (REF 718966) is suitable for all analytical HPLC columns with 1/16” fittings!

Peaks:
1. Aflatoxin G₂
2. Aflatoxin G₁
3. Aflatoxin B₂
4. Aflatoxin B₁

Detailed conditions are available online at www.mn-net.com/apps
## Product information

### Ordering information

<table>
<thead>
<tr>
<th>Step</th>
<th>Product</th>
<th>Pack of</th>
<th>REF</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>SPE</strong></td>
<td>CHROMABOND® Carbon/Alox/Celite®, 6 mL, 500 mg (on request)</td>
<td>30</td>
<td>730584</td>
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<tr>
<td></td>
<td>CHROMABOND® vacuum manifold for 12 columns</td>
<td>1</td>
<td>730150</td>
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<tr>
<td><strong>Filtration</strong></td>
<td>Disposable filters CHROMAFIL® Xtra PVD-45/25</td>
<td>100</td>
<td>729219</td>
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<td><strong>Vials and caps</strong></td>
<td>1.5 mL screw neck vials N 9, amber, label and scale</td>
<td>100</td>
<td>702284</td>
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<tr>
<td></td>
<td>Vials N 9 as before, silanized version</td>
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<td>702079</td>
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<tr>
<td></td>
<td>Ready assembled screw closures N 9, blue, center hole with septum Silicone white/PTFE red</td>
<td>100</td>
<td>702287.1</td>
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<tr>
<td><strong>HPLC 1</strong></td>
<td>Column EC 150/2 NUCLEODUR® C18 Pyramid, 3 μm</td>
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<td>Guard column EC 4/2 NUCLEODUR® C18 Pyramid, 3 μm</td>
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<td>Guard column holder Column Protection System</td>
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<tr>
<td><strong>HPLC 2</strong></td>
<td>Column EC 250/4 NUCLEODUR® C18 Gravity, 5 μm</td>
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<td>Guard column EC 4/3 NUCLEODUR® C18 Gravity, 5 μm</td>
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<tr>
<td><strong>HPLC 3</strong></td>
<td>Column EC 50/2 NUCLEOSHELL® PFP, 2.7 μm</td>
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<tr>
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<td>Guard column EC 4/2 NUCLEOSHELL® PFP, 2.7 μm</td>
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<tr>
<td></td>
<td>Guard column holder Column Protection System</td>
<td>1</td>
<td>718966</td>
</tr>
</tbody>
</table>

Visit us at [www.mn-net.com/chroma](http://www.mn-net.com/chroma) to get more helpful information

### Selection tools for

- Vials and caps
- Syringe filters

### Troubleshooting guides

- GC and HPLC

### Detailed product information

...and much more
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- Mycotoxins (Vol. 6)
- Sugars Sweeteners (Vol. 5)
- Acrylamide (Vol. 4)
- Antimicrobials (Vol. 3)
- Pesticides (Vol. 7)

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