Research on the metabolism and renal excretion of DON and DON-3-glucoside in humans.

HBM4EU, Wageningen, November 2018

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Introduction

- High presence of DON in food.

Objectives

Methodology

Results and discussion

Conclusions

- DON-3-glucoside can have a large concentration in food.

Table. Obtained results for the ratio DON-3-glucoside/DON in raw cereals.

<table>
<thead>
<tr>
<th>References</th>
<th>Ratio</th>
</tr>
</thead>
<tbody>
<tr>
<td>Berthiller et al., 2009</td>
<td>0.15</td>
</tr>
<tr>
<td>Dall'Asta et al., 2013</td>
<td>0.28</td>
</tr>
<tr>
<td>Rasmussen et al., 2013</td>
<td>0.22</td>
</tr>
</tbody>
</table>

Table. Obtained results for the ratio DON-3-glucoside/DON in breads.

<table>
<thead>
<tr>
<th>References</th>
<th>Ratio</th>
</tr>
</thead>
<tbody>
<tr>
<td>De Boevre et al., 2012</td>
<td>1</td>
</tr>
<tr>
<td>De Boevre et al., 2012</td>
<td>0.84</td>
</tr>
<tr>
<td>Vidal et al., 2016</td>
<td>0.75</td>
</tr>
</tbody>
</table>

Figure. Percentage of DON in cereals products.

References. Cano-Sancho et al., 2011.
- High presence of DON in urine.

- BIOMYC0-study (2012-2014, n=394):
  > 90% DON + DON-glucuronides.
  (Heyndrickx et al., 2015)

- High levels of DON in urine 56-69% children >>> TDI
  DON TDI: 1 µg/kg bw/day
  16-29% adults >>>

Figure. Some DON metabolites after DON intake.
- Uncertainties:

1. Some DON biomarkers in urine can come from DON-3-glucoside?

2. Morning urine or 24 h?

3. Excretion rate? (72% Turner et al., 2010; 68% Warth et al., 2013)
• Unraveling the human metabolism of DON and DON-3-glucoside.
  o Excretion pattern of DON, DON-3-glucoside and metabolites + excretion-rates.
  o Standardized method to estimate DON-intake by means of biomarkers.
- Ethical Approval.
- Trial: 20 representative subjects (4 controls).
- 55% women 45% men.
- Average age = 32 years old.
- Questionnaire (Smoking, Coffee, BMI, ...).

**Figure.** Scheme of intervention diet study.
### Analysis

**Methodology**

- **Analysis**: LC-MS/MS
- **QUECHERS method.**

<table>
<thead>
<tr>
<th>Mycotoxin</th>
<th>LOD (µg/kg)</th>
<th>LOQ (µg/kg)</th>
<th>Calibration range (µg/kg)</th>
<th>R (mean)</th>
<th>Apparent recovery (%)</th>
<th>SE</th>
<th>RSD_U (%)</th>
<th>RSD_R (%)</th>
<th>U (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>DON</td>
<td>0.2</td>
<td>0.4</td>
<td>0.5-100</td>
<td>0.99</td>
<td>103.3</td>
<td>7.6</td>
<td>5.5</td>
<td>6.6</td>
<td>14.8</td>
</tr>
<tr>
<td>DON-3-glucoside</td>
<td>0.3</td>
<td>0.6</td>
<td>0.5-100</td>
<td>0.99</td>
<td>97.8</td>
<td>8.7</td>
<td>0.9</td>
<td>3.7</td>
<td>8.3</td>
</tr>
<tr>
<td>DON-3-glucuronide</td>
<td>0.5</td>
<td>1.0</td>
<td>0.5-100</td>
<td>0.99</td>
<td>111.3</td>
<td>9.9</td>
<td>7.2</td>
<td>10.1</td>
<td>20.8</td>
</tr>
<tr>
<td>DON-15-glucuronide</td>
<td>0.4</td>
<td>0.9</td>
<td>0.5-100</td>
<td>0.99</td>
<td>108.1</td>
<td>9.9</td>
<td>7.2</td>
<td>10.1</td>
<td>20.7</td>
</tr>
<tr>
<td>DOM-1</td>
<td>0.6</td>
<td>1.2</td>
<td>0.5-100</td>
<td>0.99</td>
<td>101.3</td>
<td>0.5</td>
<td>2.8</td>
<td>5.4</td>
<td>10.0</td>
</tr>
<tr>
<td>3-ADON/15-ADON</td>
<td>0.1</td>
<td>0.2</td>
<td>0.5-100</td>
<td>0.99</td>
<td>105.1</td>
<td>7.7</td>
<td>2.5</td>
<td>4.5</td>
<td>13.2</td>
</tr>
</tbody>
</table>
Mycotoxins recovery after 24 hours of urine collection:

**DON**
- Free DON: 17%
- DON-3-glucuronide: 9%
- DON-15-glucuronide: 38%
- Unknown: 36%

Fexcretion = 0.64

**DON-3-glucoside**
- Free DON: 14%
- DON-3-glucoside: 3%
- DON-3-glucuronide: 9%
- DON-15-glucuronide: 28%
- Unknown: 46%

Fexcretion = 0.53
Average percentage of total mycotoxin excreted in urine over time (hDON DON-3-glucoside).
Excretion profile for average of total DON, DON-15-glucuronide, DON-3-glucuronide and free DON excreted after DON administration.
Correlation between DON-15-glucuronide and DON-3-glucuronide: 0.784

Results and discussion

- DON-15-glucuronide
- DON-3-glucuronide

Amount excreted (％)

Time after dosage (h)
Average of total DON, DON-15-glucuronide, DON-3-glucuronide and free DON excreted after DON-3-glucoside administration.
Gender could affect the excretion of DON.

- Men excreted less total DON than women ($p < 0.05$).
- Glucuronides were higher in women (UGT expression).
Other factors did not cause variations:

- Age (children excreted more DON than adults?)
- BMI (hydrosoluble)
- Diet
- Coffee
- Smoking
Conclusions:

- 0.64 for DON and 0.53 for DON-3-glucoside excretion rate.

- DON, DON-15-glucuronide and DON-3-glucuronide are main metabolites of DON.

- Correlation of DON-15-glucuronide and DON-3-glucuronide.

- DON represent a fast excretion, urine collection of 24 hours better than urine morning.

- Gender affects the DON excretion.

Age, coffee, smoking or BMI did not show any affect to DON.
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