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Digestibility and absorption of deoxynivalenol-3-*β*-glucoside in *in vitro* models

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Background

Certain mycotoxins may be present in plant materials as their glycosides, which may escape from routine analysis. The question is whether these so-called masked-mycotoxins may be hydrolysed into their parent compounds in the gastro- intestinal tract (GI-tract), thus increasing the exposure.

Objective

The main aim of this study was to determine the potential transformation of 3-*β*-glucoside (D3G) to deoxynivalenol (DON) in a digestion model representing the upper GI-tract and the possible transformation of D3G to DON and absorption of D3G by intestinal epithelium cells using an *in vitro* absorption model, to assess the possible increased exposure of consumers to DON.

Materials and Methods

In vitro *digestion model*

The *in vitro* digestion model, as described by Versantfoort *et al*. (2005)[1] for fed conditions (Olvarit 15M2 Nutricia, NL), was used (figure 1). Samples were spiked at 2222 µg DON/kg infant formula, 2778 µg D3G/kg infant formula or left blank. The chyme was ultrasonicated and centrifuged. An aliquot of 0.5 ml supernatant was filtered over a 0.45 µm filter and transferred to an HPLC vial. A precise aliquot of $^{13}C_{15}$-DON standard solution was added as internal standard. A 10 µl volume was injected on the LC-ESI-MS/MS system. The chyme was analysed for DON, D3G and DOM-1.

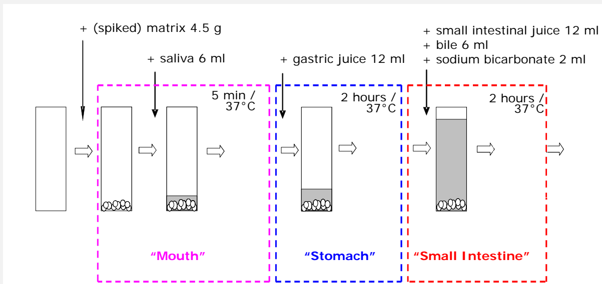

Figure 1. Schematic overview of *in vitro* digestion model

In vitro *intestinal absorption model*

The *in vitro* intestinal absorption model, using Caco-2 cells derived from human colon adenocarcinom, was applied as described by Steensma *et al*. (1999)[2] (figure 2). Caco-2 cells were seeded in Transwells. An aliquot of 1.5 ml of DMEM+++ medium containing an absolute amount of either 3.5 nmol DON, 3.6 nmol D3G or no mycotoxin (blank), was added to the apical side of the cell layer. An aliquot of 2.5 ml of blank DMEM+++ medium was added to the basolateral side.

Concentrations of DON, D3G and DOM-1 were analysed in medium samples from both the apical and the basolateral side taken after 0, 4 and 24 h respectively. One ml samples were taken from both compartments, filtered over a 0.45 µm filter. A 10 µl volume was analysed by LC-ESI-MS/MS.

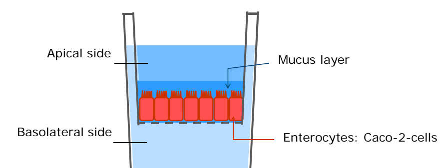

Figure 2. Schematic overview of *in vitro* absorption model

Results

In vitro *digestion model*

No DON was detected in the chyme of the samples spiked with D3G after digestion. Thus, theoretically, less than 5% (w/w) of D3G was hydrolysed to DON. No D3G was detected in the digested samples spiked with DON. No DON or D3G was detected in the blank samples and no DOM-1 was detected in any of the samples.

In vitro *absorption model*

No evidence for transformation of D3G to DON or DOM-1 by the Caco-2 cells was found. No transport of D3G from the apical side to the basolateral side was detected. When DON was added to the apical side, no D3G or DOM-1 was detected in either apical or basolateral side. A total of 20% of the apically added DON was transported to the basolateral side.

Conclusions

- No evidence was found in the *in vitro* experiments for significant elevated exposure of humans to DON from dietary D3G, since D3G was not hydrolysed to DON in the digestion model representing the upper part of the GI-tract and D3G was not hydrolysed to DON by the intestinal epithelial Caco-2 cells
- Bioavailability of D3G in humans may be low as compared to DON since Caco-2 cells did not absorb D3G, in contrast to DON.
- This work has been published in the World Mycotoxin Journal[3].

Acknowledgements

This research was (partly) financed by the Netherlands Ministry of Economic Affairs, Agriculture and Innovation.

Literature

[1] Versantfoort, C.H.M. *et al.* (2005). Food Chem Toxicol 43: 31–40.
[2] Steensma, A. *et al.* (1999) Environ Toxicol Pharmacol 7: 209 212.
[3] de Nijs, M. *et al.* (2012) World Mycotoxin Journal 5: 319-325.

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